Severe Infection in ANCA-Associated Vasculitis: Incidence, Prognostic Factors and Prediction

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Abstract

Introduction

Individuals with ANCA-associated vasculitis (AAV) are at high risk of severe infections. Using prognosis research methodology, this thesis will explore the incidence of severe infection, examine glucocorticoid exposure as a prognostic marker for severe infection and will develop prognostic models to predict severe infection events: the occurrence of first severe infection after diagnosis and early mortality following a severe infection event. Subsequent chapters will examine Covid-19 in AAV patients including prognostic factors for severe disease and the impact of SARS-CoV-2 vaccination in AAV patients treated with rituximab.

Methods

Diverse datasets were utilised to address the thesis aims. Novel semantic web technology was deployed to federate multiple European AAV registries to determine severe infection incidence. Data linkage was used to develop a large AAV dataset using national Scottish routinely-collected health data for the severe infection prognostic marker and prognostic modelling studies. A bi-national cohort of AAV patients with Covid-19 was developed through the contribution of vasculitis clinicians throughout the UK and Ireland. SARS-CoV-2 vaccination data was derived from the AAV sub-group of the UK-wide multicentre multi-disease OCTAVE study of immunosuppressed individuals. For the prognostic modelling studies, modern prediction methodologies were applied.

Results

Severe infection incidence was high, especially in the first year after diagnosis. Glucocorticoid exposure thresholds above 10 mg daily all had a substantial positive association with severe infection. Multivariable models were developed with good predictive ability for both severe infection events and early mortality following severe infection. Prognostic factors for severe Covid-19 included

immunosuppressive agents. The humoral immune response to SARS-CoV-2 vaccination was severely attenuated in AAV patients compared to controls.

Discussion

Through applying prognosis research methodology, this thesis quantified the incidence of severe infection, identified prognostic factors and developed prognostic models for severe infection in individuals with AAV. Prognostic factors relating to Covid-19 were determined.

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Author's Declaration

The work presented was completed during a Clinical Research Fellow position funded by Vifor Pharma. I declare that I am the author of this thesis and was responsible for the conceptualisation, design, conduct and analysis of all studies, under the supervision and with the guidance of my supervisors. Several colleagues collaborated with various aspects of these studies and their contributions are acknowledged below.

Chapter 2 – FAIRVASC project: Dr Karl Gisslander and Dr Kris McGlynn collaborated with me to harmonise disease terms in the registries, co-design the FAIRVASC ontology and develop the FAIRVASC technical infrastructure.

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Chapter 7 – OCTAVE – the OCTAVE study team was responsible for the overall design and conduct of the main OCTAVE study, data from which was utilised for the study reported in Chapter 7.

Definitions/Abbreviations

2012 CCHC	2012 Revised International Chapel Hill Consensus Conference Nomenclature of Vasculitides
AAV	ANCA-associated vasculitis
AAV-GN	ANCA-associated vasculitis glomerulonephritis
ACE / ACE2	Angiotensin-converting enzyme (2)
ACR 1990	American College of Rheumatology 1990 Criteria for the Classification of Vasculitis
AI	Artificial intelligence
ANCA	Antineutrophil cytoplasmic antibody
Anti-N	Anti-nucleocapsid protein antibody
Anti-S	Anti-spike protein antibody
aOR	Adjusted odds ratio
ARB	Angiotensin II receptor blockers
BVAS	Birmingham Vasculitis Score
C-statistic	Concordance statistic
C/D AUCt	Cumulative/dynamic area under the receiver operating characteristic curve
CCI	Charlson Comorbidity Index
CFR	Case fatality rate
CHCC	Chapel Hill Consensus Conference
CHI	Community Health Index
CI	Confidence interval
CITL	Calibration-in-the-large
CKD	Chronic kidney disease
CMV	Cytomegalovirus
COPD	Chronic obstructive pulmonary disease
Covid-19	Coronavirus disease 2019
CRCTU	Cancer Research UK Clinical Trials Unit, University of Birmingham
CRF	Case report form
CRP	C-reactive protein
CTCAE	Common Terminology Criteria for Adverse Events
CVD	Cerebrovascular disease
DCVAS	Diagnostic and Classification Criteria in Vasculitis
DMARD	Disease-modifying antirheumatic drug
DOB	Date of birth
DPP4	Dipeptidyl peptidase 4
DQ	Data quality
DQG	Data Quality Group

ECMO	Extracorporeal membrane oxygenation
eDRIS	Electronic Data Research and Innovation Service
eGFR	Estimated glomerular filtration rate
EGPA	Eosinophilic granulomatosis with polyangiitis
EJPRD	European Joint Program on Rare Diseases
EMA	European Medicine Agency
ENT	Ear, nose and throat
EPP	Events per candidate predictor
EPV	Events per variable
ESKD	End-stage kidney disease
EU	European Union
EULAR	European Alliance of Associations for Rheumatology
EUVAS	European Vasculitis Study Group
FAIR	Findable, Accessible, Interoperable and Reusable
FIT	FAIRVASC implementation team
FNGN	Focal necrotizing glomerulonephritis
FOAF	Friend of a friend
FP	Fractional polynomial
FVSG	French Vasculitis Study Group
GBM	Glomerular basement membrane
GC	Glucocorticoid
GCA	Giant cell arteritis
GDCN	Glomerular Disease Collaborative Network
GDPR	General Data Protection Regulation
GEVAS	German, Austrian and Swiss Vasculitis Registry
GPA	Granulomatosis with polyangiitis
GRF	Global Rheumatology Alliance
HC	Healthy controls
HE	Hematoxylin and eosin
HIT	Harmonisation team
HLA	Human leukocyte antigen
HR	Hazard ratio
HSC-PBPP	Public Benefit and Privacy Panel for Health and Social Care
HSP	Henoch-Schönlein purpura
HSV	Herpes simplex virus
ICD-10	International Statistical Classification of Diseases and Related Health Problems 10th Revision
ID	Identification number
IFNγ	Interferon gamma

IFR	Infection fatality ratio
IgA, IgG etc	Immunoglobulin A, G etc
IMID	Immune-mediated inflammatory disease
IQR	Interquartile range
IRR	Incidence rate ratio
IVIG	Intravenous immunoglobulin
KM	Kaplan-Meier
LASSO	Least absolute shrinkage and selection operator
LVV	Large vessel vasculitis
MEDS	Mortality in Emergency Department Sepsis score
MERS-CoV	Middle East respiratory syndrome virus
MeSH	Medical Subject Heading
MFP	Mulitvariable fractional polynomial modelling
MHRA	UK Medicines and Healthcare Products Regulatory Agency
MI	Myocardial infarction
MPA	Microscopic polyangiitis
MPO	Myeloperoxidase
MRC	Medical Research Council
NETs	Neutrophil extracellular traps
NIH	National Institute for Health
NRS	National Records of Scotland
NSAID	Non-steroidal anti-inflammatory drug
NSH	National Safe Haven
O/E ratio	Observed-to-expected ratio
OCTAVE	Observational Cohort trial T cells, Antibodies and Vaccine Efficacy in SARS-CoV-2
OR	Odds ratio
PAM	Periodic acid methenamine silver
PAN	Polyarteritis nodosa
PAS	Periodic acid–Schiff
PBMC	Peripheral blood mononuclear cells
PCP	Pneumocystis pneumonia
PCR	Polymerase chain reaction
PF	Prognostic factor
PHS	Public Health Scotland
PIS	Prescribing Information System
PITCH	Protective Immunity from T cells in Healthcare workers study
PMR	Polymyalgia rheumatica
POLVAS	Polish Vasculitis Registry

PPI	Patient and public involvement
PR3	Proteinase 3
PROBAST	Prediction model Risk Of Bias ASsessment Tool
PROGRESS	Prognosis Research Strategy
PVD	Peripheral vascular disease
QIT	Query implementation team
QOL	Quality of life
QUIPS	Quality in Prognosis Studies tool
R2RML	Relational to Resource Description Framework Mapping Language
RA	Rheumatoid arthritis
RBD	Receptor-binding domain
RCT	Randomised controlled trial
RDF	Resource Description Framework
REVAS	Spanish Registry of Systemic Vasculitis
RKD	Ireland Rare Kidney Disease Registry
RLV	Renal limited vasculitis
RTX	Rituximab
SARS-CoV / SARS-Cov-2	Severe acute respiratory syndrome virus (2)
SIMD	Scottish Index of Multiple Deprivation
SMR	Standardised mortality rate
SMR01	Scottish Morbidity Record admissions database
SPARQL	SPARQL Protocol and RDF Query Language
STROBE	Strengthening the Reporting of Observational Studies in Epidemiology
SVV	Small vessel vasculitis
TMP/SMX	Trimethoprim/sulfamethoxazole
TRIPOD	Transparent Reporting of a multivariable prediction model for Individual Prognosis Or Diagnosis
UKIVAS	UK and Ireland Vasculitis Rare Disease Group / registry
UofG	University of Glasgow
URI	Uniform resource identifier
URL	Uniform resource locator
VDI	Vasculitis Damage Index
VOC	Variant of concern
VOICES	Vasculitis Outcomes in relation to Care Experiences
VTE	Venous thromboembolism
VZV	Varicella zoster virus
W3C	World Wide Web Consortium
WHO	World Health Organisation

1 Introduction

1.1 Overview

Individuals with ANCA-associated vasculitis (AAV) are recognised as being at high risk of severe infections. This thesis will explore prediction epidemiology themes relating to severe infection in this population, guided by a prognosis research framework and associated methodology. The incidence of severe infections, prognostic factors and prognostic modelling will be investigated. The following introductory chapter will give context to the disease, including its history, biology and clinical management. Adverse outcomes will be discussed, including a detailed review of the epidemiology of severe infections in AAV. Finally, the PROGRESS framework, which delineates concepts in prognosis research, will be described, providing a conceptual structure on which the subsequent research studies described in the thesis are based.

1.2 What is ANCA-associated vasculitis?

The vasculitides are a heterogeneous set of over 25 distinct conditions with a core pathological common feature: inflammation in the blood vessel wall. Specifically, this is the presence of inflammatory leukocytes in the vessel wall with associated reactive damage to the vessel. Disruption of mural structures, leading to bleeding and loss of luminal patency due to inflammation and clotting, result in downstream tissue ischaemia and necrosis. The vasculitides are multiorgan diseases with heterogenous presentations. They are often serious and can result in death. These 25 separate entities are classically groups into different categories, primarily based on the size of affected blood vessels: small, medium and large vessel vasculitis. These categories are described in detail in the 2012 Revised International Chapel Hill Consensus Conference Nomenclature of Vasculitides (2012 CHCC) (Jennette et al., 2013).

Antineutrophil cytoplasmic antibody (ANCA)-associated vasculitis (AAV) group of vasculitides, that typically affects small vessels, from small arteries, through arterioles and capillaries to venules. It is therefore classified as a small vessel vasculitis (SVV). It is associated with serum ANCA or a phenotypically identical vasculitis but without ANCA. AAV can affect any blood vessel in the human body, therefore a wide range of clinical manifestations occur with substantial variability. AAV is among the most severe of the vasculitides and, untreated, is effectively universally fatal. Three main clinicopathological phenotypes are recognised: microscopic polyangiitis (MPA), granulomatosis with polyangiitis (GPA) and eosinophilic granulomatosis with polyangiitis (EGPA) (Sinico and Guillevin, 2019).

ANCAs are autoantibodies, predominantly IgG antibodies, directed against antigens predominantly expressed in the cytoplasm of neutrophils. The two most clinically relevance and well recognised antigens are Proteinase 3 (PR3) and myeloperoxidase (MPO). AAV can be classified in relation to which ANCA subtype is expressed. ANCA-negative AAV also occurs, but is clinically and pathologically indistinguishable from ANCA-positive AAV. ANCAs are discussed in more detail in section 1.7.

1.3 History

The recognition of MPA, GPA and EGPA as distinct conditions occurred over the first half of the twentieth century. The discovery of these entities began in the nineteenth century, with the identification of two clinical phenomena associated with vasculitides: purpura and arteritis. Purpura is a skin or mucous membrane rash due to bleeding from small blood vessels. In vasculitis this localised bleeding is caused by necrotising inflammation of small blood vessels. In 1808 Robert William, a dermatologist, described purpura associated with systemic features. Later in the 1800s, Eduard Henoch and Johann Schönlein described cases of children with purpura that was typically associated with abdominal pain, joint pain and nephritic syndrome. These children most likely had IgA vasculitis (previously termed Henoch-Schönlein purpura [HSP]) (Sinico and Guillevin, 2019). Later the renowned Canadian physician William Osler described a series of cases of purpura with associated multisystem features such as epistaxis, arthritis, pulmonary haemorrhage, iritis and nephritis. It is highly likely that some of the cases described in Osler's series had a systemic vasculitis such as AAV (Osler, 1914).

In 1866, Adolf Kussmaul, a physician, and his pathologist colleague, Rudolf Maier, described the first known account of a patient with systemic necrotising arteritis. They described the condition as periarteritis nodosa due to gross inflammatory nodular lesions affecting the wall, and possibly the surrounding tissues, of medium sized arteries (Sinico and Guillevin, 2019). Due to transmural inflammation later being recognised as a key feature, the condition became known as polyarteritis nodosa (PAN) (Dickson, 1908). For many decades, these terms were used to describe a multitude of diseases associated with arteritis, which were later considered to be distinct entities.

In 1923, a microscopic form of periarteritis nodosa was described by Friedrich Wohlwill, a German neurologist and pathologist (Wohlwill, 1923). This was later confirmed by Davson and colleagues as pathologically distinct from PAN (Wainwright and Davson, 1950). This was due to specific small arteries being affected: arterioles, capillaries and venules. Subsequently a link between arteritis and SVV associated with purpura, pulmonary haemorrhage and glomerulonephritis began to be established. Following on from Wohlwill's

description, a variety of conditions were identified that had characteristics similar to PAN but had unique findings that merited separate diagnoses. These included Wohlwill's description of what was essentially MPA (Wohlwill, 1923), Wegener's granulomatosis (later GPA) (Wegener, 1939), Churg-Strauss syndrome (later EGPA) (Churg and Strauss, 1951) and Kawasaki disease (Kawasaki, 1967). In the 1950, Godman and Churg grouped the sub-diagnoses of AAV together as related entities and separate from PAN (Godman and Churg, 1954). It was in this article that the term Wegener's Granulomatosis was formalised. They proposed MPA, GPA and EGPA likely shared common underlying pathogenesis. In the 1980s, ANCA were discovered: initially termed anticytoplasmic antibodies or ACPA. This confirmed that MPA, GPA and EGPA were biologically related and therefore separate entities to PAN, which was found to be ANCA-negative (van der Woude et al., 1985).

Wegener and the Nazis

In 2011, a group of vasculitis academic clinicians representing multiple nations and medical specialties published an explanation for the proposed alternative name for Wegener's Granulomatosis. The new name was Granulomatosis with Polyangiitis. This was in the spirit of a shift from historic honorific eponyms to names which better represented the clinical features, aetiology or pathogenesis of diseases. The move was prompted by evidence that Dr Friedrich Wegener was a member of the Nazi party before and during World Ward II and has close association with various Nazi organisations. Though his personal views are not known, Wegener had close professional relationships with prominent Nazis. Before the war, his head of department at the University of Breslau was Martin Staemmler, a well-known author of racial hygiene texts. During the war, Wegener was stationed in Lodz, Poland as an army pathologist. There is evidence that Wegener performed autopsies on Jews who did not survive transport from the Lodz ghetto in Poland to the nearby death camp at Chelmno (Falk et al., 2011; Woywodt et al., 2006).

Published in 2013, eosinophilic granulomatosis with polyangiitis was used in the 2012 CHCC as a more descriptive term for Churg-Strauss syndrome (Jennette et al., 2013).

1.4 Nomenclature and classification

Classification is a process that uses patients characteristics to group them in standardised classes (see definitions below). Classification in the field of vasculitis is challenging. AAV can be categorised into distinct clinicopathological entities. At a population level, this represents classification, while it would represent diagnosis at the level of an individual patient. These classes or diagnoses include microscopic polyangiitis (MPA), granulomatosis with polyangiitis (GPA) and eosinophilic granulomatosis with polyangiitis (EGPA). Organ limited AAV is recognised, such as renal limited vasculitis (RLV), however RLV can be considered a form of organ limited MPA. Notably MPA, GPA and EGPA can have pathologically identical features. GPA and EGPA can be pathologically distinguished from MPA due to the presence of necrotising granulomatous lesions. Such lesions most frequently occur in the respiratory tract. Granulomas do not occur in MPA. EGPA can be distinguished from GPA due to the presence of asthma and eosinophilia. As described above in section 1.2, ANCA can also be used to classify AAV.

Definitions around classification

Nomenclature: a system of names and definitions of diseases

Definition: the underlying disease process of the disease in question, typically describing underlying pathology.

Diagnosis (noun): the name of a disease.

Diagnosis (process): a process by which an individual patient is allocated a diagnosis. The criteria used may be similar to those used for classification. Diagnostic criteria often focus on establishing combinations of features that indicate a disease is present with a high degree of certainty.

Classification: a process by which patients are organised into well-defined groups. Primarily to allow a homogeneous group of subjects to be correctly identified for research. This may be similar to the diagnostic process, but differs in important ways. Classification criteria typically exclude disease characteristics that are common across different diseases. This is because such characteristics may not help differentiate different diseases for inclusion in a research study. Common characteristics may, however, facilitate identification of an individual with a disease and therefore are an important component of diagnostic criteria. Classification criteria are not appropriate to use for obtaining a diagnosis in an individual patient.

1.4.1 Nomenclature: Chapel Hill Consensus Conference

The first International Chapel Hill Consensus Conference on the Nomenclature of Systemic Vasculitides took place in Chapel Hill, North Carolina (Jennette et al., 1994). The principal aims of the meeting were to reach agreement on the names of non-infection related vasculitides and to derive definitions for these conditions.

2012 CHCC led to further clarification on the nomenclature of vasculitis. Similar to the output of the previous conference, it did not, however, include criteria for allocation groups of patients into these classes (classification) or for the

diagnosis of individuals (Jennette et al., 2013). AAV represents a group of SVV, but notably it spans more vessels sizes than the other forms of SVV, as depicted in Figure 1-1.

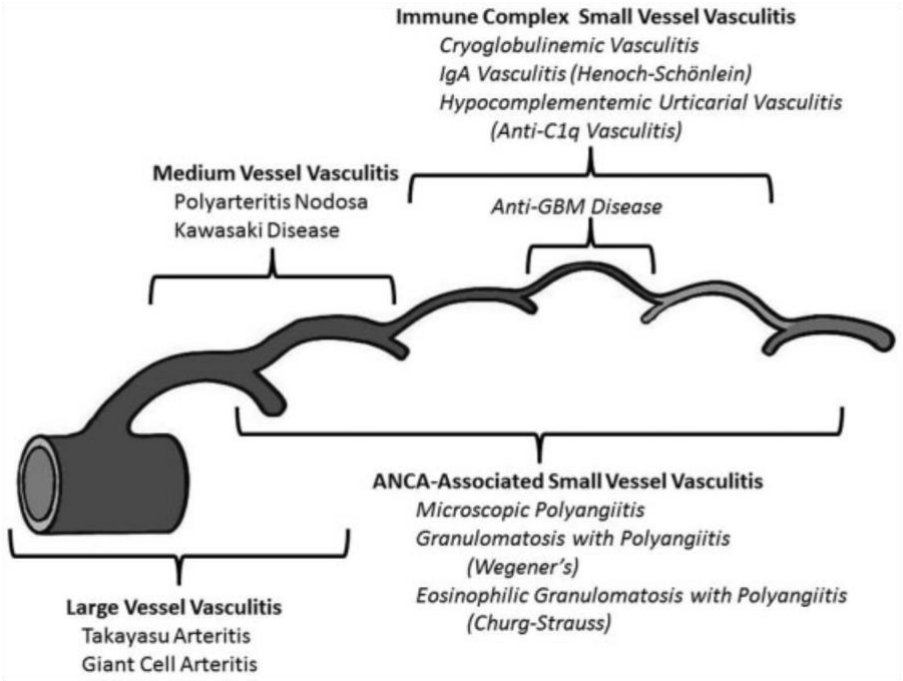


Figure 1-1 | Diagram adapted from the 2012 Chapel Hill Consensus Conference Article

An additional feature of 2012 CHCC was the incorporation of ANCA serotype into the diagnosis, for example PR3-ANCA GPA. The rationale was that both clinicopathological phenotype and serotype can influence classification, diagnosis and treatment of AAV patients.

1.4.2 American College of Rheumatology 1990 Criteria

The American College of Rheumatology 1990 Criteria for the Classification of Vasculitis (ACR 1990) was the result of the first major project conducted to formally classify vasculitis using prospective research data. A thousand patients were included from 47 rheumatology centres across the United State, Canada and Mexico. The objective was to improve communication among clinicians who care for vasculitis patients and to standardise research studies across different settings, thus facilitating comparison of studies. The initial scope was to establish criteria that would differentiate the different vasculitides. Criteria to

distinguish individuals with vasculitis from those without any form of vasculitis were not attempted. Seven forms of vasculitis were included in the study, two of which are currently recognised forms of AAV: EGPA (then Churg-Strauss syndrome) and GPA (then Wegener's Granulomatosis). The remainder were: PAN, hypersensitivity vasculitis, HSP, giant cell arteritis (GCA) and Takayasu arteritis. Of the 1,000 patients included, 85 had GPA, 20 had EGPA and 213 were excluded for having a form a vasculitis different to those listed above, such as rheumatoid arthritis (RA) or Kawasaki disease. 807 were ultimately included in the analysis. Two statistical methods were used to create the criteria after shortlists of the most discriminating variables were created. The first was labelled the 'traditional method' and involved clinicians selecting different combinations of shortlisted variables where a subject would be required to have a certain number of variables on the list present in order to be confirmed as having the disease in question. The second method was the 'classification tree' where the most discriminating variable first divides the study population into group, followed by the next most discriminating value for that branch (Hunder et al., 1990). Ultimately the include features for GPA were: evidence of red cells in urinary sediment, abnormal chest radiograph features typical for GPA, oral ulcers or nasal discharge, and granulomatous inflammation on biopsy. Two or more of these variables being present was consistent with 88.2% sensitivity and 92.0% sensitivity with the traditional methodology. The classification tree approach achieved very similar results with the same variables and the addition of haemoptysis (Leavitt et al., 1990). The decision tree approach yielded very high sensitivity and specificity for EGPA with the relatively simple criteria of eosinophilia >10% and either asthma or allergy confirming the diagnosis (Masi et al., 1990). Challenges around applying ACR 1990 in the modern era include that the criteria were published before MPA was formerly acknowledged as a distinct form of AAV. As MPA is not included in the ACR criteria, it cannot be used to identify or exclude MPA. The data on which ACR 1990 is based likely had, what would now be recognised as, MPA patients in the GPA group. It is entirely possible that certain MPA cases would be classified as GPA using ACR 1990. An additional challenge was the advent of ANCA serotype testing, which latterly has been shown to be useful in classifying and diagnosis AAV sub-types, as described in section 1.4.3.

1.4.3 EMA algorithm

Published in 2006, the European Medicines Association algorithm aimed to group the CHCC, ARC 1990 and the Lanham criteria, and to resolve inconsistencies between these approaches. At that time the ACR and CHCC approaches were widely used, but no consensus regarding how they should be applied existed. Clinician interested in vasculitis met at the European Medicine Agency in 2004 and in 2006 to develop this algorithm, which can be applied to individuals with a diagnosis of AAV or PAN. The algorithm first identifies EGPA, either via the stringent and non-validated Lanham criteria (asthma, eosinophilia and vasculitis of at least two organs) or ACR 1990 criteria (Emmi et al., 2023). It then applies ACR 1990, CHCC and the presence of positive ANCA serology to positively identify, first GPA, then MPA. If all these criteria are negative then the patient may be classified as classic PAN if consistent histology or imaging are present (Watts et al., 2007).

1.4.4 2022 ACR/EULAR

The American College of Rheumatology (ACR) 1990 criteria were widely adopted and considered effective. The consensus that was achieved around classification aided the success of international multicentre randomised controlled trials (RCTs). However, there was an increasing strong case being put for revised classification criteria. Reasons included the new widespread availability of ANCA testing, increased use of cross-sectional imaging which provides new potential classification variables, the establishment of MPA as a clinical phenomenon and the declining sensitivity of the ACR 1990 criteria. The declining sensitivity may be related to more individuals being diagnosed with modern diagnostic tests including imaging and ANCA. When the ACR criteria were applied to the DCVAS cohort (described below), one-third of patients were misclassified (Seeliger et al., 2017).

An international project to develop updated criteria was led by the ACR and the European Alliance of Associations for Rheumatology (EULAR), resulting in the 2022 ACR/EULAR criteria. The criteria were developed over five stages using modern methodology. The GPA development set had 578 GPA cases and 652 comparators. Ten data items were included in the score for GPA including nasal

involvement and PR3 positivity as factors increasing the likelihood of a GPA diagnosis and elevated eosinophil count are a factor decreasing the likelihood. A score of five or more allows classification as GPA with 93% sensitivity and 94% specificity (Robson et al., 2022). Similar scores were developed for other vasculitides including MPA and EGPA. The MPA score was developed with 149 cases and 408 comparators, it yielded sensitivity 91% and specificity 94% (Suppiah et al., 2022). The EGPA score was developed with 107 cases and 450 comparators, it yielded sensitivity 85% and specificity 99% (Grayson et al., 2022).

1.5 Epidemiology

AAV represents a set of rare diseases. A comprehensive review of AAV epidemiology by Watts et al reported descriptions of the incidence rate of AAV that ranged from 13 cases per million per year in reports from Germany and Spain to 20 per million per year in reports from Sweden and Japan. In the UK, Germany and Australia, GPA was more common than MPA, which in turn, was more common than EPGA. GPA was also more common in a report from Western Montana, USA. Other nations including Greece, Spain, Canada and Japan describe higher incidence rates of MPA compared to GPA. A possible phenomenon based on latitude may be influencing GPA and MPA incidence – GPA seems to be more common in Northern European countries and MPA in southern Europe. This latitudinal effect may also have been reflected in the southern hemisphere also, in New Zealand data (Watts et al., 2015). A more recent report from Northern Norway describes incidence for AAV overall was 24.7 per million per year over 15 years from 1999 to 2013. For the individual AAV diagnoses rates were 15.6, 6.5 and 2.7 per million per year for GPA, MPA and EGPA respectively, again possibly reflecting a latitudinal phenomenon. Prevalence rates were 350 per million for AAV overall and 261, 58 and 33 per million for GPA, MPA and EGPA respectively (Nilsen et al., 2020). A recent report from Minnesota, USA described a 33 per million per year incidence rate for AAV overall and a prevalence of 421 per million (Berti et al., 2017).

There is evidence that the incidence of AAV increased in the 20 years leading up to the turn of the century. The report by Nilsen et al from Northern Norway showed an increasing trend in incidence over 15 years from 1999 to 2013. In the UK, a report for GPA only described an incidence rate of 1.5 per million per year

from 1980 to 1986, with an increase later that decade to 6.1 per million per year in 1987 to 1989. Notably the later period coincided with the introduction of an ANCA assay. The clinical phenotype of patients did not appear to change with the exception of less severe kidney involvement at the time of diagnosis and associated improved renal outcomes (Andrews et al., 1990). The increased incidence rate over this time period likely relates to improved recognition in the context of the introduction of classification criteria and ANCA testing. Increasing rates of AAV appeared to stabilise around 2000, suggesting there is not an underlying aetiological cause for the increased incidence (Watts et al., 2022).

A study utilising DCVAS data suggested there may be differences in AAV sub-type incidence relating to ethnicity. MPO-ANCA was more common in Southern Europeans, Japanese and Chinese patients, while PR3-ANCA was more common in the other groups studied (Pearce, Craven, et al., 2017). An American study from Chapel Hill, indicated that AAV was less common in Black patients (Cao et al., 2011). A UK study did not find a difference when comparing a White population to a Black/Minority Ethnic population in terms of AAV incidence (Pearce, Grainge, et al., 2017).

There are fewer prevalence studies published relating to AAV when compared to incidence studies. On review of the literature, Watts found that prevalence estimates ranged from 46 to 184 per million population (Watts et al., 2015). This has increased in recent decades, likely reflecting improved case identification and a possible reduction in mortality due to earlier identification and better therapies.

Incidence is strongly related to age. A study by Pearce and colleagues found overall incidence of 23.1 per million person-years, but this varied substantially according to age. For age groups 16 to 39 years, 40 to 54 years, 55 to 69 years, 70 to 84 years and 85 years or older the incidence rates were 3.0, 12.2, 39.7, 86.4 and 92.4 per million person-years (Pearce et al., 2016). There is variability in the reported age of peak incidence however, with some studies reporting it to be between 55 to 64, 65 to 74 or over 75 years (Watts et al., 2015). Incidence does not vary substantially depending on sex, though some studies report a possible male predominance (Pearce et al., 2016).

1.6 Aetiology

Autoimmune conditions classically are described as having both genetic and environmental determinants. The aetiology of AAV is described in such terms yet a complete understanding of this interaction remains elusive.

1.6.1 Genetics

The genetics underlying AAV have been challenging to investigate due in part to the rarity of the condition, occurrence mainly in adults, no clear sex predominance, lack of evidence of Mendelian inheritance, rare familial clustering and seeming absence of monogenic variants (Trivioli et al., 2022). Therefore, AAV genetics are likely complex and polygenic. A significant number of candidate-gene association studies have been undertaken, yielding the identification of some genetic risk loci for AAV. Genome-wide association studies have been more fruitful, confirming some of these loci and establishing several new potentially important variants. Current understanding suggests that the human leukocyte antigen (HLA) region is the most important with respect to genetic risk. The HLA complex, located on chromosome 6, encodes membrane-bound proteins crucial for regulating the immune system. Polymorphisms in this region are implicated in the pathogenesis of many autoimmune diseases. In AAV, HLA polymorphisms associate more strongly with ANCA subtype as opposed to clinicopathological diagnoses. Other sites of genetic variation of potential importance in AAV include: *PRTN3*, which encodes PR3; *SERPINA1*, which encodes α1-antitrypsin, an inhibitor of PR3 and other serine proteases; *PTPN22* which encodes an enzyme which negatively controls T cell activation; *CTLA4*, a protein similarly involved in T cell activation; and *BACH2* which encodes a transcriptional repressor involved with the function of both B cells and regulatory T cells (Trivioli et al., 2022). Epigenetics may also be important with increased histone modifications and altered methylation status being present on the *PRTN3* and *MPO* genes of AAV subjects compared to controls (Jones et al., 2017; Yang et al., 2016).

1.6.2 Environment

An extensive systematic mapping review by Scott and colleagues explored potential environmental aetiological factors for AAV (Scott et al., 2020). That disease occurs later in life and shows seasonal and temporal peaks points to an important role for environmental factors. Seasonality studies consistently report clustering of disease onset at particular times, though there is some variability in the timing of such clustering. Most report peak onset of AAV in winter (Li et al., 2018). Increased latitude has been demonstrated in several studies to be associated with increased GPA and EGPA incidence, with lower ultraviolet radiation exposure and thus lower levels of vitamin D synthesis potentially playing a role in AAV pathogenesis (Gatenby et al., 2009). Rural versus urban living has been examined with respect to AAV incidence. Studies have reported mixed effects overall, but a large study from Scotland representing a complete national cohort found that there was increased incidence of GPA, but not MPA, in rural areas (Scott et al., 2020; Aiyegbusi et al., 2021). Environmental dust exposure likely has a role in AAV pathogenesis, with the largest body of evidence supporting an effect of silica exposure. A meta-analysis in 2013 reported a 2.5 times increased risk of AAV in individuals 'ever exposed' to silica (Gómez-Puerta et al., 2013). Increased AAV incidence has been noted after several major earthquakes which resulted in increased environmental levels of silica, such as the Great East Japan earthquake in 2011 (Takeuchi et al., 2017). Other potentially important biological effects of such natural disasters, such as psychological stress, have not been explored. Associations for other environmental exposures such as industrial solvents and carbon monoxide have been identified in some studies, but consensus has not developed in the literature. Interaction with farm animals or antigens from soil have similarly been suggested, but not consistently replicated (Scott et al., 2020).

Given that geographic, seasonal and temporal clustering has been observed it is considered likely that some form of infection has an aetiological role in AAV. The strongest evidence for the role of a microorganism is for chronic Staphylococcus aureus nasal colonisation, first identified in an observational cohort study in which colonisation was associated with relapse (Stegeman et al., 1994). This has subsequently been replicated. Weak associations between seropositivity for other infections and AAV have been described, but not substantially replicated.

These include cytomegalovirus (CMV), Chlamydia pneumoniae, Helicobacter pylori and Toxoplasma gondii (Scott et al., 2020).

Various drugs have been associated with AAV, but drug-induced AAV is usually considered a separate entity to primary AAV. Such drugs include Propylthiouracil, hydralazine, minocycline and levamisole – an anti-helminthic agent often used an adulterant for recreational cocaine. Drug-induced AAV typically runs a milder course, improves with drug cessation and auto-antibodies fall with drug cessation. Occasionally severe organ involvement occurs and sometime immunosuppression is utilised (Grau, 2015).

1.7 Pathogenesis

1.7.1 MPO and PR3

The blood vessel inflammation and necrosis that leads to organ damage in AAV are mediated through the pathogenic role of ANCA directed against PR3 and MPO. PR3 and MPO are both enzymes found in azurophil granules in neutrophils. PR3 is an elastinolytic serine protease which catabolises various human proteins. In addition to being expressed in neutrophil granules, PR3 is also expressed on the cell surface of some healthy individuals and at varying frequencies. This is genetically determined and a predicative factor for developing AAV (Witko-Sarsat et al., 1999; Schreiber et al., 2003). One hypothesis is that the loss of self-tolerance in individuals who exhibit PR3 membrane expression leads to the development of AAV (Wiik, 2000). MPO is a peroxidase enzyme which exerts antimicrobial activity though the formation of highly reactive chloride-based oxidants. MPO results in the green colour of pus and due to its bright colour it was initially called verdoperoxidase (Klebanoff, 2005). There is evidence that MPO adheres to the neutrophil cell surface and has cytokine and cell adhesion mediation properties, though whether these properties occur in healthy individuals and whether they contribute to AAV pathogenesis is unknown (Lau et al., 2005; Johansson et al., 1997).

1.7.2 Development of ANCA

MPO and PR3 are not typically exposed to the immune system. When neutrophils degranulate they are promptly inhibited: MPO by ceruloplasmin and PR3 by α1-

antitrypsin. There are several hypotheses attempting to explain the development of autoantibodies to neutrophil self-antigens. One explanation is prolonged exposure of the immune system to the proteins due to defective neutrophil apoptosis or diminished elimination of apoptotic neutrophil fragments. Another is molecular mimicry, whereby antigens from microorganisms bear similar molecular structure to host proteins, leading to antibodies directs at the infectious agent targeting MPO or PR3. Auto-antibodies against LAMP2 (anti-LAMP2), a neutrophil protein expressed on the cell surface may play a role in the molecular mimicry theory. Most severely affected AAV patients are positive for anti-LAMP2 and these antibodies decrease rapidly following successful induction immunosuppression (Kain et al., 2012). Rats injected with anti-LAMP2 develop a pauci-immune focal necrotizing glomerulonephritis (FNGN) – the pathological hallmark of AAV-glomerulonephritis. The most common human LAMP2 epitope shows complete homology for FimH, an adhesin protein that enables Escherichia coli to attach to host epithelia. Rats immunised with FimH developed anti-LAMP2 and associated pauci-immune FNGN (Kain et al., 2008). A further proposal is the complementary peptide hypothesis which is less well developed. Complementary PR3 (cPR3) is a protein generated from the antisense DNA strand of *PRTN3*, the gene which codes for PR3. cPR3 peptides may be produced after infections, resulting in autoantibodies which not only react with cPR3 but also sense PR3 (Pendergraft et al., 2004). NETosis is an additional process which may contribute to the exposure of self-antigens. Neutrophil extracellular traps (NETs) are extracellular threads which trap and kill invading pathogens. They are primarily comprised of DNA and various proteins such as MPO and PR3. NETosis is the process of NET formation. Evidence for a role in AAV comes from studies of drug-induced AAV where causative drugs can induced NETosis (Lood and Hughes, 2017).

1.7.3 ANCA pathogenicity

Clinical, animal model and in vitro experiments demonstrate that ANCAs have a pathogenic role in AAV. The presence of ANCAs in up to 90% of individuals with AAV, their response to treatment and their correlation with disease activity are clinical observations consistent with this (Boomsma et al., 2000). That various drugs stimulate an AAV phenotype alongside ANCAs further supports a pathogenic role. A clinical case of pulmonary renal syndrome in a newborn

represents a human model indicating the pathogenic role of ANCA. The mother experienced resurgent MPA during pregnancy and cord blood analysis of the neonate indicated the cause of disease was placental transmission of MPO-ANCA (Schlieben et al., 2005). Mouse models where MPO-ANCA is infiltrated results in pauci-immune FNGN (Xiao et al., 2002). PR3-ANCA models have been significantly more challenging to develop due to primed murine neutrophils expressing minimal surface PR3 and lack of human PR3-ANCA reactivity with murine antigens, however, mice with a humanised immune system have been shown to develop an AAV phenotype in response to human PR3-ANCA infiltration (Little et al., 2012). In vitro incubation of human neutrophils with both PR3 and MPO leads to the release of damaging enzymes, oxygen radicals, NETosis and promotion of autoimmunity (Falk et al., 1990; Kessenbrock et al., 2009).

1.7.4 Complement

Complement activation was initially not thought to play a major role in AAV pathogenesis due to the lack of complement deposition in glomerular AAV lesions. The alternative complement pathway was, however, shown to be important in an experiment where C4 knockout mice developed disease in response to MPO-ANCA, while C5 knockout animals did not (Xiao et al., 2007). Neutrophils have been shown to be primed in murine models via C5a receptor 1, activation of this receptor promotes both ongoing autoimmunity and release of reactive oxygen species (Dick et al., 2018). The CLEAR and ADVOCATE randomised controlled trials of avacopan, a selective C5a receptor antagonist, have shown this therapy to be effective in AAV in humans, confirming the fundamental role of the alternative complement pathway (Jayne et al., 2017, 2021). It has long been recognised that primed neutrophils activate the alternate complement pathway in vitro: this likely leads to a amplification loop in AAV pathogenesis (Shingu et al., 1992).

1.7.5 From ANCA to tissue damage

The above phenomena provide the basis of how the genesis of ANCAs leads to the life-threatening tissue damage seen in AAV. Infection or inflammation lead to complement or inflammatory cytokines priming neutrophils. This leads to ANCA antigens (MPO or PR3) moving from granules to the surface of neutrophils.

ANCA, present though mechanisms described above, cause neutrophils to activate and degranulate through binding to their antigens. Monocytes can also be activated by ANCA, which can lead to granuloma formation in PR3 disease. Activated neutrophils stimulate C5a production, creating a positively reinforcing feedback loop whereby more neutrophils are recruited and activated. Neutrophils adhere to the vascular endothelium. Once activated, they cause endothelial destruction through the release of damaging enzymes and reactive oxygen species. NETosis enhances endothelial injury. Deposition of PR3 and MPO and cytokine release recruit monocytes and autoreactive T cells, increasing injury. Spill-over of plasma and coagulation products through the damaged mural structures lead to the familiar pathological lesions of fibrinoid necrosis in blood vessels and to glomerular crescents. Later, macrophages and T cells remove apoptotic neutrophils. Where tissue injury continues, fibroblasts deposit collagen leading to fibrosis and sclerosis of the damaged structures (Sinico and Guillevin, 2019).

1.8 Pathology

AAV is known as a necrotising pauci-immune vasculitis, as it is associated with no or few immune deposits, specifically immunoglobulin or complement deposition. This distinguishes AAV immunopathologically from immune complex-mediated vasculitis and anti-glomerular basement membrane antibody (anti-GBM) disease.

The pathology of AAV is characterised by segmental neutrophil-dense inflammatory and necrotic lesions. Necrotic lesions lead to physical defects in the blood vessel wall. This allows the components of plasma to flow from vessels into interstitial tissue or adjacent areas, such as the urinary space next to glomerular blood vessels and the alveoli associated with alveolar capillaries. Thrombogenic proteins, such as tissue factor, are present in interstitial tissue and are also released in response to blood vessel damage. This leads to the clotting process being initiated around the site of blood vessel necrosis and ultimately to fibrin formation, therefore the damage process associated with AAV is termed fibrinoid necrosis. A pathological picture typically present is leukocytoclastic vasculitis, commonly present in venules and arterioles. Leukocytoclastic vasculitis results when invading leukocytes undergo cell death leading to a pattern called leukocytoclasia, caused by nuclear fragmentation.

Figure 1-2 shows classic histopathological lesions in AAV. It is adapted from (Ishizu et al., 2023) and (Almaani et al., 2021) under the terms of Creative Commons Attribution-NonCommercial 4.0 License (https://creativecommons.org/licenses/by-nc/4.0/). This license requires credit to authors (above), a link to the licence (above) and description of any changes (below). Sections A. to G. were taken from (Ishizu et al., 2023). Section F. was taken from (Almaani et al., 2021), resized and cropped to match the rest of the Figure.

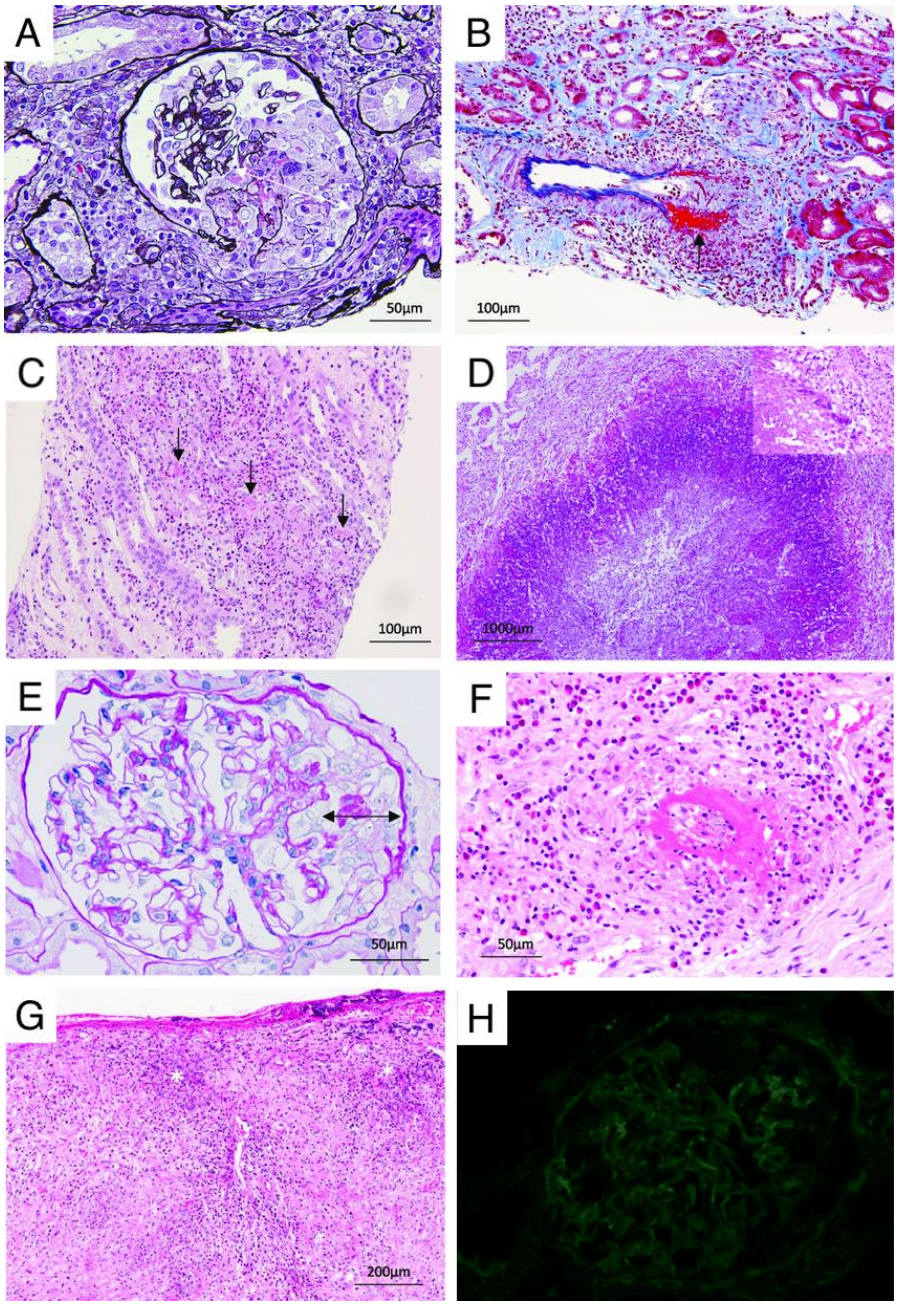


Figure 1-2 | Histopathology of ANCA-associated vasculitis
Adapted from (Ishizu et al., 2023) and (Almaani et al., 2021) under the terms of Creative Commons Attribution-NonCommercial 4.0 License (https://creativecommons.org/licenses/by-nc/4.0/).
A. Cellular crescent (white arrow) in a glomerulus in MPA (PAMS-HE staining)
B. Necrotising arteritis in a renal interlobular artery (arrow, elastic Masson staining)
C. Red cell extravasation and infiltration of neutrophils around renal medulla capillaries in MPA. Interstitial fibrin deposition (arrows, HE staining)
D. Pulmonary GPA (HE staining). Inflammatory cells, including multinucleated giant cells, forming a necrotising granuloma (inset: higher power view).
E. Cellular crescent in glomerulus (arrow), GPA (PAS staining)
F. Hepatic EGPA (HE staining). Eosinophilic infiltration and fibrinoid necrosis associated with injured vessel wall.
G. Dura mater (HE staining). AAV causing hypertrophic pachymeningitis. Microabscess-like inflammatory cell foci (asterisks).
H. Negative-to-very-weak immunofluorescence for immunoglobulins in a glomerulus, illustrating the classic pauci-immune nature of AAV.
(See abbreviation section for abbreviations of histopathological stains.)

1.9 Clinical features

The clinical signs and symptoms of MPA and GPA have substantial overlap and will be discussed first. While EGPA shares many clinical features, there are important differences which merit a separate description.

Theoretically any blood vessel in humans can be involved in GPA and MPA, leading to a wide variety of affected organs and resultant diverse presentations. Severity is highly variable, with some presenting with mild organ-limited disease, but many present with severe organ or life-threatening disease. Descriptions such as “non-severe” or “limited” disease have been used in reference to AAV initially presenting without obvious major organ involvement. However, a large majority of those presenting with such apparently mild disease will go on to develop severe organ involvement, therefore the above descriptions may have limited clinical utility. The proportion of those presenting with such disease is approximately 25%, with symptoms such as polyarthropathy and nasal crusting. They are typically younger, female, more likely to have recurring disease and more likely to have destructing upper airway disease (Stone and Wegener’s Granulomatosis Etanercept Trial Research Group, 2003). A summation of organ involvement frequency across AAV sub-diagnoses is presented in Table 1-1.

Table 1-1 | Percentage organ involvement at time of diagnosis in MPA, GPA and EGPA

	MPA	GPA	EGPA
Constitutional	86%	78%	70%
Cutaneous	30%	35%	50%
ENT	26%	82%	59%
Mucous membrane or eyes	13%	38%	-
Respiratory	63%	63%	100%
Cardiovascular	15%	11%	28%
Abdominal	22%	19%	35%
Renal	82%	59%	21%
Neurological	37%	31%	65%

ENT = ear, nose and throat. Percentages for MPA and GPA are derived from DCVAS data reproduced in (Kronbichler et al., 2024). EGPA data is median percentages in the clinical features articles featured in (Sinico and Guillevin, 2019).

Constitutional symptoms are common in AAV, including EGPA. Typical such symptoms include anorexia, weight loss, fever, malaise, myalgia and arthralgia.

These are often the first to develop and are notably non-specific. As a result, the classic presentation of AAV is often vague, at least before more obvious organ-specific features develop. Many individuals with AAV are therefore misdiagnosed with other more common conditions that can present with similar symptoms such as infection, cancer and inflammatory joint disease.

ENT presentations include hearing loss (both conductive and sensorineural), earache, otorrhoea, otitis media, sinusitis, nasal discharge including epistaxis, nasal ulcers, oral ulcers and polychondritis. GPA patients are much more likely to have destructive disease affecting bone and cartilage leading to saddle nose deformity, cranial nerve entrapment and upper airway and retroorbital masses.

Respiratory manifestations can involve the airways and lung parenchyma. Symptoms include breathlessness, cough, haemoptysis, stridor, hoarseness, wheeze and pleuritic pain. Examination findings include those of tracheal or subglottic stenosis, pleural effusion and consolidation. Interstitial lung disease and pulmonary hypertension can occur, more commonly with MPA or MPO-ANCA disease, and may represent a separate disease entity (Sebastiani et al., 2020). Chest imaging may reveal a diverse set of features such as adenopathy, nodules, opacification and patchy or diffuse opacification.

Kidney involvement in GPA and MPA is classically a rapidly progressive glomerulonephritis represented by rising serum creatinine, haematuria, proteinuria and hypertension. Urine microscopy typically reveals red cell casts. Proteinuria is typically in the sub-nephrotic range. Kidney involvement may be severe and require dialysis on presentation. In GPA 20% will have excretory renal dysfunction at diagnosis, but this increases over time with around 80% going on to have renal involvement (Hoffman et al., 1992). Renal dysfunction is more common in MPA at presentation.

Skin manifestations are common. The classic lesion is palpable purpura – a “vasculitic rash”. Other skin presentations from most to least common are ulcers, livedo reticularis, nodules and urticaria. Eye involvement may present as ophthalmoplegia, episcleritis, scleritis, uveitis, conjunctivitis, corneal ulceration, retinal vasculitis, nasolacrimal duct blockage and retro-orbital masses. The typical nervous system lesion is mononeuritis multiplex, but central

nervous system masses, granulomatous meningeal disease, sensory neuropathy and cranial nerve involvement may occur. Venous thromboembolism is more common in AAV and may be part of a non-classical presentation. Less common presentations include clinically apparent gut involvement, genitourinary manifestations and cardiac disease. Pericarditis, myocarditis and dysrhythmias can occur (Sinico and Guillevin, 2019).

EGPA shares many clinical features with GPA and MPA, but there are important differences, including the frequency of involvement of various organs. EGPA is a systemic vasculitis almost always accompanied by asthma. It is likely to be associated with allergic rhinitis or nasal polyps and significant eosinophilia. Any organ system can be involved but respiratory manifestations are most common, followed by cutaneous. Involvement of the nervous system and heart are more prominent than in GPA or MPA. Renal involvement is less common than other AAVs but can be severe. It generally manifests in three stages: late-onset severe asthma occurs first, sometimes with allergy features; this is followed by lung infiltrates and eosinophilia; with features of systemic vasculitis becoming apparent later.

1.10 Diagnosis

Diagnosis is challenging in AAV. Often patients attend multiple specialists in the months leading up to an AAV diagnosis. Pearce et al reported that 20% of individuals attended more than one specialist in the period prior to diagnosis (Pearce, Hubbard, et al., 2018).

Jayne described a pathway for vasculitis diagnosis, which can be applied to AAV specifically. This first includes identifying a compatible clinical syndrome, which often requires a high index of suspicion on the part of the clinician. A chronic inflammatory condition for which a diagnosis remains elusive is a typical clinical scenario where vasculitides such as AAV should be considered. This is followed by non-invasive investigations such as ANCA serology or diagnostic imaging. If there is a plausible clinical phenotype with test results supporting the diagnosis, then tissue biopsy should be sought to confirm disease histologically. Disease mimics and secondary causes should be considered and excluded. Mimics include infective endocarditis and paraneoplastic syndromes. Secondary causes of

vasculitis include infections such as tuberculosis and drugs such as those described in Section 1.6.2. Lastly, clinical observation over time can help improving diagnostic certainty (Jayne, 2009).

Notably there are no published diagnostic criteria for AAV. The use of classification criteria has often been extended to diagnosis, beyond their intended scope for use in research studies. Classification criteria are described in Section 1.4. Such criteria are likely overly specific and poorly sensitive for diagnostic purposes, but they may assist clinicians where applied with the knowledge that those with an incomplete clinical syndrome may be missed. Surrogates that would enable a diagnosis of GPA over MPA include features such as destructive processes in nasal cartilage or bone and destructive nodular or cavitating lung lesions. These features may be apparent clinically in the case of saddle-nose deformity, on endoscopy or on imaging.

1.11 Management

Therapy in AAV is classically described in two phases. The remission induction phase aims to promptly bring about suppression of disease activity. Intense immunosuppressive therapy is typically used, especially in the case of organ or life-threatening disease. Disease remission is ideally achieved within three months. The induction phase is followed by a maintenance phase, whereby less intense immunosuppression is used to prevent relapse of disease. Representation of EGPA in important trials of AAV therapy is low, due to the rarity of the condition and potential differences in underlying pathogenesis. EGPA will briefly be discussed after reviewing remission induction and maintenance therapy for MPA and GPA.

1.11.1.1 Remission induction

Oral glucocorticoids have been a cornerstone of AAV induction treatment for several decades. In combination with other agents, this transformed the outlook for AAV patients: AAV was once considered to be universally fatal, whereas now the vast majority of patients survive over the course of the first year of therapy (Fauci et al., 1971). Intravenous methylprednisolone is typically used at the start of therapy, though this is not underpinned by controlled studies. While ongoing

high dose oral glucocorticoids are commonly used for ongoing management, adverse effects such as severe infections are well recognised. The PEXIVAS trial was a two-by-two factorial design RCT including one arm which evaluated plasma exchange while the other examined reduced dose glucocorticoid therapy (Walsh et al., 2020). It is the largest RCT yet to be performed in AAV. It found that a glucocorticoid regimen with a substantially reduced total cumulative dose was similar to the standard dose regimen in terms of achieving disease remission, but there were fewer severe infection events. Similar results were achieved in the LOVAS trial, a multicentre RCT based in Japan (Furuta et al., 2021).

Cyclophosphamide, in combination with glucocorticoids, transformed the outlook for AAV patients when it was first studied in individuals with GPA in the 1970s. Prior to the use of cyclophosphamide, as described above, AAV was considered universally fatal. Fauci and colleagues, working at the National Institute for Health (NIH) in Bethesda, Maryland, demonstrated that over 90% of cyclophosphamide and glucocorticoid treated patients achieved complete remission (Fauci et al., 1983). The application of cyclophosphamide has been explored in various RCTS. It can be administered by either the oral or the intravenous route as a pulsed therapy. These approaches were compared in the CYCLOPS trial. There were no significant differences in major outcomes such as mortality, renal outcomes or adverse effects. There was a lower cumulative dose and less leukopenia in the intravenous group, however there were fewer disease relapses in the oral group at 20.8% compared to 39.5% (Harper et al., 2012).

B cell depletion using rituximab has been employed in both induction and maintenance strategies in AAV. The RAVE and RITUXVAS trials showed that rituximab regimens were non-inferior with respect to remission induction compared to cyclophosphamide-based regimens. No difference in adverse events was detected. RAVE did not enrol individuals with severe renal disease, while RITUXVAS did. Notably RITUXVAS included two or three pulses of intravenous cyclophosphamide as part of its regimen, therefore there is no RCT evidence that rituximab alone has efficacy in the setting of severe renal disease. RAVE included patients with relapsing disease where rituximab demonstrated greater

efficacy over cyclophosphamide (Stone et al., 2010; Jones et al., 2010). These trials are the basis for recommendations that rituximab and cyclophosphamide are equally effective at remission induction, except for the setting of relapse where rituximab is recommended.

The C5a receptor inhibitor avacopan represents a significant advance in AAV induction therapy. In the ADVOCATE RCT avacopan added in to standard induction regimens with minimal glucocorticoid use was compared to a regimen utilising a tapering glucocorticoid regimen. Avacopan was non-inferior to glucocorticoid taper at 26 weeks and superior at one year with respect to sustained disease remission. Overall adverse events were similar. The avacopan group had a mean cumulative glucocorticoid dose that was approximately one third that of the comparator group (Jayne et al., 2021).

Plasma exchange is considered an adjunctive therapy in severe AAV. Its mechanism is not fully understood, though removal of pathogenic ANCA and cytokines may play a role. PEXIVAS showed a numerical reduction in the primary composite outcome of death or end-stage kidney disease (ESKD), but this finding was not statistically significant (Walsh et al., 2020). A subsequent meta-analysis of nine trials including over one thousand participants showed no important effect on mortality, but a significant reduction in the risk of ESKD and an increased risk of severe infection. Therefore plasma exchange remains a therapeutic option in some patients with severe disease such as severe renal involvement or diffuse alveolar haemorrhage (Walsh et al., 2022).

Other therapies that can be employed for remission induction include methotrexate, mycophenolate mofetil and intravenous immunoglobulin (IVIG). The initial results of the NORAM trial showed non-inferiority of methotrexate compared to cyclophosphamide in the setting of non-severe AAV, but a later longer term analysis demonstrated increased use of glucocorticoids and worse disease control (Faurschou et al., 2012). The MYCYC trial compared mycophenolate mofetil to cyclophosphamide in the setting of non-severe relapse. This study did not show mycophenolate mofetil to be as effective. Therefore it is not usually considered an option for first line treatment (Tuin et al., 2019). IVIG is not routinely used but can be employed in severe disease, refractory disease or typical immunosuppression is not deemed safe such as

ongoing severe infection. One small placebo controlled trial of 34 patients showed reduced disease activity, though this effect was not sustained (Jayne et al., 2000).

1.11.1.2 Maintenance

Long-term follow-up of initial studies of cyclophosphamide and glucocorticoids of patients with GPA at the NIH showed that the condition has a high propensity to relapse without maintenance therapy (Hoffman et al., 1992). The CYCAZAREM trial showed that azathioprine did not have increased rates of relapse compared to cyclophosphamide (Jayne et al., 2003). Although the rates of adverse events were similar in the two arms, azathioprine maintenance became the standard of care given the recognised toxicity associated with long-term cyclophosphamide use. The MAINRITSAN series of RCTs from the French Vasculitis Study Group have explored the use of rituximab as maintenance therapy. MAINRITSAN (the first of three trials in the series) showed a decreased relapse rate and similar rates of adverse events with rituximab when compared to azathioprine. The outcome of MAINRITSAN has recently been confirmed in RITAZAREM, an international multicentre RCT comparing rituximab and azathioprine (Smith et al., 2023). MAINRITSAN2 compared a standard schedule of rituximab to tailored dosing guided by plasma B-cell repopulation. There was no difference in relapse rate, but the tailored dosing group received few infusions of rituximab (Charles et al., 2018). After the first two years of treatment with rituximab, MAINRITSAN3 compared two further years of rituximab therapy to placebo. Relapse free survival was 97% in the prolonged rituximab group and 74% in the placebo group, serious adverse events being similar in both arms (Charles et al., 2020).

The REMAIN trial explored duration of maintenance therapy, comparing 24 months of therapy with azathioprine or glucocorticoid with 48 months. Higher relapse rates were found with the early withdrawal group, but more adverse events in the continuation group. An optimal strategy for maintenance therapy duration remains unclear (Karras et al., 2017). Other agents trialled for use as maintenance therapy include methotrexate, mycophenolate mofetil and etanercept but these have not demonstrated advantages over the standard of care (Kitching et al., 2020).

1.11.2 Treatment of EGPA

Severe EGPA is typically managed in a similar fashion to MPA and GPA. Severity can be defined using the five-factor score. This scoring includes low renal excretory function, proteinuria, gastrointestinal involvement, cardiomyopathy and central nervous system involvement. A small RCT of participants with at least one five-factor item indicated that 12 pulses of cyclophosphamide were better at controlling severe disease compares to 6 pulses (Cohen et al., 2007). Mepolizumab is an anti-interleukin 5 monoclonal antibody therapy and is used in chronic eosinophilic asthma. An RCT in refractory or relapsing EGPA using this therapy showed increased time in remission, fewer relapses and lower glucocorticoid exposure. Remission did not occur in 47% of the mepolizumab group, demonstrating the resistance of this sub-set of EGPA to currently available therapy (Wechsler et al., 2017).

Non-severe EGPA is often managed with glucocorticoids and additional immunosuppressive agents to limit the high glucocorticoid doses often required to control disease activity. A small trial in vasculitis patients included a high proportion of EGPA patients and compared the addition of azathioprine to glucocorticoids alone. This did not show improved outcomes in terms of disease activity or steroid sparing (Puéchal et al., 2017).

1.12 Outcomes

1.12.1 Survival

Published in the British Medical Journal, one of the earliest reports of outcomes in AAV was of 56 patients from Northern England. These individuals had GPA, then described as “giant-cell granuloma of the respiratory tract”. Early mortality was high, with 44 of 56 (79%) patients dying within 12 months of diagnosis. Half of the patients died of uraemia, while the other common causes of death were respiratory failure, cardiovascular causes and sepsis (Walton, 1958). An analysis of four EUVAS trials explored AAV outcomes in the era of effective immunosuppressive therapy. Survival was 89.3%, half of those who died did so following an infection (Little et al., 2010). A later population-based study from southern Sweden covered the era during which rituximab treatment was

introduced. The study reported one-year, two-year, five-year and ten-year patient survival in 195 patients at 87%, 82%, 70% and 55% respectively. Mortality was substantially increased compared to an age, sex and time-matched comparator group. (Heijl et al., 2017).

1.12.2 Relapse

With modern therapy, disease remission is achieved in a high proportion of patients. In the RAVE trial, defined as a Birmingham Vasculitis Score (BVAS) of zero, remission was achieved in 86% of participants (Miloslavsky et al., 2013). In an analysis of four clinical trials undertaken by the European Vasculitis Study Group (EUVAS), 38% of 535 participants experienced at least one relapse over five years (Walsh et al., 2012). The competing risk of death without relapse should be taken into account, notably 18% died without experiencing disease relapse. Relapse is not necessarily associated with worse mortality, as many relapses are ENT related. However, such relapses are still associated with reduced quality of life and increased exposure to potentially toxic therapies (Sinico and Guillevin, 2019). Relapse rates may now improve in the era of rituximab: long term analysis of the MAINRITSAN trial showed relapse-free survival rates of 71.9% for major relapses and 57.9% for all relapses at 5 years (Terrier et al., 2018).

1.12.3 Renal Outcomes

Although not always present at diagnosis, the prevalence of renal involvement in AAV is 75 to 95%. Kidney disease is typically worse in MPA or MPO-ANCA disease compared to GPA or PR3-ANCA disease, this is in part due to extrarenal manifestations leading to earlier presentation and resultant earlier treatment in the latter phenotypes. The Berden classification derived histological scoring to predict renal outcomes. In a validation cohort at five years, renal survival was 93% for the focal group, 76% for the crescentic class, 61% for the mixed class and 50% for the sclerotic class (Berden et al., 2010).

1.12.4 Comorbidities and adverse events

Severe infection is the most common adverse event in the first year of therapy for AAV and is discussed in greater depth in section 0.

The Vasculitis Damage Index (VDI) is a comprehensive, validated list of 64 items relating to incident comorbidities and adverse events following AAV diagnosis (Exley et al., 1997). All accrued damage is recorded, regardless of whether the cause is considered to be vasculitis related, treatment related or otherwise. As it records chronic damage, acute events such as severe infection are not included. Robson and colleagues used VDI items to analyse data from six EUVAS RCTs to explore the frequency of incident comorbidities and adverse events in MPA and GPA. Long-term follow up data were available for 87% of patients. At a mean of 7 years after diagnosis, the most frequent VDI items were hypertension (41.5%), osteoporosis (14.1%), malignancy (12.6%) and diabetes (10.4%), with the overall burden of damage increasing over time. Complications potentially related to glucocorticoid toxicity such as diabetes, osteoporosis and cataract (which occurred in 9.3%) are notably prominent in this data. Cardiovascular disease is a substantial cause of morbidity and mortality in AAV patients (Wallace et al., 2020). In the study by Robson, cardiovascular items were frequent at long-term follow up with angina/coronary bypass, stroke and myocardial infarction having occurred in 8.1%, 3.7% and 4.4% respectively. There was no direct comparison to rates of complications expected in the general population (Robson et al., 2015). Venous thromboembolism (VTE) is not included in VDI, but is recognised to be higher in AAV. The WeCLOT study prospectively enrolled individuals with GPA. It found a 7 per 100 person-years incidence rate of VTE, higher than reported rates in the general population and other autoimmune conditions (Merkel et al., 2005). In the rituximab era, hypogammaglobulinemia has become recognised as a complication of rituximab therapy in a non-dose dependent fashion. In one study, 4.2% of rituximab treated patients required immunoglobin replacement for recurrent infection (Roberts et al., 2015).

Sarica and colleagues investigated multimorbidity in AAV through a Scottish population-based study of over 543 AAV patients. Patients were individually matched by age, sex and locality to 2,672 general population controls. Median 5.1 years of follow-up data was available. Substantially increased risk of several morbidities was identified. In order of decreasing incidence rate ratios these included: osteoporosis, pulmonary circulation disorders, hypothyroidism, valvular heart disease, hypertension, cardiac dysrhythmias, chronic respiratory

disease, diabetes mellitus and cardiovascular disease. Notably hypertension and cardiovascular disease were the most frequently occurring morbidities in AAV patients at 19.7% and 12.6% respectively, but the magnitude of difference was less compared to the general population relative to other conditions. Hypertension and cardiovascular disease occurred at 9.4% and 9.5% respectively in the general population. Osteoporosis had the greatest difference in terms of magnitude. AAV patients had a 5.4% frequency of osteoporosis compared to 0.8% of controls, equating to an incident rate ratio of 8.0 (Shifa H. Sarica et al., 2020).

1.12.5 Quality of life

The importance of quality of life (QOL), in addition to clinical outcomes such as survival and disease remission, is increasingly being acknowledged as of high importance to patients. Importantly patients and physicians have differing perspectives on the relative importance of such outcomes. A multicentre international survey of the burden of disease in vasculitis patients, a large majority of whom had AAV, was conducted between 2006 and 2007. This work concluded that fatigue, loss of energy, weight gain, joint paint and sinusitis were the highest ranked symptoms of concern for patients. Ninety-five percent of patients reported fatigue and energy loss, most rated this severe. Severe organ involvement was considered less of a priority. Pain, musculoskeletal symptoms, financial aspects and anxiety were also prominent concerns (Herlyn et al., 2010). A multicentre case-control QOL study by Basu and colleagues compared 410 AAV cases to matched chronic disease and general population controls. Across physical and mental domains, AAV patients had similar QOL to chronic disease controls, but substantially worse QOL when compared to the general population. Fatigue, sleep disturbance, depression and anxiety were symptoms identified as strongly associated with poor QOL in a multivariable model. High serum C-reactive protein levels and high glucocorticoid dose were additional disease-related factors identified in the model, these may relate to disease activity and are potentially modifiable. Only 7% of AAV group had ESKD, therefore the effect of advanced kidney disease in AAV patients may not be accounted for in this study (Basu et al., 2014).

1.12.6 Fertility and pregnancy

Fertility can be adversely affected, both by disease activity and certain AAV therapies. Chronic kidney disease (CKD) is a recognised cause of reduced fertility in women and men (Dumanski and Ahmed, 2019). Data in systemic lupus erythematosus demonstrates a high incidence of ovarian failure with cyclophosphamide treatment. Higher cumulative dose appears to be a risk factor (Mok et al., 1998). Cyclophosphamide can severely affect sperm counts in men, though there is more potential for recovery on the cessation of therapy (Cigni et al., 2008). Teratogenicity is an important concern in AAV therapy. Cyclophosphamide, mycophenolate mofetil and methotrexate are teratogenic and rituximab depresses neonatal B-cells (Sinico and Guillevin, 2019).

1.13 Severe infection

1.13.1 Background

The evolutionary success of microorganisms has enabled them to occupy virtually every ecological niche on Earth. This includes utilising other organisms as potential habitats, such as humans and other mammals. Where this relationship is beneficial to the host, it is described as symbiosis, but where it is detrimental infection results. Infection is the pathological invasion of body tissues by microorganisms, followed by reproduction in the host and the subsequent host immune response to the pathogen and any associated toxins. Most commonly caused by bacteria and viruses, infections can also be caused by fungi, parasites and prions. The spectrum of infection varies from asymptomatic colonisation to severe illness, involving tissue destruction due to microbial toxin release or a damaging inflammatory response. The nature of a toxin, and the site of its action; or the site of bacterial replication, and type of immune response induced, determine the phenotype of the associated disease. Where the immune response involves overwhelming systemic inflammation, sepsis occurs, with associated circulatory compromise and frequently death (Maskell and Wood, 2020). As a clinical phenotype representative of severe infection, sepsis is a major cause of morbidity and mortality globally. A retrospective cohort study of 2.9 million individuals admitted to hospital in the United States found a sepsis incidence of 6% (Rhee et al., 2017). An analysis for the Global Burden of Disease

Study estimated that 48.9 million cases of sepsis occurred in 2017. These cases led to 11 million deaths, accounting for 20% of all deaths globally (Rudd et al., 2020). Modern management of infection including antimicrobials, vaccination and infection control measures have changed the outlook for individuals with severe infection.

1.13.2 Immune-mediated inflammatory disease and infection

Individuals with immune-mediated inflammatory diseases (IMID) typically are at increased susceptibility to infection. AAV is an archetypal example of this phenomenon and will be discussed in the next section (1.13.3). The mechanisms underlying vulnerability to infection in IMID likely relate to factors due to the underlying disease processes, such as immune dysregulation, and immunosuppressive therapy aimed at treating the disorder in question. Immunosuppressive therapies target different immune pathways. Depending on the pathway targeted, susceptibility to different organisms may be apparent (Doran et al., 2002; Cannon et al., 2023).

A large US based registry study of over 10,000 patients with IMID including RA, inflammatory bowel disease, psoriasis and spondyloarthropathies found high rates of serious infections. The RA group had approximately 8 events per 100 person-years. A dose-dependent relationship was demonstrated between baseline glucocorticoid use and infection rates (Grijalva et al., 2011). A systematic review of a broad range of connective tissue diseases found that 29% of patients developed a serious infection and, of those patients, there was a 24% mortality rate due to the infection. Typical bacterial infections represented the majority of these events, though opportunistic infections, such as fungal infections, also occurred. In a small number of the included studies, glucocorticoid exposure was studied. Use of intravenous methylprednisolone and overall cumulative glucocorticoid exposure were identified as prognostic factors (Falagas et al., 2007). A systematic review carried out to inform EULAR vaccination recommendations described the incidence and prevalence rates of vaccine preventable infections in individuals with autoimmune inflammatory rheumatic diseases. It reported increased risks of influenza, VZV, human papillomavirus and pneumococcal infections in the disease groups when

compared to the general population. Prevalence of hepatitis B was not different to the general population based on serological data (Furer et al., 2019).

Focusing on studies of specific IMIDs, a retrospective cohort study matched RA patients to general population controls. A hazard ratio of 1.83 was found for increased risk for hospitalisation related to infection for the RA group (Doran et al., 2002). Rituximab is a commonly utilised therapy in a range of IMIDs, including RA. A study pooled clinical trials of RA patients who received rituximab. It found a 4.3 per 100 patient-year rate of serious infections (van Vollenhoven et al., 2010). A systematic review of RCTs and observational studies compared glucocorticoid exposure to non-exposure in RA. Observational studies found a significant increase in risk associated with glucocorticoids, with relative risk 1.67. Analysis of RCTs in the report found a null effect, but with wide confidence intervals such that both clinically important increased risk and decreased risk were plausible. That individuals in observational studies tended to have increased cumulative glucocorticoid exposure compared to those in RCTs may partially explain this finding (Dixon et al., 2011). A large retrospective cohort study of US claims data in RA found a dose-dependent relationship for increased infection risk with increased glucocorticoid exposure over a prior 90-day window (George et al., 2020). The same group analysed the impact of low dose glucocorticoid exposure and described an increased risk for those exposed to up to 5 mg daily prednisone equivalents. The incidence of infection necessitating admission to hospital was 8.0 per 100 person-years in the non-exposed group, compared to 11.7 per 100 person-years in the exposed group (George et al., 2020). A case-control analysis of Canadian data in the setting of elderly patients with RA showed that while current and recent glucocorticoid exposure carries the highest associated risk, exposure over the prior 2-3 years also carries risk of infection (Dixon et al., 2012).

A prospective multicentre study of 1,000 SLE patients found that over 10 years of follow up, 36% presented with infection. Mortality was 6.8%, with infection being cited as one of the most common causes at 25% of those who died (Cervera et al., 2003). A Canadian matched cohort study utilised administrative health data comparing over 5,000 individuals with SLE. It reported adjusted hazard ratios of 1.82 for first severe infection and 1.61 for infection-related death

(Zhao et al., 2021). In a report of SLE patients treated with rituximab, prognostic factors for serious infections included the presence of CKD and higher background glucocorticoid exposure (Sun et al., 2024). Sjögren's syndrome is also associated with severe infections. A French nationwide population-based retrospective study of over 25,000 individuals with Sjögren's syndrome found an increased risk of hospitalisation due to infection with an adjusted hazard ratio of 1.29, when compared to matched controls (Goulabchand et al., 2022).

Individuals with IMID are susceptible to severe infections, both typical bacterial or viral infections and opportunistic infections such as PCP. Host and treatment factors likely play a role. AAV is no exception, and the following section will provide evidence that individuals with AAV are among the most vulnerable to this potentially life-threatening complication.

1.13.3 AAV and infection

Infection is recognised as one of the most important complications of AAV and its therapy (Kitching et al., 2020). For this thesis, a systematised review (defined below) was carried out to explore the incidence, prognostic factors and outcomes of severe infection in AAV. Defining severe infection in the epidemiological setting can be challenging. Many studies use an adapted form of the criteria for severe infection described in the Common Terminology Criteria for Adverse Events, initially developed for use in oncology clinical trials (Colevas and Setser, 2004). CTCAE grades for a typical infection are described in Table 1-2. Definitions vary in epidemiological studies, but often represent Grade 3 and above such that a severe infection is one that results in the need for hospital admission, intravenous antimicrobial therapy or death.

Grade	Descriptor
1	N/A or minimally invasive non-drug intervention indicated
2	Oral therapy indicated
3	Intravenous therapy or invasive procedure indicated
4	Life-threatening infection or urgent intervention indicated
5	Death

Table 1-2 | Common Terminology Criteria for Adverse Events example grades for infection

1.13.3.1 Systematised review

A systematised review is a methodological exploration of the scientific literature that does not meet all the requirements to be designated a systematic review. As is the case with the current review, often only one reviewer performs a systematised review and a formal risk of bias assessment may be excluded (Booth et al., 2022). Literature searches were performed in early 2020 to identify deficiencies in the prognosis research literature relating to AAV and infection. This process directly informed the scientific questions addressed in this thesis. A formal literature search of two major biomedical academic databases, Medline and Embase, was performed on 1 March 2022. This ensured all relevant studies had been used to inform this thesis, including the current introductory chapter. Studies from early 2020 and prior are described in this section, as these informed the development of thesis scientific questions. Relevant studies from after this period are considered in the appropriate substantive thesis chapter discussions. The search strategies can be found in 9.1. An adapted list of terms designed to identify epidemiological studies was used (Li et al., 2019). The search yielded 1,301 and 3,350 results from Medline and Embase respectively. After removal of duplicates there were 3,609 unique records. Article titles and abstracts were screened, leading to 342 articles to be considered for inclusion. Prospective studies, multicentre studies, population-based studies, studies with appropriate methodology and those with sufficient size for the study question were prioritised for inclusion. Where many studies reiterated similar findings to more rigorous studies, where only one AAV subtype was studied or multiple IMIDs were studied, such articles were not necessarily included. Where many studies were summarised by a systematic review, reference will be made to such a review, as opposed to summarising all studies contained within the work.

1.13.3.2 Incidence

Observational studies report variable rates of infection in AAV. Variable patient inclusion criteria, variable definitions of and degrees of severity and variable follow-up time make comparison between studies challenging. One of the earliest reports of long-term outcomes relating to infection in AAV comes from the NIH group led by Anthony Fauci which pioneered treatment with

cyclophosphamide and glucocorticoids. This NIH cohort comprised 158 patients with GPA in whom 46% developed an infection leading to hospitalisation over a mean follow-up period of eight years, ranging from 6 months to 24 years, totalling 1,229 patients-years (Hoffman et al., 1992).

A report by Reinhold-Keller and colleagues, described an early experience of patients with AAV including the long-term outcomes of 155 consecutive patients with GPA in Germany. Patients were diagnosed between 1966 - 1993 and were followed for median seven years. Most were treated with cyclophosphamide and glucocorticoids, while approximately half were treated with trimethoprim/sulfamethoxazole (TMP/SMX) and half with methotrexate at some stage. There were 56 serious infections requiring hospitalisation reported, occurring in 41 patients – 26% of the cohort. Most infections were described as pneumonia or sepsis. Three patients developed a CMV infection and one had PCP infection (Reinhold-Keller et al., 2000).

A multicentre registry report of patients managed with a modern cyclophosphamide-based regimen comes from the Glomerular Disease Collaborative Network (GDCN). McGregor analysed the outcomes of 147 AAV patients in this US glomerular disease registry, covering several states. The study aimed to examine the impact of glucocorticoid exposure from six months following diagnosis, therefore notably patients who died before six months were excluded. Patients with ESKD at presentation or lacking 12 months of sufficient follow-up data were also excluded. Patients diagnosed over 10 years from the year 2000 were included and all had induction therapy with cyclophosphamide and glucocorticoids. Evaluating the whole cohort, 87 patients (59%) experienced at least one infection over median follow-up of just under 3 years. A subsequent study from the same group examined a broader cohort from the GDCN registry. The cohort of 489 patients included biopsy-proven AAV patients treated with cyclophosphamide or rituximab-based regimens. Median follow-up was 2.8 years. Importantly the authors again did not include patients with ESKD at presentation, a group more vulnerable to infection. Infection rates were reported at 1, 2 and 5 years. The cumulative incidence of any infection was 51%, 58% and 65% at the respective time points, while the equivalent rates for severe infection were 22%, 23% and 26%. A significant proportion experienced three or

more infections: 11% over 1 year and 22% over 2 years. Patients who suffered multiple infections were also more likely to suffer a severe infection. Antimicrobial prophylaxis was used in many patients but was not universal. Proportions of patients receiving prophylaxis were not possible to ascertain. Pneumocystis pneumonia occurred in only one patient at 6 weeks following diagnosis (McGregor et al., 2012, 2015).

A large retrospective cohort study from Beijing, China described infection rates and survival in 398 consecutive patients with AAV-GN. Immunosuppression was cyclophosphamide-based and more intense than modern regimens with slower prednisone wean and longer courses of cyclophosphamide. PCP prophylaxis was not used routinely. Median follow up was just over two years. Severe infection occurred in 44% of the cohort over all follow-up. Most infections, 82%, were in the first year of therapy. The one-year infection-free survival rate was 61% (Lai et al., 2014).

Other cohort studies include the Spanish Registry of Systemic Vasculitis (REVAS) retrospective study, where 40% of 450 AAV patients developed an infection over median 6.8 years follow-up. Opportunistic infections occurred in 15%, including 14 cases of pneumocystis pneumonia and 12 cases of CMV. These patients were treated with regimens that predated widespread use of rituximab. An era comparison was performed, showing higher incidence of bacterial infection before 2000 at 55%, compared to after at 33%. Differing patterns of immunosuppression may have contributed to this finding (Solans-Laque et al., 2013). Remission Induction Therapy in Japanese patients with ANCA-associated vasculitis (RemIT-JAV) was a national, prospective cohort study including patients diagnosed with AAV in 2009 and 2012. An analysis of 156 registry patients identified 63 severe infections in 42 patients within six months of induction therapy, an incidence rate of 88 per 100 patient-years. Pulmonary infections were the most common infection site, followed by VZV skin infections. There were 23 opportunistic infections: 12 cases of fungal pneumonia, six PCP, four CMV respiratory infection and one case of respiratory tuberculosis (Watanabe-Imai et al., 2017). A retrospective Korean study examined 154 AAV patients. 14.9% had at least one hospitalisation due to infection in the first year, there were no deaths in the first year, giving a severe infection-free survival

rate of 85.1. Five year and ten year equivalent rates were 77.9% and 72.7% respectively (Yoo et al., 2018).

In the UK, retrospective survey data from the north of England showed that 24% of patients had an infection within the first 6 months of treatment with cyclophosphamide (Pearce, McGrath, et al., 2018). Scottish multicentre population-based data examining 379 AAV cases has shown that 35.6% of AAV patients developed a severe infection, 55.4% developed a laboratory-confirmed infection and 74% received an antibiotic prescription from primary care over median 3.5 years follow-up. The rates of severe infections were highest in the first 30 days following diagnosis at over 400 events per 1000 person-years. Rates decreased over time, but remained substantially higher than controls at all time points, including at 8 years following diagnosis (Sarica et al., 2018).

A systematic review and meta-analysis of studies of AAV patients treated with rituximab included over 1400 patients. It reported a cumulative incidence of 15.4% for severe infection. Median follow-up ranged from 0.6 to 5 years across studies. Opportunistic infection occurred in 1.5% with 0.2% PCP infections. The overall incidence of severe infection was estimated to be 6.5 per 100 person-years. The incidence of PCP was 1.1 per 100 person-years. There was significant heterogeneity across studies (Thery-Casari et al., 2020).

Case-control population-based cohort studies provide evidence of the rates of events in AAV patients compared to general population matched controls. Such a study from the southern Sweden compared 186 AAV patients to 744 matched controls. Median follow-up was 4.8 years and 6 years for cases and controls respectively. AAV cases experienced 116.2 severe infection events per 1000 person-years compared to 25.6 events per 1000 person-years in the controls, an incidence rate ratio (IRR) of 4.53 (Mohammad et al., 2017). The multicentre study from Scotland by Sarica, described above, compared 179 AAV cases to 1859 controls. Severe infection, laboratory-confirmed infection and prescription of antibiotics by primary care was more likely in AAV patients compared to population controls with incidence rate ratios 4.4, 7.3 and 2.2 respectively. Co-trimoxazole prophylaxis was prescribed to 76% of the AAV group. *Escherichia* was the most frequently observed genus in both groups, IRR 4.9. The pathogens with the highest incidence relative to controls were *Herpes*, *Candida* and *Clostridium*

with IRRs 12.5, 11.4 and 9.2 respectively. Varicella zoster virus (VZV) was not evaluated. (Sarica et al., 2018).

Understanding of severe infection incidence in AAV is hampered by studies with significant limitations. Such limitations include retrospective design, small sample size and lack of important data items to provide context. Retrospective design can result in incidence studies which are vulnerable to survival bias – patients who survive for longer may be more likely to be included in retrospective studies. This is potentially problematic when assessing severe infection incidence, as severe infection is associated with mortality. Retrospective studies may be biased against inclusion of patients with severe infection, potentially leading to underestimation of the incidence. Retrospective studies are vulnerable to non-standardised and missing data. Data in such studies is frequently not standardised for research purposes, as the reason for its collection is non-research related such as for clinical or administrative purposes. Where important variables are not available in the information source for the study, such as the clinical record or administrative record, then it is often impossible to retrospectively acquire this data. Retrospective studies are also more vulnerable to p-hacking, where multiple analyses are performed until “interesting” or “significant” results are obtained. Prospective studies with pre-specified analyses are less likely to suffer from these issues and are more likely to have high quality data. Missing information in studies of infection incidence in AAV make interpretation difficult. For example, many studies are unclear what proportion of subjects with treated with antimicrobial prophylaxis against PCP. Given that PCP prophylaxis is now largely considered a standard of care, the risk of severe infection is unclear for patients treated with modern regimens. Achieving adequate sample size is challenging in rare disease research. Many studies of infection incidence in AAV are small, frequently with sample sizes of 100 individuals or fewer. This makes it difficult to obtain accurate measures of the frequency of complications. There is a clear need in the AAV literature for studies of severe infection incidence with adequate sample size, variables to provide full context and high quality, complete underlying data to inform the epidemiological question.

1.13.3.3 Prognostic factors

Age has been identified as a risk factor for infections in AAV across multiple studies. The Beijing study by Lai identified an additional decade of age as an independent predictor of infection over one year with hazard ratio 1.3 (Lai et al., 2014). A similar effect was reported in an analysis of French RCTs which enrolled 733 patients (Lafarge et al., 2020). McGregor found an association between age and any infection with odds ratio 1.01, however when severe infections were analysed in a multivariable model, age was not statistically significant (McGregor et al., 2015). In a rituximab-treated cohort from two tertiary centres, one from the UK and one from Austria, Kronbichler identified age in years as predictive of severe infection with hazard ratio 1.03 (Kronbichler et al., 2018).

Female sex was identified by McGregor in a multivariable model to be associated with both any infection and severe infection, with odds ratios 1.75 and 1.83 respectively (McGregor et al., 2015). Women were far more likely to experience urinary tract infection but were also more likely to have upper respiratory tract infection. Other infections were similarly distributed according to sex (McGregor et al., 2015). Conversely, Watanabe-Imai reported female sex as a protective prognostic factor in relation to serious infection, with hazard ratio 0.47. Interaction with smoking history, which was strongly associated with sex in this study, may account for this finding (Watanabe-Imai et al., 2017). The French analysis of RCT data did not find an association between severe infection and sex using a competing risk model (Lafarge et al., 2020).

Exposure to cigarette smoking has been identified as a prognostic factor for severe infection. The RemIT-JAV study found a hazard ratio of 2.6, though this may have been confounded by sex (Watanabe-Imai et al., 2017). A retrospective cohort from China of 248 AAV patients reported a hazard ratio of 2.3 for smoking in relation to severe infection (Yang et al., 2018).

Glucocorticoid exposure was recognised early as a prognostic factor associated with subsequent severe infection in the treatment of patients with AAV. In the 1970s the NIH group identified that patients receiving daily prednisone had a higher incidence of infections compared to those taking prednisone on alternate

days or those treated with cyclophosphamide alone (Hoffman et al., 1992). The relationship between high glucocorticoid exposure was further evidenced in an early French RCT comparing oral to intravenous cyclophosphamide. Glucocorticoid weaning was slower in this trial compared to alternative regimens, with patients receiving an average of 55 mg daily prednisone at three months. Rate of infections were high at 55% across the whole trial population. The oral cyclophosphamide group demonstrated a 70% infection incidence, while 41% of the intravenous group experienced this adverse event. Notably this study recruited some patients from the critical care setting, a population at particular high risk of infection (Guillevin et al., 1997). In the US report from McGregor, from 6 months after diagnosis, a significantly higher incidence of infections was found among those treated with glucocorticoids beyond 6 months, at 0.42 infections per person-year, compared to those who were not receiving glucocorticoids, at 0.23 per person-year ($P<0.0001$) (McGregor et al., 2012). Glucocorticoids were identified in the Japanese cohort as associated with serious infection. Their multivariable models contained initial prednisolone dose ≥0.8 mg/kg/day as a prognostic factor, with approximate hazard ratio of 3. This dose threshold equates to above 56 mg per day for a 70 kg person. Not all studies found a clear association with glucocorticoids. Lei reported that total glucocorticoid exposure did not predict severe infection, but confidence intervals for this finding were not available (Lai et al., 2014). No studies were available which examined the impact of different glucocorticoid dose thresholds in AAV.

Other treatments may impact the rates of infections. Rates of severe infections were not notably different in RCTs comparing rituximab and cyclophosphamide (Stone et al., 2010; Jones et al., 2010). In the systematic review by Thery-Casari, a meta-regression indicated that severe infection was associated with the cumulative dose of rituximab (Thery-Casari et al., 2020). Both RCT and observational data indicate that there is not a significant difference between oral or intravenous cyclophosphamide in terms of severe infection incidence (Pearce, McGrath, et al., 2018; Harper et al., 2012). Kronbichler identified TMP/SMX antimicrobial prophylaxis as strongly predictive of decreased rates of severe infection with hazard ratio 0.3 in a multivariable model (Kronbichler et al., 2018).

Comorbidities are important prognostic factors for infection in both the general population and AAV. McGregor identified steroid-induced diabetes as a strong predictor of severe infection with odds ratio 1.91 (McGregor et al., 2012). Kronbichler identified chronic obstructive pulmonary disease (COPD) as predictive with hazard ratio 6.3 in a multivariable analysis. In the same study, diabetes was predictive of severe infection on univariable analysis, but was not significant in the multivariable model. Cardiac involvement, represented by prior myocardial infarction or reduced left ventricular systolic function, was predictive of severe infection on univariable analysis, but was not significant in the multivariable model (Kronbichler et al., 2018).

Worse renal function has shown to be a predictor of severe infection across most studies. Lai and colleagues found that an increased creatinine clearance of 10 mL/min at baseline carried hazard ratio 0.93 (Lai et al., 2014). Lafarge reported numerically increased rates of infection with lower eGFR, though confidence intervals spanned both protective and deleterious effects (Lafarge et al., 2020). Kronbichler found higher eGFR to predict lower rates of severe infection in a rituximab-treated cohort, but this was not significant in a multivariable model (Kronbichler et al., 2018). A multivariable model in AAV-GN suggested that lower estimated glomerular filtration rate (eGFR) was statistically significantly associated with fewer severe infection events, with odds ratio 0.99. The thresholds of eGFR incorporated into the model were unclear, therefore this data is difficult to interpret (McGregor et al., 2015).

Other organ system involvement may predict infection. Lai found that lung involvement predicted infection within one year, with hazard ratio 2.3 (Lai et al., 2014). Lafarge's analysis of French RCT data revealed a hazard ratio of 1.81 for pulmonary involvement predicting severe infection (Lafarge et al., 2020). Similar results were found in the Korean study by Yoo, with hazard ratio 2.4 (Yoo et al., 2018). Kronbichler identified endobronchial disease activity as predictive of severe infection in the rituximab-treated tertiary centre cohort (Kronbichler et al., 2018). Yoo reported BVAS as associated with hospitalised infection events (Yoo et al., 2018). Increased organ system involvement implies more extensive and severe disease: worse immune dysfunction at baseline may contribute to

increased infection, but the relationship may also be confounded by subsequent high intensity of immunosuppression.

Certain laboratory or immune parameters predict infection. Lei reported that an increase in peripheral lymphocytes at diagnosis of 1 x 10^9/L was protective with hazard ratio 0.7 for severe infection (Lai et al., 2014). Leukopenia and neutropenia were associated with increased rates of severe infection in the Kronbichler study, though this was not significant in the multivariable model (Kronbichler et al., 2018). A small case-control study from France previously reported that lymphopenia prior to and during therapy was more prevalent in GPA cases who developed PCP, compared to GPA cases who did not (Godeau et al., 1995). ANCA subtype was not predictive of infection in the French RCT data (Lafarge et al., 2020). A study of vaccine response in 91 AAV patients reported that hypogammaglobulinaemia, reduced B cell count and reduced CD4-positive lymphocyte count predicted infection (Morgan et al., 2016).

Several studies have sought to identify prognostic factors for severe infection in AAV, but these are hampered by similar reasons to those described in relation to incidence studies, detailed in section 1.13.3.2 above: retrospective design, small sample size and missing important variables. The impact of these deficiencies becomes worse in the setting of prognostic factor and prognosis modelling. Studies with retrospective design are more vulnerable to selection bias, as inclusion criteria may be associated with the prognostic factor under investigation (Geneletti et al., 2009). This may lead to a biased estimate of the strength of association between prognostic factor and outcome. Sample size is highly important in studies that build statistical models using multiple variables. Such models are used in both prognosis factor and prognosis modelling research. An insufficient sample size can result in overfitting, with inaccurate estimates of prognostic factor model coefficients. The modelling conducted in studies of infection in AAV patients is highly vulnerable to this phenomenon – many studies have inadequate sample size for the type of modelling undertaken and none use recommended sample size calculations.

A further problem in the AAV prognosis literature is poor differentiation between the type of research undertaken: whether the scientific question relates to aetiology or prediction research (see section 1.14 for a more detailed discussion

of this issue in epidemiology). It is an almost universal weakness of the literature that studies in AAV where multivariable modelling is undertaken do not specify whether the goal is aetiological or predictive research. This is important because different modelling strategies should be used depending on the goal, specifically different variables should be included depending on the goal. For an aetiological study consideration of whether candidate variables are confounders, colliders or mediators is crucial. All confounders should be included in the modelling process, but colliders or mediators must be excluded. Prediction, or prognostic modelling, studies may consider all variables. As this thesis focuses on prognosis research, a detailed discussion of variable selection for aetiological studies is out with the scope of the thesis. Consideration of such issues can be found elsewhere such as a review by Heinze and colleagues (Heinze et al., 2018). The impact can be substantial however, as the model effect size of a given variable may be very different depending on the approach taken. Notably, no prognosis modelling studies relating to infection in AAV were found following the literature search.

Glucocorticoids are well recognised as contributing to severe infection, but studies of this exposure as a prognostic factor are limited for many of the reasons described above. In adjacent IMIDs, such as RA, well-conducted studies of the impact of different dose thresholds have been conducted (Dixon et al., 2011). Such studies have not been conducted in the AAV setting.

1.13.3.4 Outcomes

The early German study by Reinhold-Keller reported 22 (14%) deaths over median 7 years follow up in 155 patients. Five deaths (23% of all deaths) were due to infection, though active disease was also present. Overall, 3% of patients died due to infection. One death was associated with pneumonia without an identified pathogen and four were associated with *Staphylococcus aureus* bacteraemia. The NIH group reported a similar number of infection related deaths, despite a greater overall number of severe infections. As described above this variation may be related to glucocorticoid exposure. Further early experience was reported in a multicentre British case series of 265 patients diagnosed with GPA between 1975 and 1985. Although fewer patients were treated with cyclophosphamide compared to other similar reports at the time,

12% of the cohort died of infection (Anderson et al., 1992). A longitudinal analysis of 77 patients who were enrolled for the ACR vasculitis classification study showed that 10% of patients died of infection over mean 7.1 years follow-up (Matteson et al., 1996). In the French RCT conducted by Guillevin and colleagues, infection-related mortality was 18% over a mean follow-up period of approximately 28 months. This was significantly higher than infection mortality rates reported in other studies. High levels of glucocorticoid exposure may have contributed, though may also have been related to the high risk nature of the cohort (Guillevin et al., 1997).

McGregor described causes of death in the larger US registry report of individuals with AAV and renal involvement. Of 421 patients with 12 months complete follow-up, 31 (7%) died. The study also identified an association between the number of infections experienced in the first year of treatment and death. Infection resulted in at least 13% of deaths and 29% died with active vasculitis. Cardiovascular disease accounted for 10% of deaths, with the remainder described as causes after reaching ESKD or unknown causes. For individuals who did not experience an infection mortality was 3%, 1-2 infections were associated with 10% mortality rate, while those who suffered 3 or more infections had 13% mortality in the first year. Severe infection was associated with 19% mortality in the first year, compared to 4% in those who did not experience severe infection. A multivariable analysis revealed a strong association for severe infection with death, with hazard ratio 4.2 (McGregor et al., 2015).

The Chinese study from Lai and colleagues also reported that severe infection was a strong predictor of mortality within the first year, with adjusted hazard ratio 4.0. It found that the most frequent cause of early mortality was infection at 47%, followed by active vasculitis (22%), then cardiovascular disease (12%). After the first year of therapy, the most common cause of death was cardiovascular disease (29%), closely followed by infection (27%) (Lai et al., 2014). The REVAS study described a similar association between infections and mortality with odds ratio 3.7 (Solans-Laque et al., 2017). In the RemIT-JAV study, there were five deaths due to infection over the six month observation period following induction, 3.2% of the cohort (Watanabe-Imai et al., 2017).

Flossmann and colleagues performed a follow-up survey of patients recruited to four early EUVAS RCTs. Infection was the most common cause of death in the first year following enrolment at 48%. After the first year, infection remained an important contributor to mortality, responsible for 20% of deaths, with more deaths being caused by cardiovascular disease and malignancy at 26% and 22% respectively (Flossmann et al., 2011). The same data was evaluated by Little and colleagues, aiming to determine the contribution of adverse events to early mortality. The analysis demonstrated that infection was a strong independent predictor of mortality within 12 months following diagnosis (Little et al., 2010). As described, many studies report infection as the most common cause early mortality following AAV diagnosis, with some describing it as causing the majority of deaths. In many reports with longer follow-up, infection is overtaken as the most common cause of death by other causes, such as cardiovascular events. It is important to consider standardised mortality rates (SMR) when evaluating the cause of death, as excess deaths within a population deserve greater attention and are likely to be more amenable to interventions. Wallace and colleagues performed such an analysis in a large, contemporary AAV cohort of 484 patients, over mean follow-up of 7 years. Cardiovascular disease was responsible for the most deaths, but infection was identified as causing the most excess deaths by a substantial margin. The SMRs for infection, renal disease, cancer and cardiovascular disease were 13.9, 4.3, 2.7 and 2.3 respectively (Wallace et al., 2020).

1.13.3.5 AAV and Covid-19

At the outset of the Covid-19 pandemic, there was widespread concern among patients, clinicians, researchers and governments about the potential impact of the novel infectious disease on immunosuppressive individuals. Due to their vulnerability to severe infections, individuals with AAV were a specific population of concern. At the time of initial literature searches which informed this thesis, there was no published data on the incidence, prognostic factors or outcomes relating to Covid-19 in individuals with AAV.

1.13.3.6 AAV and infection: summary

Studies relating to severe infection in AAV have accumulated in recent years, providing increasing awareness of the frequency of this complication, prognostic factors and impact. The cumulative incidence over all follow-up time in studies described above ranges from 26 to 59%, with median 40%. In the first year 15 to 22% will experience a severe infection. The early incidence rate varies from 22 to 88 severe infections per 100 person-years, while the rate over more prolonged follow-up times ranges from 7 to 12 severe infections per 100 person-years. Over variable follow-up periods, infection as a proportion of the cause of death ranges from 10 to 48%, with median 20%.

Important patient-related predictive factors include age, diabetes, pulmonary disease and smoking history. Disease related factors likely include renal function and the presence of lung involvement. Therapy related factors are glucocorticoid exposure and possibly the use of cyclophosphamide over rituximab, although increased rituximab exposure has also been shown to be associated with greater number of infections. TMP/SMX is likely to be protective against severe infection. Leukopenia and hypogammaglobulinaemia are biomarkers of severe infection susceptibly.

Much of the literature has methodological limitations. Studies are frequently small, leading to inaccurate estimates and potentially spurious results. Retrospective studies are commonplace and prone to missing and inaccurate data, as well as survival bias. RCTs do not report infections in a standardised manner (Kronbichler et al., 2015). Important areas of deficiency exist in the AAV infection literature, which are highlighted above. It is clear, however, that severe infections are common and can lead to devastating outcomes for AAV patients. Although causality is difficult to determine, there is acceptance in the medical literature that necessarily intense immunosuppression leads to infections, many of which are severe in nature and can lead to early mortality. A large body of observation data from a variety of settings, and with varying analytical approaches, support this position.

1.14 Prognosis research

In clinical epidemiology, studies typically have descriptive, aetiological or predictive objectives. While descriptive studies, which typically aim to quantify incidence or prevalence of diseases and their outcomes, are intuitively understood, there is often conflation between the objectives of aetiological and predictive studies. Aetiological studies aim to determine whether an exposure causes an outcome. A predictive study uses multiple factors to predict the presence of a diagnosis or future occurrence of an outcome. This is regardless of where the factors incorporated into the tool are causal (Ramspek et al., 2021). Well-conducted predictive studies, also referred to as prognostic modelling studies, use advanced statistical methods to develop models which include multiple predictive factors. Such studies fall under a wider spectrum of scientific endeavour known as prognosis research. The PROGRESS framework was developed to help clarify concepts, improve study design and enhance standards of reporting in this field. This thesis will aim to develop aspects of prognosis research relating to the prediction of severe infection in AAV, utilising the underlying structure of the PROGRESS framework, described below.

1.14.1 The PROGRESS framework

Prognosis is the aspect of clinical medicine whereby the likelihood of future clinical events is determined in individuals or populations with a specific disease or within a particular clinical scenario. Prognosis research aims to establish the relationship between baseline health status and subsequent clinical events. The ultimate aim is to be able to accurately predict future outcomes, using information known about individuals at baseline, in order to improve outcomes for patients (Hemingway et al., 2009).

While the impact of high-quality prognosis research is not in doubt, the field has been less prominent, and methodological standards less rigorous, than in other important clinical fields such as RCTs and genomics. The field has suffered from variable terminology and poorly defined concepts. Many studies are low quality due to being underpowered, inappropriate methodology, vulnerability to publication bias and failing to replicate previous findings. The Prognosis Research Strategy (PROGRESS) was developed to improve research standards in

prognosis research. Four main types of prognosis studies have been defined to assist this objective: overall prognosis research, prognostic factor research, prognostic model research and stratified medicine research (Hemingway et al., 2013).

Overall prognosis research explores the average frequency of clinical outcomes of a specific disease or at-risk group within a particular population, at a specific time and within a defined location or healthcare setting. This type of study aims to answer questions such as “In females over 65 years in Scotland with hypertension diagnosed in primary care, what proportion will experience a major adverse cardiovascular event over the following 10 years?”. This contributes to understanding the impact of disease and allows comparisons across locations and time (Hemingway et al., 2013).

Prognostic factor research investigates whether specific characteristics of individuals with a disease impacts the frequency of an outcome. For the same group described above, an example might be “In females with hypertension does variation in the aldosterone synthase gene predict increased cardiovascular events over 10 years?”. A wide array of characteristics can be considered, such as environmental exposures, genes, clinical features or treatments. Such studies often examine the added predictive ability of novel characteristics, such as laboratory biomarkers, to more traditional characteristics, such as age or clinical observations (Riley et al., 2013).

Prognostic model research comprises developing and validating statistical models aiming to predict outcomes in individual patients. Such models typically include multiple prognostic factors. Where appropriate datasets are available, the clinical impact of the model should be evaluated. The QRISK3 model uses 21 patient characteristics to predict cardiovascular events over 10 years (Hippisley-Cox et al., 2017). Many models are used in routine clinical practice to make individualised decisions with patients (Steyerberg et al., 2013).

Stratified medicine research, also described as “predictors of treatment effect research”, aims to determine whether sub-groups of patients with particular prognostic characteristics will or will not benefit from specific treatments. An example might be: for an individual with hypertension and a specific

combination of prognostic factors, Drug A will likely provide more clinical benefit and cause fewer adverse effects than Drug B (Hingorani et al., 2013).

1.15 Aims

Individuals with AAV are vulnerable to an array of adverse clinical outcomes, with severe infection being among the most concerning. However, there is a lack of clearly defined, high quality prognosis research in this setting. As detailed above, incidence studies are hampered by incomplete variables, small sample size and retrospective design. There are no studies examining specific glucocorticoid dose thresholds as prognostic factors. There are no prognosis modelling studies examining severe infection risk from the time of diagnosis or mortality risk following a severe infection. Studies relating to Covid-19 prognosis research in AAV were not available. This thesis will seek to explore the impact of severe infection in AAV and develop methods for prediction, utilising prognosis research themes and methodology as described in the PROGRESS framework. The aims are as follows:

- To determine the incidence of severe infection in individuals with AAV across European registries using novel web technology (overall prognosis research, Chapter 2)

- To explore the predictive ability of glucocorticoid exposure, including dose thresholds, in relation to severe infection in individuals with AAV (prognostic factor research, Chapter 3)

- To develop and internally validate a predictive model for severe infection events in individuals with AAV (prognostic model research, Chapter 4)

- To develop and internally validate a predictive model for early mortality following a severe infection in individuals with AAV (prognostic model research, Chapter 5)

- To identify prognostic factors for severe Covid-19 in individuals with AAV and confirmed Covid-19 (prognostic factor research, Chapter 6)

- To explore the immunological response to SARS-CoV-2 vaccination and factors which predict SARS-CoV-2 infection in individuals with AAV (overall prognosis research/prognostic factor research, Chapter 7)

1.16 Summary

This introductory chapter has described AAV from epidemiologic, biological and clinical perspectives. While outcomes for individuals with AAV have improved with modern methods of diagnosis and evidence-based application of immunosuppressive therapy, adverse events are frequent and have serious impact on quality of life and longevity. Severe infections are among the most concerning adverse events and are the most common cause of early mortality and overall excess mortality. Through the studies alluded to above, this thesis will explore: the incidence of infection in AAV, prognostic factors for severe infection and prognostic modelling relating to severe infection, including examination of a novel infectious disease, Covid-19.

2 Data quality and incidence of severe infection in AAV registries: a retrospective cohort study

2.1 Overview

Chapter 1 provided a detailed overview of ANCA-associated vasculitis (AAV). It established that severe infection occurs frequently in AAV and can be life threatening. Studies conducted in this area often have limited sample size, therefore the resulting estimations of severe infection incidence lack precision. This chapter will describe the FAIRVASC initiative, a project undertaken by a European consortium which seeks to federalise vasculitis patient registry data using novel web technology. First, a data quality analysis of registry data will be described. Then, severe infection incidence will be reported, following one of the first applications of the FAIRVASC infrastructure to interrogate registry data in this manner. Due to challenges with the availability and quality of severe infection data, only two of seven registries were suitable to use. Regardless, this work represents the largest known study of severe infection incidence in AAV.

2.2 Abstract

2.2.1 Background

Individuals with ANCA-Associated Vasculitis (AAV) are at increased risk of severe infections. Understanding the incidence of severe infection in AAV patients across different settings is desirable. The FAIRVASC project seeks to federalise seven AAV registries using semantic web technology. Prior to the data uplift required for federalisation, the FAIRVASC consortium sought to perform a data quality (DQ) analysis of the data contained within the registries. This chapter describes two studies: a DQ analysis of FAIRVASC registry data prior to semantic web uplift and an assessment of severe infection incidence in AAV patients using the prototype FAIRVASC architecture.

2.2.2 Methods

A quantitative DQ assessment of core data items was performed at each FAIRVASC pilot registry. Eight representative variables were analysed across four DQ domains: uniqueness, consistency, completeness and correctness. The formal DQ assessment did not include infection data, but infection DQ was assessed qualitatively. The underlying FAIRVASC technology was described. Registry data was mapped according the FAIRVASC ontology and uplifted to local triplestores. Via SPARQL queries, the FAIRVASC interface was used to retrieve severe infection incidence data over discrete time periods.

2.2.3 Results

There were 6,104 participants across seven registries. Uniqueness was 100% across all registries in terms of unique identification numbers. Duplicate patients existed below 1% for UKIVAS and Czech registries and at 3.6% for the POLVAS registry. Consistency of data type was 100% across all registries and variables. Consistency with respect to logic tests and plausibility tests was 95-100%. Completeness varied between 53-100%, but was greater than 95% for most variables in most registries. Correctness was greater than 95% for most variables in most registries. Due to specific issues with the presence and quality of infection data, only two registries were appropriate to use to determine severe infection incidence. There were 1,120 participants in these registries. The

combined incidence of severe infection was 179.2 events per thousand person-years (95% confidence interval 153.9 – 207.6) for the first year after diagnosis and was progressively lower over subsequent time periods.

2.2.4 Conclusions

A quantitative assessment of core data items determined that DQ in the FAIRVASC registries was of an appropriate standard for future FAIRVASC research studies and activities, however a qualitative assessment of severe infection data resulted in only two of seven registries being able to be used to evaluate severe infection incidence. Although fewer registries were able to be used than anticipated, application of the FAIRVASC architecture demonstrated that severe infections were a major complication, most frequently occurring in the first year after diagnosis.

2.3 Introduction

2.3.1 Severe infection incidence in AAV

Individuals with ANCA-associated Vasculitis (AAV) are at increased risk of severe infection due to the requirement for potent immunosuppressive therapy to control disease activity. Disease related factors, such as disruption of the innate immune defence with respect to the respiratory tract, also contribute to infection susceptibility. As summarised in Chapter 1, a variety of studies have shown that AAV patients are highly susceptible to infections, including registry based cohort studies, population-based cohort studies and case-control studies.(McGregor et al., 2015; S. H. Sarica et al., 2020; Rathmann et al., 2021) Infection is the leading cause of early mortality.(Little et al., 2010) Studies of the incidence of infection from different regions with comparative methodology are lacking. Quantifying the risk of severe infections across different geographical and healthcare settings is key to a deeper understanding of the impact of this disease complication and how it might best be tackled, both from a basic and clinical research perspective, and a health care delivery perspective. Making such data available to vasculitis researchers is a principal goal of the FAIRVASC project. FAIRVASC is a large European consortium aiming to combine data from multiple AAV registries. Key to understanding the project are the current European data protection landscape and the underlying technology, the semantic web.

2.3.2 Data protection landscape

In 2016 the European Parliament and Council legislated on the use of personal data through the General Data Protection Regulation (GDPR) (REGULATION (EU) 2016). This aimed to strike a balance between two key societal values: protecting personal data whilst preserving the ability for individuals and organisations to be able to legitimately use personal data for the benefit of citizens. Prior to the introduction of GDPR, the basis on which scientists and clinicians were able to use patient data was contingent on either the consent of participants or anonymisation of the data in question. For data to be fully anonymised various important attributes of the data often must be removed, potentially rendering it less useful or, in some cases, no longer fit for the

purpose of addressing the original scientific question. Anonymisation is particularly challenging in rare disease research where subjects are at increased risk of being identifiable due to the rarity of their underlying condition. In the case of scientific research, GDPR allows for limited exemption from some obligations, such as the necessity for the subject's consent for data processing, where there is deemed to be sufficient public good achievable from the research in question. However, these exemptions are determined at a Member State level, leading to variability across national borders in terms of what data sharing or data processing is permitted. This leads to significant challenges for cross-border health research projects. Complicated multi-party data sharing agreements are often required, with input from the legal teams of all participating institutions. For many projects, funding and resources are not available to overcome this challenge. FAIRVASC seeks to utilise semantic web technology to analyse subject level data in a manner that is fully compliant with GDPR without that data leaving the host institution. The aim is for rare disease research to be able to be performed across national boundaries, simultaneously removing the need for complicated legal agreements and comprehensively protecting personal data in a secure manner.(Donnelly and McDonagh, 2019)

2.3.3 The Semantic Web

The semantic web, also known as the web of data, is a movement to have, theoretically all, online data being uniquely identifiable, clearly described and have defined relationships with other data. The concept was first popularised in the early 2000s. Each data item should be uniquely identified with a uniform resource identifier (URI), much in the way all webpages are uniquely identified with a web address, the uniform resource locator (URL). Data, and the relationships between data items, should be clearly described using ontologies. An ontology is a formal description of concepts within a specific domain and the relationship of the concepts to other concepts. Well know examples outside of the biomedical domain include FOAF (Friend of a friend), an ontology for describing relationships between people, and the DBpedia ontology, a cross-domain ontology which originated from data on Wikipedia.(FOAF collaboration, 2022; DBpedia collaboration, 2022) Important ontologies in biomedicine include SNOMED CT, a large collection of medical terms and definitions, and the Gene Ontology, a compilation of genes and gene products across all species.(SNOMED

International, 2022; Gene Ontology Consortium, 2022) Data in the sematic web is stored in a semantic graph comprised of nodes and edges. In this graph structure, data points are represented by the nodes and relationships between data points are represented by the edges. To give an example of data represented in this manner, consider two data points: a patient's identification number (ID) and a doctor's ID, where the relationship between the data points is that the patient is under the care of that doctor. In semantic graph structured data these two data points ("Patient ID" and "Doctor ID") can be conceptualised as nodes, while the edge is the relationship between the data points ("is under the care of"). This simple example can be expanded upon infinitely and flexibly – one of the key strengths of the semantic web. This is represented graphically in Figure 2-1. This contrasts with traditional means of storing data in relational databases, which are comprised of separate tables and keys in each table relating them. Notably, relationships between data points are not described in the latter. The graph structure has several strengths including being highly flexible, infinitely extensible, machine readable and suited to data structures with complicated relationships between variables. Ascribing these properties to data essentially makes it machine readable, meaning that computers can analyse the relationships between different data items. Using computation, scientists can take advantage of this semantically structured data to make new discoveries. As an example, linking a genetics database to a protein database using semantic technology, allowed new gene-protein interactions to be discovered, thus providing potential novel therapeutic targets. Semantic web technology has seen widespread adoption and the amount of semantically described online data has increased dramatically over the past two decades. Major administrations, such as American and UK governments, publish a large proportion of public data as semantic data and all major technology companies, including IBM, Alphabet and Meta utilise semantic web technology.

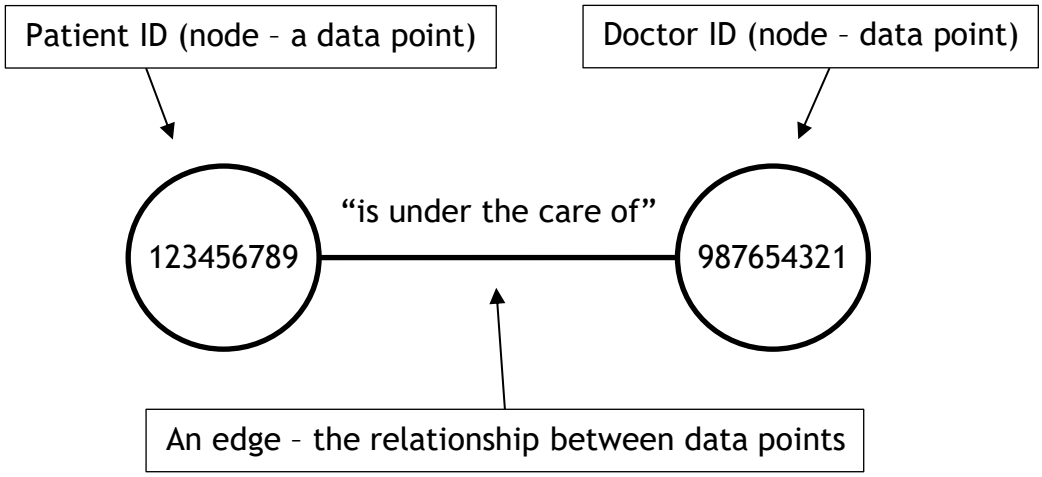


Figure 2-1 | Example of nodes and edges in a simple semantic graph

2.3.4 FAIRVASC

The concept of FAIR, developed by the GO-FAIR initiative, aims to make scientific data Findable, Accessible, Interoperable and Reusable.(Wilkinson et al., 2016) FAIRVASC is a European Joint Program on Rare Diseases (EJPRD) funded study, with additional support from industry, which seeks to make vasculitis data FAIR in order to answer new research questions. It does so by combining multiple AAV registries using semantic web technology. AAV represents a rare set of diseases with the incidence for AAV as a whole being reported as 1.4 – 38.2 cases per million person-years.(Kitching et al., 2020) Epidemiological research relies on data with sufficient sample size to permit reliable statistical inference. Achieving such a sample size in the rare disease setting - such as AAV research – is not always possible, particularly within a single nation or region. This is one of the principal challenges that FAIRVASC seeks to address. Various solutions have been proposed, traditionally a form of multinational registry. Traditional multinational registries have two main designs: central and network. In the central registry model, there is a single research protocol and data is submitted to a central database administered by a coordinating centre. Participating sites collect local patient data using a single case report form (CRF), possibly with some minor variability such as language or adaptation for local services. There are several registries which align with this model, covering a diverse set of both rare and common clinical conditions, such

as Gaucher disease and acute coronary syndrome.(Khan et al., 2012; Zubaid et al., 2014) The network model takes a different approach. This model seeks to align separate registries, each of which has its own protocol, CRF and database. There is often a common data model whereby registries collect specific shared variables. These variables can then be submitted to a central platform and combined. Various groups subscribe to this model with clinical areas including psoriasis and medical devices.(Lecluse et al., 2009; Sedrakyan et al., 2014) These models have strengths and limitations. Strengths include a consistent data structure for the central model and ease of analysis for both, as the data ultimately exists within one database for analysis. Limitations include cost, other resource consumption, lack of efficiency, requirement for complicated ethical approvals and privacy concerns. Setting up a central registry can take resource away from existing local registries. In the rare disease context, there are often many existing heterogeneous registries and heightened concerns regarding data privacy due to the increased identifiability of rare disease patients. Taking advantage of recent technological developments, the federated model seeks to address many of these challenges. In FAIRVASC, the federated approach utilises semantic web technology to make aggregated data available without the data ever leaving the server of the registry's host institution. As a result, the process complies with both the spirit and the letter of European data protection legislation, without necessarily requiring complicated data sharing agreements. The semantic approach requires comprehensive data harmonisation which results in carefully aligned data before federation takes place. The model for aligning data is described in the FAIRVASC ontology (see methods).

2.3.5 Data quality

There is variability in the literature as to what constitutes data quality (DQ). Defining DQ, and its constituent domains, remains an active area of research. One common definition, however, is that DQ is characterised by the extent to which data is fit for purpose.(Fadahunsi et al., 2019) Overall DQ is typically represented by different DQ domains.(Batini et al., 2009) Notably, there is also significant variability in the nomenclature of DQ domains in the literature.(Weiskopf and Weng, 2013) Some domains are quantitative and can be used to evaluate variables contained within a database, producing DQ statistics. Examples of such domains are accuracy (also described as correctness),

consistency, completeness and timeliness. Qualitative DQ domains include governance, availability, trustworthiness and relevance. It is not known what level of DQ is required for reliable scientific research. It is likely that the degree of necessary DQ will vary depending on the subject area, methodology and purpose of the study. Some domains will also be of greater or lesser importance depending on the setting. However, it is generally accepted that organisations should assess DQ and where necessary take steps to improve it, in order to improve the reliability of scientific results based on that data.

2.3.6 Aims

The first aim of this chapter is to describe a pilot study of various data quality metrics for the FAIRVASC pilot registries and to report summary measures across all registries as an indication of the suitability of the data for the stated aims of FAIRVASC. The second aim is to utilise the FAIRVASC infrastructure to extract and report severe infection incidence from multiple registries. This represents one of the initial components of prognosis research, whereby it is key to establish the incidence of an adverse clinical outcome, before going on to determine prognostic factors which are associated with the outcome or to derive multivariable models to predict the outcome.

2.4 Methods

2.4.1 Methods overview

First the FAIRVASC project methodology will be briefly summarised. The methodology for the DQ study and the severe infection incidence study will then be described. Reporting follows guidance according to the Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) statement.(von Elm et al., 2007)

2.4.2 FAIRVASC

A framework to deliver the core infrastructure was developed iteratively with input from clinicians, researchers, computer scientists, health data experts and, crucially, patients and patient representatives. This framework is depicted graphically in Figure 2-2. A detailed description of the FAIRVASC approach has

been published.(McGlinn et al., 2022) Harmonisation refers to the process of identifying similar or identical terminology used across systems to describe phenomena. In the case of rare disease registry integration this involved identifying variables across registries that either represent the same information but may be labelled differently or identifying variables which could be transformed, sometimes utilising other data in the registry, to represent the clinical concept in question. Data dictionaries from each pilot registry were collated and compared. Potentially common variables were identified across registries in several categories including demographics, vasculitis diagnosis, comorbidities, investigations, treatment and complications, such as cardiovascular disease, malignancy and infection, and outcomes such as mortality. The output from assessment of data dictionaries then informed initial discussions of the Query Implementation Team (QIT), a group of vasculitis clinician scientists with representation from across the pilot registries. The focus of QIT was to determine research questions which would be of scientific importance to eventual users of the FAIRVASC infrastructure, many of whom are likely to be clinician scientists. The summary output is requested from the registries through the FAIRVASC web interface through a point-and-click web application that is easily used without the need for computer science expertise. This request is transformed into a SPARQL Protocol and RDF Query Language (SPARQL) query. SPARQL is a query language designed to retrieve data from a semantic data graph, specifically one in the Resource Description Framework (RDF) format. RDF is a World Wide Web Consortium (W3C) standard for describing semantic web data. The Harmonisation Team is predominantly made up of registry representatives with close working knowledge of registry metadata, structure and use. Once potential queries were developed by QIT, the Harmonisation Team (HIT) then established how each registry would derive the necessary variables. This informed the development of the FAIRVASC ontology, published online at http://ontologies.adaptcentre.ie/fairvasc/. This web page includes an interactive visualisation of the FAIRVASC ontology. Screenshots of the ontology visualisation and a small section of the structure of the ontology are available in the appendix. The FAIRVASC Implementation Team (FIT) is made up of a representative from each registry with an information technology background and trained by semantic web experts within the consortium on semantic web technologies. Based on output from HIT, FIT transformed registry

data to RDF format using the Relational to Resource Description Framework Mapping Language (R2RML). RDF data was then uplifted to a local ‘triplestore’, a database designed for the storage and retrieval of semantic data. Registry triplestores were then able to be queried remotely using SPARQL via the FAIRVASC interface. An iterative approach to ontology design and implementation has been used, with continuous input from QIT, HIT and FIT. A ‘core team’ of three individuals, one computer scientist and two clinical researchers (including the thesis author), has coordinated the interaction between these groups. Ultimately a protype interface was developed. This enables the user to determine counts, percentages and confidence intervals (CI) for a range of outcomes and can be stratified by various baseline characteristics, including by registry.

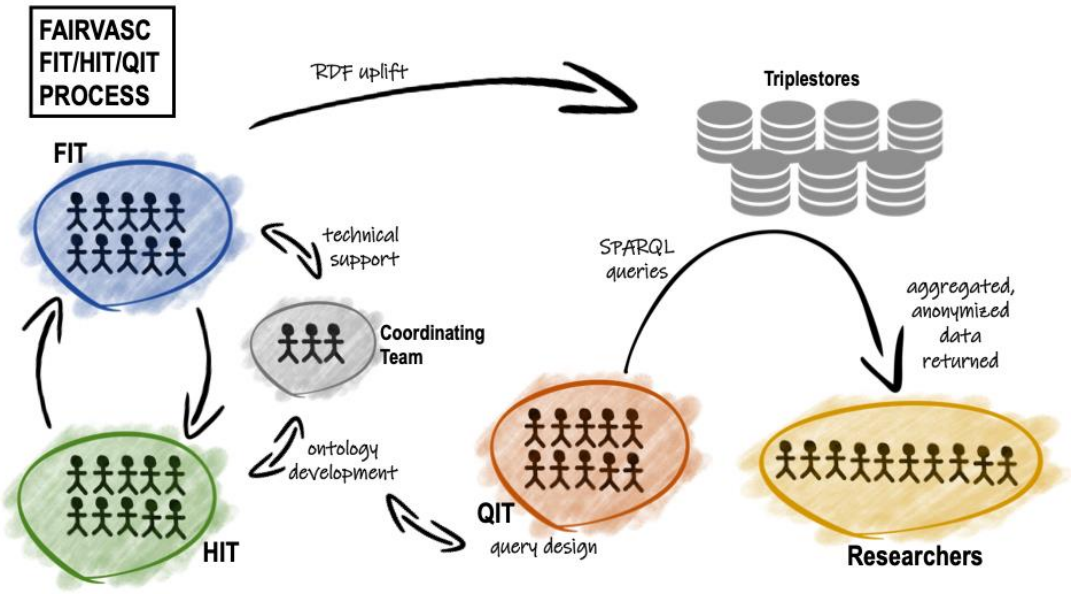


Figure 2-2 | Framework underpinning the development of the FAIRVASC infrastructure FIT = FAIRVASC implementation team, HIT = Harmonisation implementation team, QIT = Query implementation team, RDF = resource description framework, SPARQL = SPARQL protocol and RDF query language. These terms are described above.

2.4.3 Study design and setting

The DQ study is a cross-sectional pilot analysis of the seven pilot FAIRVASC registries, which are either registries dedicated to AAV or broader registries covering other vasculitides, but with substantial numbers of AAV participants. The DQ study was a global and quantitative assessment of registry data and was not specific to infection, therefore variables relating to severe infection were not included. The severe infection incidence study is a registry-based, mixed retrospective-prospective cohort study. With respect to severe infection, a qualitative DQ assessment was undertaken by surveying registry owners to ascertain the presence, consistency, completeness and correctness of relevant data. The study design, setting, location, period of recruitment, follow-up, data collection and summary of qualitative DQ relating to severe infection for each registry is described in Table 2-1.

2.4.4 Participants

Eligible participants for the DQ study were all AAV patients within the seven FAIRVASC pilot registries as defined by recognised international standards such

as the American College of Rheumatology 1990 criteria for the classification of Wegener's granulomatosis (ACR 1990), the 2012 revised International Chapel Hill Consensus Conference Nomenclature of Vasculitides (CHCC 2012) or the European Medicines Agency (EMA) classification algorithm (Leavitt et al., 1990; Jennette et al., 2013; Watts et al., 2007). Data for the incidence of severe infection study were from AAV patients registered with FAIRVASC registries where severe infection data was available (Table 2-1). Severe infection was defined as an infection associated with an admission to hospital. Sources and methods of selection of participants and methods of follow-up for each registry are described in Table 2-1.

2.4.5 Variables

Eight variables were assessed as part of the DQ study: sex, date of birth, serum ANCA autoantibody, a single comorbidity (chosen by the local registry team, diabetes was recommended), Birmingham Vasculitis Activity Score (BVAS), serum creatinine at baseline, date of death (if death had occurred) and date of End Stage Kidney Disease (EKSD; if ESKD had occurred). Serum ANCA autoantibodies and serum creatinine assays varied across, and within, registry sites. Presence of comorbidity was defined by the local investigator's clinical judgement. BVAS was version 3.(Mukhtyar et al., 2009) ESKD was defined as requirement for dialysis or kidney transplantation.

Baseline variables for the severe infection incidence study were sex, AAV diagnosis, ANCA autoantibody status and mortality status. Ideally age would have been included, but at the time of retrieving analyses via the FAIRVASC interface this variable had not yet been incorporated into the available queries. The outcome variable was incidence rate of severe infection as defined by the number of severe infections per thousand person-years.

2.4.6 Data sources / measurement

One registry (FVSG; French) was predominantly derived from national clinical trials. The Skåne registry (Sweden) identifies possible AAV patients through health district administrative codes, then utilises the local clinical record to confirm this diagnosis before inclusion and use of the clinical record to populate

the registry. All other registries recruited patients with AAV prospectively and data was sourced from the local clinical record or through direct patient interaction as part of registry data collection. Where some patients are recruited after their vasculitis diagnosis, some retrospective data collection is carried out (Czech AAV registry, GEVAS, POLVAS). All seven registries were analysed for the data quality study. Due to infection DQ issues, only two registries were appropriate to analyse for the severe infection incidence study, discussed further below.

DQ domains were selected by the FAIRVASC Data Quality Group (DQG) which has a broad range of clinical, statistical, health informatics and computer science expertise, in addition to representation from all pilot registries. These domains were prioritised by investigator consensus from a pool of nine candidate domains drawn from the literature and developed through prior published research.(Aerts et al., 2021) The identified domains were uniqueness, completeness, consistency and correctness. Definitions for DQ domains can vary across subject areas. For this study DQ domains were defined as follows. Uniqueness was defined as the degree of unwanted duplication within a variable. It was reported as the proportion of non-duplicated values for a variable with respect to the total number of values. Completeness represents the extent to which missing data has been minimised and was defined as the proportion of all subjects with non-missing data for a given variable. Consistency is the extent to which data is in a format which aligns with the registry's data dictionary. Correctness, synonymous with accuracy, is the degree to which data represents the true, real-world object or event.(Arts et al., 2002) Completeness, consistency and correctness were also reported as the proportion of values which aligned with the definition. A DQ work sheet was designed and disseminated to local registry DQ analysts. Uniqueness was assessed in two ways. First, the total number of unique patient identifiers (IDs) were analysed for duplicates. Then possible duplicate patients entered under different IDs were assessed. This was done by identifying patients with the same sex and date of birth (DOB). Local DQ analysts then further compared these individuals across additional variables such as approximate date of diagnosis and date of death to determine whether the case was indeed a duplicate. Consistency was examined with three approaches. First, all eight DQ variables were examined for appropriate data type or format, according to the

local data dictionary. Potential data types include character string, numeric, integer and dates. Then two logic tests were applied to three variables: was date of death “greater than” date of birth and was date of death “greater than” date of diagnosis. The last consistency checks were two plausibility tests: was BVAS at diagnosis within the possible range (0 – 63) and was serum creatinine at diagnosis within a biologically plausible range (0 – 5000 micromol/L). Completeness was checked across all variables by assessing the amount of missing data, this was reported as “percentage complete”. For date of death and date of ESKD, the denominator was the number of these events that had occurred. Where possible, correctness of values in each registry was assessed against the source data for at least 10 patients per registry. Where the variable did not exist in a given registry this was reported as “NA”. Accessing source data was not possible in the required time frame for the French registry, therefore all correctness data was reported as “NA”.

Severe infection data is not collected in all registries. It is not collected in UKIVAS and POLVAS collect whether any infection has taken place, but do not count individual infections. GEVAS at present likely has incomplete infection data. The Czech registry is not designed for complete capture of infection data. French registry infection data was not in a suitable format for transformation into RDF. Therefore, due to variation across registries in how severe infection data is collected and represented, we included severe infection data from registries where this was likely to be complete and there was sufficient granularity in terms of number and severity of infection. Therefore, the ideal included registries for the severe infection incidence analysis were RKD and Skåne.

2.4.7 Bias

Data quality study: Variables were intentionally selected to gain a representative sample of both those likely to have a high level of data quality (e.g. age) and those which may be suspected to have a significantly lower level of overall data quality due to complexity of entry (e.g. BVAS). To maximise external validity, variables which were considered likely to be useful in epidemiological studies were selected. For correctness assessment, participants were selected randomly to avoid selection bias.

Severe infection incidence study: As above, where significant missing data on severe infection occurrence was likely, such registries were excluded from the severe infection analysis. The data quality analysis informed registry inclusion. Survival bias, a form of selection bias, is a potential concern in any cohort study where the outcome can potentially affect study participation. Although the large majority of participant data was collected prospectively from diagnosis, some retrospectively entered participants were included. An exact proportion is not available. In this instance, survival bias would occur whereby a patient may not be recruited to a study due to having the event of interest – a severe infection, which may result in death, prior to the time of potential recruitment. This may result in systemic bias where the cohort experiences a lower incidence of severe infection compared to the ideal target population. Information bias may be an issue in some AAV cohorts. AAV cohorts are often recruited at tertiary centres. Some patients who experience a severe infection may be admitted to a local, secondary care centre. Data relating to such events may not be accessible to investigators from the tertiary site. These forms of bias were addressed in this study by only selecting registries that had a large proportion of prospectively recruited participants (RKD) or a population-derived cohort (Skåne), which have high levels of estimated complete outcome data.

2.4.8 Study size

The sample size was determined by the maximum number of cases available in registries with minimal missing data regarding incidence of infection and low loss to follow-up.

2.4.9 Quantitative variables

Due to the nature of the federated approach, it was not possible to gain access to patient level data. Ideally statistical adjustment would have been carried out using dichotomised variables such as age and sex using the Cochran-Mantel-Haenszel method. However, at the time of analysis age was not available as a variable for stratification in the FAIRVASC web interface, therefore adjustment was not possible.

2.4.10 Statistical methods

Baseline characteristics were reported as counts and percentages. DQ metrics were reported as counts and/or percentages. Severe infection was reported as incidence rates for each individual registry and the combined registries. CIs were calculated by approximation to a normal distribution.

2.5 Results

2.5.1 Description of the FAIRVASC registries

Table 2-1 shows a descriptive table of the FAIRVASC pilot registries. The total number of patients in each registry varied from 106 to 2644 individuals with AAV. Some registries were purely dedicated to AAV, while most collected data on all patients with systemic vasculitis. All registries utilised a recognised international standard for the definition of AAV, such as ACR 1990, CHCC 2012 or EMA.(Leavitt et al., 1990; Jennette et al., 2013; Watts et al., 2007) Other aspects of the registries, such as study design, setting, location, recruitment period, follow up, data collection and availability of severe infection data, are described. Severe infection data was not a component of the formal, quantitative DQ assessment, but a qualitative evaluation was carried out. Two registries (RKD and Skåne) contained severe infection data deemed complete, consistent (i.e. in a useable format) and correct by registry owners. Two registries (Czech and GEVAS) contained severe infection in their data dictionaries, but registry owners reported that such data was not routinely collected. This was reflected in implausibly low severe infection incidence. The French registry contained infection data, but in a “free text” format that was not possible to quantitatively analyse. POLVAS collected minimal severe infection data: a binary variable relating to the prevalence of severe infection data, but no count data relating to number of infections or associated timestamp. UKIVAS did not collect severe infection data. Therefore only two registries were included in the incidence study, described in section 2.5.3.

Table 2-1 | Descriptive table of registries

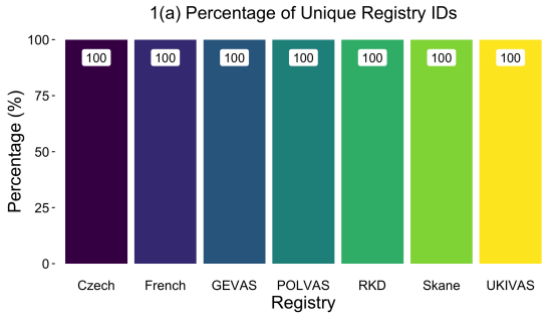
	Czech	GEVAS	French	POLVAS	RKD	Skåne	UKIVAS
n	268	106	2644	878	698	325	1185
Disease area	AAV, nephrology predominant	All systemic vasculitis	All systemic vasculitis, majority AAV	All systemic vasculitis	All systemic vasculitis, nephrology predominant	AAV	Systemic vasculitis including AAV
AAV definition	EMA	CHCC 2012 (all) ACR 1990 (GPA & EGPA) Clinical (Renal limited)	ACR 1990 and/or CHCC 2012	Combined CHCC 2012 and ACR 1990	EMA	EMA	CHCC 2012
Study design	Mixed retrospective / prospective	Mixed retrospective / prospective	Prospective cohort study	Retrospective (prospective component not in FAIRVASC)	Mixed retrospective / prospective	Mixed retrospective / prospective	Mixed retrospective / prospective
Setting	Secondary / tertiary care, 16 recruitment sites	Vasculitis centres	Secondary / tertiary care, 75 recruitment sites	Secondary care	Secondary / tertiary care, 8 recruitment sites	Population based	Secondary care
Location	Czech Republic - approximately 60-65 % of Czech AAV patients recruited	Germany (future: Austria, Switzerland)	France	10 centres covering 60% of population	Ireland - approximately 75% of Irish AAV patients recruited	Skåne, Sweden	UK and Ireland
Period of recruitment	2009 – present	June 2019 – present	1983 – present	March 2016 – present	September 2012 – present	1997 – 2019	
Follow-up	To present	Up to 2022	To present	Up to 2021	To present	Up to 2020	Plan for follow up with data linkage
Data collection	Encounter based, minimum annually	At clinical visit	At clinical visit (typically	Review of clinical record	Encounter based	Review of clinical record	Initial clinical visit / clinical record

			biannual)				
Loss to follow up	n/a – date of last encounter marks end of follow up	3%	n/a – date of last encounter marks end of follow up	Nil (for retrospective component)	n/a – date of last encounter marks end of follow up	Estimated 0.5% for those attending clinical services	n/a
Severe infection data available / DQ	Yes / substantially underreported	Yes / substantially underreported	Yes / inaccessible format	No / -	Yes / sufficient DQ	Yes / sufficient DQ	No / -

ACR = American College of Rheumatology criteria, CHCC = Chapel Hill Consensus Conference criteria, DQ = data quality, EMA = European Medicines Agency criteria for AAV, EGPA = eosinophilic granulomatosis with polyangiitis, GPA = granulomatosis with polyangiitis, n = number of AAV participants

2.5.2 Data quality

Issues with severe infection DQ are addressed in section 2.5.3. With respect to overall DQ as determined in the main DQ assessment, uniqueness was described both in terms of the number of unique IDs, but also the number of unique patients who in could be entered into the registry with more than one unique ID. Across all registries, participant IDs were 100% unique (Figure 2-3). The number of patients who had possible duplicate entries was zero or low across registries, however the POLVAS registry did have higher than expected duplicated patients at 3.6% (32/878). Consistency of data type was 100% across all variables. In some registries certain variables were not present and therefore were not able to be assessed for consistency, e.g. ANCA auto-antibodies in UKIVAS and comorbidity in Czech, POLVAS and UKIVAS (Figure 2-4). Consistency for logic tests of date of birth, date of diagnosis and date of death was between 95% to 100% and between 99.8% to 100% for plausibility test for BVAS and serum creatinine (Figure 2-5). Completeness was more variable across registries (Figure 2-6). Completeness for sex, date of birth and date of ESKD was 95% to 100% across all registries. Completeness for serum ANCA autoantibodies was similarly high, with the exception of the French registry at 52% (1382/2644). Due to the nature of registry data structures, comorbidity completeness could only be performed in the French registry. Many of the other registries collected data on comorbidities, but stated when these were present and not explicitly when absent. As a result, it was not possible to determine completeness in those registries for this variable. Completeness for BVAS scoring varied from 57% to 99% and for serum creatinine from 71% to 99%. Completeness for date of death was 94% to 100%, with the exception of 67% for GEVAS. Notably GEVAS is the most recently initiated registry, having started recruitment in 2019. At the time of analysis only three individuals were deceased, and date of death was missing for one individual.


1(a) Percentage of Unique Registry IDs
Percentage (%)
100
75
50
25
0
100
100
100
100
100
100
100
Czech
French
GEVAS
POLVAS
RKD
Skane
UKIVAS
Registry

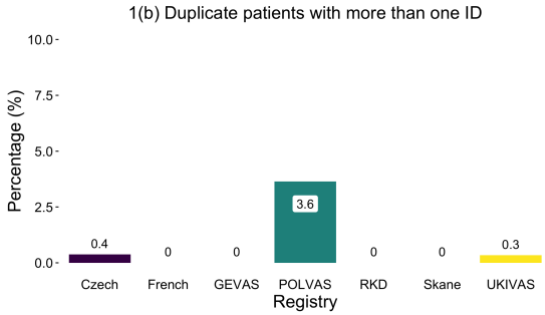

1(b) Duplicate patients with more than one ID
Percentage (%)
10.0
7.5
5.0
2.5
0.0
0.4
0
0
3.6
0
0
0.3
Czech
French
GEVAS
POLVAS
RKD
Skane
UKIVAS
Registry

Figure 2-3 | Data quality metrics by registry – Uniqueness

2(a) Consistency - Date of Birth

2(b) Consistency - ANCA

2(c) Consistency - Comorbidity

2(d) Consistency - BVAS

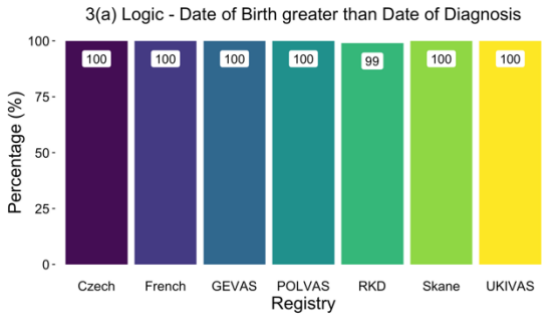
2(e) Consistency - Creatinine

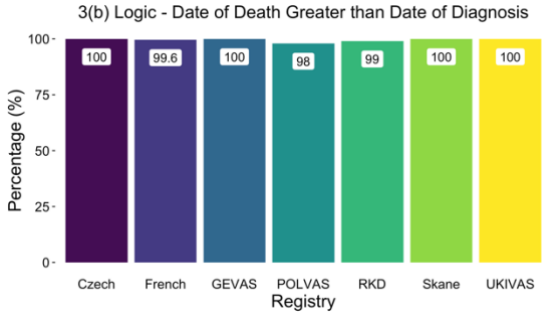
2(f) Consistency - Date of Death

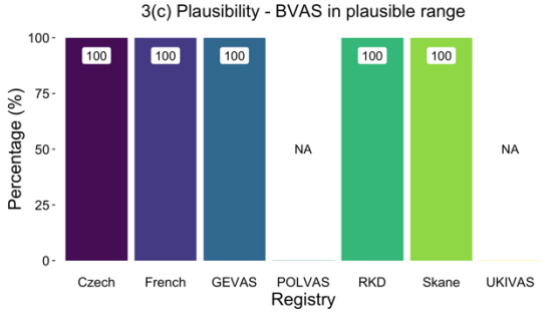
2(g) Consistency - Date of ESKD

Figure 2-4 | Data quality metrics by registry – Consistency: data dictionary format
BVAS = Birmingham vasculitis activity score, ESKD = end stage kidney disease. “NA” represents where a variable was not available for a registry.

2 95


3(a) Logic - Date of Birth greater than Date of Diagnosis
Percentage (%)
100
75
50
25
0
100
100
100
100
99
100
100
Czech
French
GEVAS
POLVAS
RKD
Skane
UKIVAS
Registry


3(b) Logic - Date of Death Greater than Date of Diagnosis
Percentage (%)
100
75
50
25
0
100
99.6
100
98
99
100
100
Czech
French
GEVAS
POLVAS
RKD
Skane
UKIVAS
Registry


3(c) Plausibility - BVAS in plausible range
Percentage (%)
100
75
50
25
0
100
100
100
NA
100
100
NA
Czech
French
GEVAS
POLVAS
RKD
Skane
UKIVAS
Registry

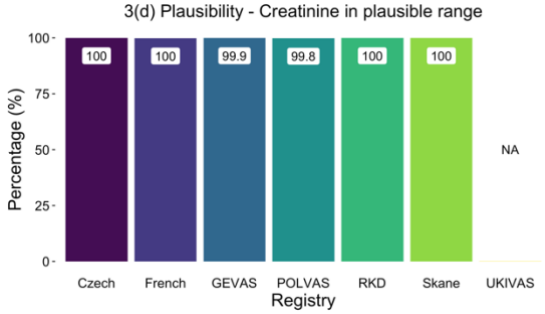
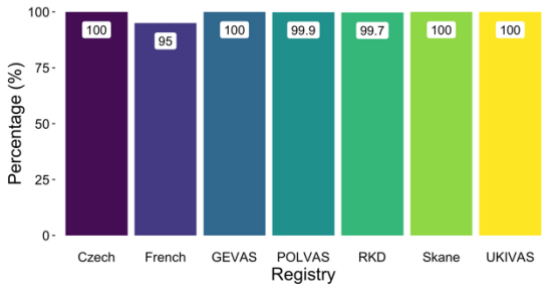

3(d) Plausibility - Creatinine in plausible range
Percentage (%)
100
75
50
25
0
100
100
99.9
99.8
100
100
NA
Czech
French
GEVAS
POLVAS
RKD
Skane
UKIVAS
Registry

Figure 2-5 | Data quality metrics by registry – Consistency: Logic and Plausibility Tests
ESKD = end stage kidney disease. “NA” represents where a variable was not available for a registry.

4(a) Completeness - Sex

4(b) Completeness - Date of Birth



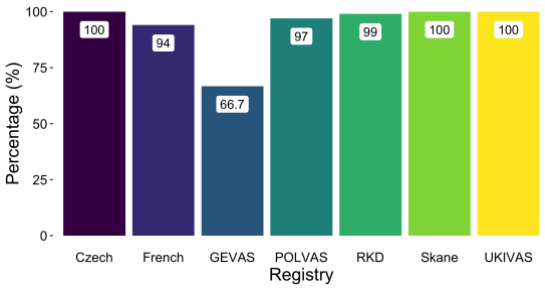
4(c) Completeness - ANCA

4(d) Completeness - Cormorbidity

4(e) Completeness - BVAS

4(f) Completeness - Creatinine

4(g) Completeness - Date of Death



4(h) Completeness - Date of ESKD

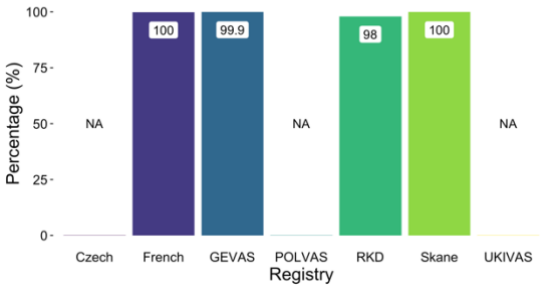


Figure 2-6 | Data quality metrics by registry – Completeness
BVAS = Birmingham vasculitis activity score, ESKD = end stage kidney disease. “NA” represents where a variable was not available for a registry.

5(a) Correctness - Sex

5(b) Correctness - Date of Birth

5(c) Correctness - ANCA

5(d) Correctness - Comorbidity

5(e) Correctness - BVAS

5(f) Correctness - Creatinine

5(g) Correctness - Date of Death

5(h) Correctness - Date of ESKD

Figure 2-7 | Data quality metrics by registry – Correctness
BVAS = Birmingham vasculitis activity score, ESKD = end stage kidney disease. “NA” represents where a variable was not available for a registry.

2.5.3 Severe infection: Cohort baseline characteristics and incidence

Baseline characteristics, mortality and follow-up of the severe infection cohort, both per-registry and combined, are summarised in Table 2-2. Missing data in the core registry variables is summarised in the data quality analysis under 'completeness'. At present data on completeness for infections is not available. Number of severe infections, total time-at-risk and incidence of severe infection for various time periods (less than one year, between one and two years, between two and five years and greater than five years) are summarised in Table 2-3. The incidence rate for severe infections was highest in the first year following diagnosis at 179.2 (95% CI 137.8 – 201.1) severe infections per thousand person-years. It was sequentially lower in subsequent time periods. The latest time period, beyond five years, demonstrated a severe infection incidence of 35.1 (95% CI 29.4 – 41.6) events per thousand person-years.

Table 2-2 | Registry baseline characteristics

Variable		RKD	Skåne	Combined
Total		n = 746	n = 374	n 1120
Mean age (years)		59.2	65.0	61.1
Sex	Female	327 (43.8)	174 (46.5)	501 (44.7)
	Male	419 (56.2)	200 (53.3)	619 (55.3)
AAV diagnosis	GPA	316 (42.4)	192 (51.3)	508 (45.4)
	MPA	385 (51.6)	159 (42.5)	544 (48.6)
	EGPA	45 (6.0)	23 (6.1)	68 (6.1)
ANCA autoantibody specificity	PR3	336 (45.0)	187 (50.0)	523 (46.7)
	MPO	363 (48.7)	161 (43.0)	524 (46.8)
	Negative	34 (4.6)	26 (7.0)	60 (53.6)
	Unknown (missing)	13 (1.7)	0 (0)	13 (1.2)
Mortality	Alive	613 (82.2)	187 (50.0)	800 (71.4)
	Deceased	133 (17.8)	187 (50.0)	320 (28.6)
Mean follow-up (years)		8.1	8.0	8.0

EGPA = eosinophilic granulomatosis with polyangiitis, GPA = granulomatosis with polyangiitis, MPA = microscopic polyangiitis, MPO = anti-myeloperoxidase antibody, PR3 = anti-proteinase 3 antibody, RKD = Ireland Rare Kidney Disease registry

Table 2-3 | Incidence rate of severe infections over different time periods post diagnosis

Registry	Time period	Number of Infection events	Total time-at-risk (years)	Event rate per thousand person years (95% CI)
RKD	Less than 1 year	110	649	169.5 (137.8, 201.1)
	Between 1 and 2 years	27	573	47.0 (29.3, 64.8)
	Between 2 and 5 years	43	1315	32.7 (22.9, 42.5)
	More than 5 years	68	2277	29.9 (22.8, 36.9)
Skåne	Less than 1 year	68	344	197.6 (153.5, 250.6)
	Between 1 and 2 years	30	315	95.2 (64.2, 136.0)
	Between 2 and 5 years	44	781	56.3 (40.9, 75.6)
	More than 5 years	66	1538	42.9 (33.2, 54.6)
Combined	Less than 1 year	178	993	179.2 (153.9, 207.6)
	Between 1 and 2 years	57	888	64.2 (48.6, 83.1)
	Between 2 and 5 years	87	2096	41.5 (33.3, 51.2)
	More than 5 years	134	3815	35.1 (29.4, 41.6)

95% CI = 95% confidence interval, RKD = Ireland Rare Kidney Disease registry

2.6 Discussion

2.6.1 Data quality – results

A pilot DQ analysis of core variables was undertaken. Potential strategies to improve registry DQ are discussed below. The DQ analysis covered all pilot registries and evaluated a range of variables that were likely to be representative of broader data collected by the registries. Variables likely to have a high level of DQ, such as age and date of birth, but also variables with potential for lower DQ, such as BVAS and creatinine, were evaluated. We found that uniqueness of registry IDs was 100% across all registries. Duplication of patients, but with separate unique identifiers, was similarly low with the exception of POLVAS. POLVAS had 3.6% of patients possibly entered more than once under different unique identifiers. This uncovered an issue requiring further investigation and provided an opportunity for practical improvement steps to be considered, as discussed below. Consistency with respect to data

dictionary format was 100% across all registries and variables. This likely represents data validation systems in the various software platforms used by the registries, whereby it is only possible to enter data in the correct format for the variables that were checked. Logic tests of consistency for important dates such as date of birth, diagnosis and death were typically 100% and were 98% at lowest in one registry. Plausibility tests of consistency for BVAS and creatinine were also typically 100%. The lowest reported compliance with plausibility for creatinine was 99.8%. This suggests a low frequency of major errors for these variables but does not exclude more subtle errors. Completeness was high for certain variables such as sex, date of birth and date of death. However, there was substantial variability in completeness for other variables, such as serum ANCA autoantibody status, which varied from 52% in the French registry to 100% in the GEVAS, RKD and Skåne registries. This likely reflects the fact that the French registry was established around a decade before the introduction of ANCA autoantibody testing. A basic test of correctness was performed by each registry team. A minimum of 10 patients from each registry was sampled by the local DQ analyst. The eight DQ variables for each patient were then compared to a gold standard source of information - the respective clinical record for all registries. Correctness was high across most variables at 90% to 100%. This was lower for RKD for Comorbidity, BVAS and Creatinine at 71%, 85% and 85% respectively.

2.6.2 Impact of data quality

On review of the rare disease registry medical literature, while many evaluations of data quality of such registry data have been undertaken, it is clear that standards which indicate adequacy of data quality are lacking (Aerts et al., 2021; Taruscio et al., 2014; Trama et al., 2017). A qualitative commentary is often provided, suggesting that DQ levels are “good” or “high”, with recommendations typically made for approaches for improving DQ, however beyond intuitive statements that high quality data is necessary for reliable research and the provision of good clinical care, clear evidence for adequate levels of DQ is lacking. Similar themes emerge from literature beyond the rare disease setting in clinical domains such as cancer and trauma (Chiang et al., 2015; O’Reilly et al., 2016). This is not surprising, due to the complexity of quantifying required DQ for any given scientific investigation and that different

thresholds of DQ may be required depending on the specific research question or DQ domain. The levels of DQ demonstrated in the current analysis of the FAIRVASC registries suggest that DQ is similar to other registries considered to have a high level of DQ (Aerts et al., 2021; Taruscio et al., 2014). This provides a reasonable level of confidence that systematic bias will be minimised in the eventual outputs of the project. Planned projects include cluster analysis to determine novel AAV phenotypes, prediction analysis for death and ESKD as well as making summaries of key variables available via the main FAIRVASC interface. The interface will provide a portal to explore the impact of AAV in terms of outcomes, helping to assess unmet need, and will supply information regarding the demographics and epidemiology of AAV across different nations, thus facilitating the planning and design of future clinical studies.

2.6.3 Strategies to improve data quality

From the outset the FAIRVASC consortium has not viewed DQ assessment as a ‘one-off’ task. We have elected to integrate a DQ culture into the FAIRVASC project and plan future regular iterations of DQ assessment. A repeat DQ study is already underway and is evaluating additional variables, specifically induction treatment and serum C-reactive protein. Ultimately, we aim to achieve a cycle of assessment and improvements, leading to FAIRVASC containing high quality data. This data will be suitable for a variety of scientific analyses, the results of which the vasculitis community can have confidence in, with bias having been minimised. Notably this pilot study did not include infection as a variable for formal DQ assessment. Future DQ studies will seek to evaluate this variable.

Possible duplicate patients under different IDs were identified at a significant level in the POLVAS registry but also in the Czech and UKIVAS registries. Further detailed analysis of these cases is warranted to confirm the likelihood of duplication. Following further assessment, subjects confirmed as duplicates could easily be removed from an analysis at a data cleaning stage, improving the reliability of subsequent inferences based on this dataset. Systems for data entry could be enhanced, for example an automated warning when a patient is registered that a similar patient already exists in the registry could be directed at the user, allowing them to check likelihood of duplication at an early stage.

This could help reduce unnecessary work for registry personnel, conserving limited local resources.

A potential improvement step for consistency could be a further enhancement of data validation systems, if suitable systems are not already in place. For example, for an individual performing data entry, it would be easy to mistakenly add an extra digit to a numerical value, making the value erroneous by a factor of ten. If a data entry system only allowed entry of plausible values, some of these erroneous values could be avoided. Notably, many registries already utilise such data validation approaches at the data entry stage. More elaborate systems are possible. For example, if a blood test value is entered that is substantially different to previous values or a previous trend, then a warning message could be shown to the user. Alternatively, automated input of certain variables from a laboratory database to a registry could be applied, reducing the capacity for human error. This would also reduce manual workload for registry personnel.

Strategies to improve completeness include targeted additional data collection for specific variables where funding and resource allow. Missing data is well recognised as a DQ issue across multiple study designs and various strategies exist for dealing with this at the data analysis stage. A useful additional to the FAIRVASC interface would be a function to allow an assessment of missing data when exploring the data, helping to inform which strategies may be useful to mitigate against low completeness. For some analyses, for example where completeness is independent of confounders (i.e. data that is missing completely at random (MCAR)), a complete case analysis may be valid.(Ross et al., 2020) Multiple imputation is an alternative option and can help produce results with minimal bias, even with low levels of completeness.(Blazek et al., 2020) One could envisage a sub-study of FAIRVASC, where multiple imputation is carried out on datasets at the registry level before being uplifted to local triplestores, proving interface users with a multiple imputation version of the FAIRVASC dataset.

2.6.4 Severe infection incidence – main results and comparison with other studies

Incidence rate was highest in the first year following diagnosis at 179.2 (95% CI 153.9 – 207.6) events per thousand person-years and decreased over subsequent time periods. This highlights the well-recognised impact of the potent immunosuppression used in the initial months after AAV diagnosis to control disease activity as well as increased susceptibility due to disease related factors. That infection is common in the first year following diagnosis is well recognised. The largest published study on this topic is by McGregor and colleagues, who included 489 patients – less than half the size of the current study at 1120 participants. McGregor et al evaluated the risk of infection, including severe infection, over different time points in a longitudinal registry based AAV cohort in the USA. They found 22% had at least one severe infection within the first year following diagnosis. Over two years of follow-up, most infections occurred within the first year and the greatest number of infections occurred within the first three months. The number of patients who experienced at least one severe infection over the two-year period was reported at 96 out of a population of 374. This equates to at least 128 severe infections per 1000 person-years.(McGregor et al., 2015) Sarica et al undertook a multicentre matched cohort study of 379 AAV cases which utilised data linkage to national records in Scotland. 35% of individuals experienced at least one severe infection over median 3.5 years follow up. Incidence was highest in the first year following diagnosis, particularly in the first 30 days when the incidence rate ratio was 10.6 (95% CI 4.0-28.0) compared to general population controls. The incidence rate ratio for the 181 – 365 day period was 6.6 (95% CI 4.1-10.5) (S. H. Sarica et al., 2020). Rathmann et al performed a population-based cohort study using the Skane registry, therefore the underlying patient data was very similar to that reported in the current study. This study also reported incidence rates over the first and second year. In 129 AAV patients, the incidence rate over the first year was 22.1 per 100 person-years (95% CI 16.7-27.4) and for the second year was 11.4 per 100 person-years (95% CI 9.5-13.3) (Rathmann et al., 2021). The results of the current study demonstrate a similar trend to the key studies highlighted, in that severe infection incidence is highest soon after diagnosis but then falls substantially over subsequent years. The reported incidence rate in the current study of 179.2 severe infections per 1000 person-years appears consistent with

findings from the literature, as described above (McGregor et al., 2015; Rathmann et al., 2021).

The larger registry in the current severe infection incidence study was RKD, a nephrology registry. Lower renal function is a recognised prognostic factor for severe infection. It would therefore be expected that the severe infection incidence would be higher in RKD, a nephrology registry, compared to Skåne, a population-based registry. This was not the case in the current study. That the mean age of the Swedish cohort was 5.8 years older may, at least partly, account for this. Other possible explanations include the proportion with renal dysfunction not being substantially different across the two registries. Renal data was not available to aid further exploration of this. Other possibilities include variability in data collection technique, immunosuppression regimens or geographical variation.

2.6.5 Strengths

FAIRVASC represents the largest amalgamation of AAV data yet assembled. Achieving an adequate sized epidemiological study is challenging in the rare disease setting, but is essential in order to achieve sufficient statistical power and to enable reliable estimates of association to be determined. FAIRVASC has deployed state of the art web technology to enable federalisation of diverse data sets and thus establish this large data resource. It has achieved this whilst maintaining high standards of data protection and is fully compliant with modern European legislation ("REGULATION (EU) 2016/679 OF THE EUROPEAN PARLIAMENT AND OF THE COUNCIL of 27 April 2016 on the protection of natural persons with regard to the processing of personal data and on the free movement of such data, and repealing Directive 95/46/EC (General Data Protection Regulation)," 2016). This is of particular importance in the rare disease setting where, due to the rarity of conditions such as AAV, individuals are at greater risk of being identifiable despite attempts at anonymisation. The size of the FAIRVASC data sets is significantly larger than many existing datasets on which previous research has been based. The aim is that new insights into the epidemiology of AAV will be possible. While FAIRVASC has successfully federalised the data of 6,104 participants, the presence and quality of severe infection data was not sufficient for all registries to be included in the incidence

study of this complication. Only two registries with a total of 1,120 AAV participants were used for the incidence study – much smaller than the intended goals of the FAIRVASC project. While a smaller study than anticipated, the current study still represents the largest study of severe infection incidence in AAV to the author's knowledge. The three closest comparable studies in terms of size have between 400 to 500 participants, while the current study is over double this size (Lai et al., 2014; McGregor et al., 2015; Solans-Laque et al., 2017).

Crucial to the aims of FAIRVASC being achieved is that the underlying data is of high quality. The core DQ data reported in the current study indicates that, for the variables assessed, FAIRVASC data is of a similar standard to other carefully maintained registries, as explored above. A broad range of DQ domains were explored in a variety of clinically important variables. A continually improving DQ culture has been established in FAIRVASC. This will give confidence to researchers about the validity of output.

Data from the FAIRVASC project will be highly generalisable to real-world populations with AAV. This is partly evidenced by demonstration of similar baseline demographics to a typical AAV population (Kitching et al., 2020). A broad range of European countries were represented, covering variable demographics and healthcare systems. FAIRVASC has been designed such that existing registries will be able to easily transform their data to be made available through the FAIRVASC interface. Expansion is planned internationally, with current interest from many national AAV registries including Spain, Turkey, Denmark and Australia. This will further enhance the generalisability of analyses emerging from the project.

AAV registries vary considerably in size and maturity. As a result, the underlying structures of the datasets varies substantially. In many cases identical clinical phenomena were described with different terminology. In other cases, similar concepts were recorded requiring cross referencing of different data fields within the registries such that the same information could be derived from each. FAIRVASC benefitted from a dedicated team of clinicians, computer scientists and health data experts in aligning these disparate data sources in a reliable manner.

Forms of bias were mitigated against to limit any impact in the severe infection incidence study. This includes survival bias and information bias. Survival bias is when either death or the study outcome, in this case severe infection, occurs and adversely affects inclusion or subsequent participation in a study. A large majority of patients in the study were prospectively identified and comprehensive follow-up was performed via clinical records. This would have the effect of minimising survival bias and minimising information bias in the form of “loss to follow-up”.

2.6.6 Limitations

There were limitations that are important to acknowledge. With respect to the core DQ assessment, levels of data quality were variable across registries. Aspects of the DQ analysis could be improved upon such as the biological plausibility tests, particularly with respect to serum creatinine. In this study the biologically plausible range for serum creatinine was defined as 0-5000 micromol/L. On further reflection, a serum creatinine of zero is implausible and, based on the clinical experience of the thesis author and supervising team, is not observed clinically. The upper limit of 5000 micromol/L could also be revised: there are published case reports of higher values being observed. One report (published after the DQ approach was designed) described a serum creatinine value of 73.8 ml/dL, approximately equivalent to 6200 micromol/L (Persaud et al., 2021). Logic tests such as date of birth being greater than date of diagnosis will have high sensitivity for major consistency errors, but are at risk of missing less obvious errors. Correctness, potentially the most important DQ domain, was examined to a limited extent in the study. Ten individuals were checked for correctness across a range of variables. This may be too few registry participants to accurately identify levels of correctness and it will be difficult to determine if changes over time are statistically significant due to a lack of power with respect to this measure. Due to limited local resource at the participating registries it was not possible for more extensive correctness checks to be undertaken.

While a systematic assessment of DQ was carried out, infection data was not part of this assessment. A qualitative assessment of registry severe infection data was conducted in the form of a survey of registry owners. This revealed

that registry severe infection data was variable in terms of overall presence and quality. Where infection data was collected, registries were not typically directly funded for this data capture, therefore substantial missingness may result. Some registries did not collect severe infection data at all. Another registry collected such data, but this was in free-text format and was not possible to assess or use.

In the incidence study, we noted a substantial decrease in severe infection events over time. While this is biologically plausible, it remains possible there was degree of detection bias, whereby severe infections that occur in later years are less likely to be detected, as follow-up is typically not as frequent at this stage of the disease. Selection bias may also occur, in that individuals who present to hospital more frequently, for example with infection, may be more likely to encounter clinicians involved in the registry and therefore become a registry study participant. In theory this could inflate severe infection events.

Ideally infection outcome data would have been presented for multiple registries, but unfortunately this data was not available. Age was available as a mean, but due to the prototype nature of the FAIRVASC web interface, was not available in dichotomised age brackets. Had age been available in a dichotomised form, it would have been desirable to perform an age and sex adjusted analysis of infection rates using direct standardisation (Naing, 2000). This would have allowed comparison of severe infection incidence across different registries, while taking demographics into account. Future iterations of the FAIRVASC infrastructure will seek to use novel encrypted federated learning techniques which will allow more advanced statistical approaches to be undertaken remotely on fully encrypted patient level data. Within the FAIRVASC consortium this has been successfully trialled using fabricated data with logistic regression. More complex techniques such as survival analysis and machine learning methods are also technically feasible.

2.6.7 Conclusion

A pilot DQ study of the initial FAIRVASC registries was carried out and indicated high levels of DQ across several core variables and DQ domains. The data was deemed sufficient for the initial FAIRVASC research activities, namely cluster

analysis, predictive modelling and presenting count data of baseline characteristics and outcomes via the FAIRVASC web interface. The FAIRVASC web interface utilises semantic web technology to federate AAV registry data across several European nations. The interface was used to conduct a study of severe infection incidence. Due to issues identified with severe infection data quality, only two registries were used for the severe infection incidence study. This still resulted in the largest study yet undertaken of this complication, to the author's knowledge. This study highlights that severe infection is a major complication in AAV. Severe infection incidence was substantially higher in the first year following diagnosis. This work highlights severe infection in AAV as a research priority for the AAV clinical and scientific communities.

2.7 Summary

This chapter found that European AAV registries contain data of suitable quality for the principal research activities of the FAIRVASC EU consortium. Novel semantic web technology was used to federalise the registries, but with the underlying data staying in the host institution in a secure manner. A severe infection incidence study was performed using the prototype FAIRVASC infrastructure, though due to DQ issues only two registries were used for this. Severe infection occurred with high frequency, especially in the first year after diagnosis. Chapter 3 will seek to identify prognostic factors that predict severe infection, namely glucocorticoid dose thresholds. Subsequent chapters utilise the knowledge derived by developing prognostic models aiming to help clinicians and scientists predict severe infection in AAV and its consequences.

3 Glucocorticoids as a prognostic factor for severe infection in ANCA-associated vasculitis (AAV): dose relationship retrospective cohort studies

3.1 Overview

The previous chapter investigated the frequency of severe infection using the novel FAIRVASC federated rare disease platform. This represented ‘overall prognosis research’: one of the four main study types described in the PROGRESS framework which classifies prognosis research. Now that we have established that severe infection occurs with high frequency, and with the knowledge that its consequences can result in death, the importance of being able to determine the likelihood of severe infection based on patient characteristics is clear. The next step in prognosis research is to identify prognostic factors: patient characteristics that are associated with the outcome of interest. Glucocorticoids are well recognised as contributing to severe infection risk, but the impact of different dose thresholds has not been explored in AAV. This chapter will utilise a large, real-world, observational dataset to ascertain the value of glucocorticoid exposure as a prognostic factor in individuals with AAV.

3.2 Abstract

3.2.1 Background

For decades, glucocorticoids have been a cornerstone of AAV management. In recent years, efforts have been made to find strategies to reduce or replace their use due to a wide range of adverse effects. Based on observational and randomised data, one of the most concerning adverse effects is susceptibility to severe infection. While this is well recognised, it is unclear whether different thresholds of glucocorticoid exposure are associated with differing risk for severe infection. This chapter will evaluate different glucocorticoid thresholds as prognostic factors for severe infection.

3.2.2 Methods

AAV patient data was identified from national Scottish datasets. Linked datasets such as those for hospital admissions and community prescribing were used to ascertain comorbid conditions and severe infection outcome, and glucocorticoid exposure, respectively. Four separate Cox proportional hazards models were developed. Glucocorticoid exposure was represented at daily prednisolone equivalents. Severe infection was defined as a hospital admission associated with infection. The observation period was 12 months. Glucocorticoid thresholds evaluated included: 1) 0 mg, >0-10 mg and >10 mg; 2) zero mg, >0 to 5 mg, >5 to 10 mg, >10 to 20 mg and >20 mg, 3) 0 mg vs 5-7.5 mg. A fourth model evaluated glucocorticoid exposure as a continuous variable.

3.2.3 Results

Suitable data was available for analysis in 978 individuals with AAV. For model 1) glucocorticoid exposure >10 mg had hazard ratio (HR) 1.93 (95% CI 1.19 – 3.13, p = 0.008). The referent for all models was zero glucocorticoid exposure. Model 2) showed similar hazard ratios above 5 mg, with >20 mg having HR 2.05 (95% CI 1.23 – 3.43, p = 0.015). In model 3) the 5 mg to 7.5 mg group had HR 2.55 (95% CI 0.72 – 8.94, p = 0.145). Model 4) showed a HR 1.01 (95% CI 1.00 – 1.02, p = 0.047) for every milligram increase in glucocorticoid exposure.

3.2.4 Conclusions

Glucocorticoid exposure thresholds above 10 mg daily all had a substantial positive association with severe infection. For lower exposures an association was not clearly determined. For every milligram increase in glucocorticoid exposure, the odds of severe infection increased on average by 1%. Glucocorticoid dose threshold may have value as prognostic factors for severe infection.

3.3 Introduction

3.3.1 Background

Glucocorticoids were one of the first immunosuppressive therapies to be introduced for treatment of AAV. Initial reports in the 1950s described cases of GPA, then known as Wegener's granulomatosis, being managed with cortisone (Fahey et al. 1954). Glucocorticoids appeared to ameliorate the disease in some individuals and perhaps extended survival, but ultimately did not prevent the progression to death (Hollander and Manning 1967). A major turning point came from studies on the effect of cytotoxic therapy on AAV, at the National Institute of Heath (NIH) in Maryland, USA. Led by Anthony Fauci, these studies showed that, combined with glucocorticoids, cyclophosphamide was shown to induce disease remission (Fauci et al., 1971). Disease prognosis was transformed from a condition considered universally fatal to one where most individuals were alive at the end of the first year following treatment.

3.3.2 Glucocorticoids: mechanism of action

Glucocorticoids act through pleotropic mechanisms. Potent immunosuppressive effects are predominantly mediated via cytosolic glucocorticoid receptors. These receptors exert genomic effects that result in upregulation of anti-inflammatory proteins and suppression of proinflammatory proteins. Non-genomic effects include action on cellular membranes, on membrane-located glucocorticoid receptors and non-genomic actions of intracellular glucocorticoid receptors (Stahn and Buttgereit 2008).

3.3.3 Glucocorticoid therapy: outcomes and adverse events

Therapy for AAV has been evaluated in several multicentre randomised controlled trials, a major success for a rare condition (Wallace and Miloslavsky 2020). However, trials of glucocorticoid treatment were lacking until the PEXIVAS study, the largest RCT yet conducted in AAV, was reported. In this open-label trial 704 AAV patients were randomised in a two-by-two factorial design to evaluate the use of plasma exchange versus no plasma exchange and a standard-dose glucocorticoid regimen versus a reduced-dose regimen. With respect to the glucocorticoid comparison, the reduced-dose regimen was found

to be non-inferior to the standard regimen, both in terms of overall patient survival and progression to end-stage kidney disease (ESKD) (Walsh et al., 2020). Severe infections were observed less frequently in the reduced-dose group compared to the standard-dose group, with an incident rate ratio of 0.69 (95% CI 0.52-093). Similarly in LoVAS, a multicentre trial conducted in Japan, a low-dose glucocorticoid regimen was non-inferior with respect to the primary outcome of remission at six months and showed reduced severe infections, when compared to a standard-dose regimen (Furuta et al., 2021).

There have been various observational studies published describing outcomes and adverse effects of glucocorticoid therapy in AAV, but most are small or methodologically flawed. McGregor *et al* sought to explore the effects of glucocorticoids in a small cohort totalling 147 individuals. In this report, patients receiving glucocorticoids beyond six months following therapy initiation suffered a higher rate of infections, when compared to patients not receiving glucocorticoids beyond six months. No significant differences were apparent with other important outcomes such as occurrence of ESKD or mortality (McGregor et al., 2012).

Current guidance recommends a reduced-dose glucocorticoid regimen consistent with the PEXIVAS trial (Hellmich et al., 2023). Following stepwise glucocorticoid tapering over the initial months of the regimen, this notably includes a prolonged duration of low-dose glucocorticoid for over six months, at 5 mg daily. After 12 months, current guidelines recommend that ongoing glucocorticoid dosing is left to the discretion of the clinician. This may result in further prolonged treatment at this dose. It is unclear what impact chronic low-dose glucocorticoid therapy has on individuals with AAV. Studies in other settings have found an association between low-dose glucocorticoid therapy and severe infection, such as a large observational study in rheumatoid arthritis which showed an association at the low dose of 5 mg (George et al., 2020).

3.3.4 Optimising the use of glucocorticoids

While glucocorticoid treatment approaches have been compared to other treatments, such as plasma exchange in MEPEX and avacopan in ADVOCATE, to date only two studies have compared glucocorticoid regimens in an RCT setting:

the PEXIVAS study (Jayne et al., 2007, 2021; Walsh et al., 2020) and LoVAS (Furuta et al., 2021). In PEXIVAS, the cumulative glucocorticoid dose in the reduced-dose arm was less than 60% of that in the standard-dose arm. While PEXIVAS represented significant progress in the care of vasculitis patients, the optimal application of glucocorticoids in AAV management remains to be clarified. Several aspects need elucidation: optimum duration, dose, total cumulative dose, speed of taper and long-term dosing. Prognostics markers, such as clinical disease activity, or prognostics scores, aimed at predicting outcomes or adverse events, could be utilised in dynamically adapting the dose over time. Decisions regarding investigation of these research questions must take into consideration other urgent research needs in the care of AAV patients. These include optimisation of other existing treatments, introduction of new treatments, introduction of new tests and evaluation of the use of prognostic scores – all of which, ideally, should be evaluated using RCT methodology. However due to restrained resources and a limited number of participants available for enrolment, the vasculitis research community must prioritise clinical studies which have the maximum potential utility for patients. It therefore is logical to utilise existing data sources to inform decisions about which research agenda to pursue. Through observational, real-world data, a better understanding of the relationship between glucocorticoids, outcomes and adverse effects can be achieved. An important concern around the use of glucocorticoids in AAV is the recognised impact on frequency of severe infections. This both in terms of the magnitude to which glucocorticoid exposure is associated with severe infections and whether there is a glucocorticoid dose threshold at which risk increases.

3.3.5 Rationale for this investigation

While investigation of the causal role of glucocorticoids in relation to severe infection is important, the focus of the current study was to determine the ability of glucocorticoid dosing to predict severe infection as a prognostic factor.

It is well established that glucocorticoids cause adverse effects across many settings. The contribution of glucocorticoids to the occurrence of severe infection has been evidenced in observational and randomised data (McGregor et al., 2012; Walsh et al., 2020). Severe infection, being the main cause of early

mortality in AAV patients, is a particular concern for patients and clinicians (Little et al., 2010). Being able to predict the occurrence of severe infection would be highly desirable in the setting of AAV patients. It would have the potential to help clinicians and patients make treatment decisions regarding the intensity of immunosuppression, it could be a useful research tool for targeting higher risk patients for recruitment to interventional studies and would give patients clearer expectations about the risk of adverse events.

Given that glucocorticoids are generally accepted as having a causal role in contributing to the occurrence of severe infections, it follows that glucocorticoid use should be predictive of severe infection. In this sense it would be described as a prognostic factor – a clinical parameter in a given disease population which can be used to determine an individual's likelihood of a future event occurring (Riley et al. 2009). Prognosis factor research falls under the purview of the PROGRESS (PROGnosis RESearch Strategy) framework, described in the introduction chapter, section 1.14.1 (Hemingway et al., 2013).

The current study should be considered prognostic factor research – stage 2. If clear associations are found between glucocorticoids and severe infection, particularly if a dose-response relationship can be demonstrated, then this would also be supportive evidence for a causal role for glucocorticoids. However specific study designs and methodology would be more appropriate for addressing this causal question as a primary aim, such as: target trial emulation with respect to design, considering the time varying nature of medication and specifically only including confounders in a multivariable analysis. It would also be important to include a wider range of covariates such that all recognised potential confounding variables are included – this is not strictly necessary in a prognostic factor study. (Hernán and Robins 2016)

3.3.6 Aims

The aims of this study are to explore the association between glucocorticoids, in various prespecified dose groupings, and severe infection in AAV. This will be achieved through the development of separate multivariable models for each different dose grouping and including glucocorticoid dose as an exposure in each model. Models will be designed in a manner suitable for prediction, as opposed

to primarily aiming to support a causal role for glucocorticoids – an important but separate research question.

3.4 Methods

The Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) statement guidance for cohort studies was used to guide reporting of this study. (Vandenbroucke et al. 2007)

3.4.1 Data source, study design and setting

Vasculitis Outcomes in relation to Care Experiences (VOICES) was a services mapping study of vasculitis care and outcomes with projects covering both Scotland and the whole of the United Kingdom. VOICES included a Scottish data linkage matched cohort study which aimed to examine patterns of vasculitis care and outcomes. This dataset was developed using the advanced data linkage capabilities in Scotland via services provided by the electronic Data and Research Service (eDRIS) within Public Health Scotland (PHS). This dataset was utilised in the current study in line with the overall objectives of VOICES. The dataset identified all patients in Scotland with a relevant International Statistical Classification of Diseases and Related Health Problems 10th Revision (ICD-10) code for AAV and Giant Cell Arteritis (CGA) from the Scottish Morbidity Record admissions database (SMR01). SMR01 captures hospital admission data from all non-obstetric and non-psychiatric inpatient and day case care episodes. Patients were identified from a start date of 1st April 1996, when ICD-10 was adopted in Scotland, to 31st October 2020, the latest available update. Ten controls were identified, matched by age, sex and health board of residence. Patient identifiers were used to link to administrative health care data from multiple national datasets. These datasets are described in Table 3-1. An index date representing probable AAV diagnosis was defined as the earliest admission date associated with an AAV ICD-10 code. A look-back period of 5 years allowed comorbidities that occurred prior to the index date to be determined, according to established methodology (Quan et al. 2005). Data linkage was performed by PHS using a robust approach that has been demonstrated to result in highly accurate and complete data (Evans and MacDonald 1999; Scottish Public Health Observatory 2022). Application to use these data sets typically requires ethical

approval and a successful application to the NHS Scotland Public Benefit and Privacy Panel for Health and Social Care (HSC-PBPP), the body in Scotland which scrutinises applications for research using national health data and considers the potential public benefit of proposed work. An application to HSC-PBPP was successful, specific ethical approval was not required as this was covered by existing National Safe Haven (NSH) approvals. In order to access study data, all individuals in the study team were required to have up-to-date UK information governance training, such as that provided by the UK Medical Research Council Regulatory Support Centre (MRC Regulatory Support Centre 2023). Data was accessed through the secure NSH, using two-factor identification. Additional information regarding the study is available online (VOICES, University of Aberdeen 2022).

Table 3-1 | Public Health Scotland linkage datasets – description and availability

PHS dataset	Description	Years data available*
General Acute/Inpatient (SMR01)	SMR01 captures data at the encounter level on all inpatient and day case care episodes, with the exception of mental health and obstetric specialties. An entry in the dataset is created at the end of an episode of care. This may be being discharged to home, change of care to a different specialty, change of hospital or death. Over one million entries are generated each year.	1981 onwards
National Records of Scotland (NRS) Death Records	All deaths occurring in Scotland. Typically 55,000 deaths are registered annually.	1974 onwards
Prescribing Information System (PIS)	PIS captures data on all community prescriptions in Scotland. Most are written by General Practitioners (GPs), but hospital prescriptions that are dispensed in the community are also included. Approximately 100 million data items are captured every year.	January 2009 onwards

*** 'Years data available' represents data available for linkage for researchers via Public Health Scotland, data may exist for earlier time periods. Data linkage became more practical in 2009 following application of Community Health Index (CHI) numbers.**

The dataset for this study was derived from the overall VOICES dataset as follows (and described in results section Figure 3-1): first GCA cases were excluded, then individuals whose index date was over six months before the start date of PIS records were excluded, following this AAV controls were excluded then, finally, individuals who died before the end of the glucocorticoid exposure observation period were excluded. This resulted in a dataset of individuals with AAV from the VOICES cohort who were alive at the end of glucocorticoid exposure period and had PIS prescription data available for the

whole of the glucocorticoid exposure period. Patients who did not have any PIS data available during PIS era were assumed not to be taking any medication.

The setting covers the entire Scottish population who could have been identified as having vasculitis in relation to an inpatient admission. Data collection was undertaken as part of routine heath care and national records processing, such as clinical coding and death registration. In Scotland, at the end of each hospital or day case admission, trained clinical coders assign codes to describe clinical events and comorbid conditions. This is according to standardised guidance (Public Health Scotland, 2023). As such this study is a retrospective cohort study utilising administrative data.

3.4.2 Patient and public involvement

Collaboration with the Aberdeen Centre for Arthritis and Musculoskeletal Health's user group and patients identified through national support groups (Vasculitis UK and the Lauren Currie Twilight Foundation) was undertaken as part of VOICES. This identified patient and carer priorities. This process informed the design and aims of the current study. (VOICES, University of Aberdeen 2022)

3.4.3 Participants

The VOICES study sought to identify all adults, ages 16 years or older, identified within the SMR01 database with a code for AAV or GCA. For this study, individuals with a clinical code for AAV were included. The following ICD-10 codes were used to identify patients: M31.1 (granulomatosis with polyangiitis), M31.7 (microscopic polyangiitis) and M30.1 (eosinophilic granulomatosis with polyangiitis). The first SMR01 admission associated with a clinical code for AAV was designated as the index date. Patients were included if their index date occurred between 31st July 2008 (six months prior to the start of PIS data) up to 31st October 2020 (the end of the observation period). The start date was selected so that all patients included in the study had full data available for the glucocorticoid exposure period. Patients were followed up from the end of the exposure period to 365 days post-index, to the time of death or to the end of the observation period, whichever occurred first.

3.4.4 Exposure

The prognostic factor of interest was glucocorticoid exposure. Due to PIS dataset being based on prescribed medication that was obtained at community pharmacies, the exact prescribed dose of a given medication is not known at any specific time point. The dataset includes medication that was dispensed each month with variables including drug name, the preparation strength and number of units (e.g. tablets) dispensed. Using a 90-day period of this information, it was possible to estimate the glucocorticoid exposure according to a simple formula (preparation strength x number of units / 90 days). The 90-day interval was selected based on existing literature relating to glucocorticoid exposure and infections in RA, described below. As described in Chapter 1, in studies of AAV patients where prognostic factors for severe infection are explored, the time window for glucocorticoid exposure was frequently not clearly specified. Where the time window was specified, this was typically at baseline, another specific time period such as a six months or over an initial index period such as the first month following diagnosis (Kronbichler et al., 2018; McGregor et al., 2012; Sakai et al., 2019). No study specified an exposure window over which cumulative glucocorticoid exposure was calculated. Therefore, studies in RA, as an adjacent immune-mediated inflammatory disease (IMID), were deemed a useful alternative source of methodology to derive a relevant cumulative glucocorticoid exposure period. A systematic review and meta-analysis of the association between glucocorticoid exposure and the likelihood of infection in the setting of RA found many observational studies showing steroid dosing captured over a three-month period was associated with subsequent infection. Of the groups reported with a known time window of possible exposure to glucocorticoids, this group had the strongest association with subsequent infection, with a mean relative risk of 1.7 (95% CI 1.47-1.97). Other groups were glucocorticoid exposure at baseline, within 6 months of the outcome, glucocorticoid administered ‘ever’ or an unclear time window (Dixon et al., 2011). Another study supporting the 90-day time window was Dixon et al 2012. This was a large case-control study performed in RA patients in Quebec, Canada. It showed that current and recent glucocorticoid doses had the greatest impact on the risk of severe infection, with “any use in the past 90 days” carrying the largest odds ratio at 2.26 (95% CI 2.02-2.54) (Walsh et al., 2020).

Almost all individuals prescribed glucocorticoid were prescribed prednisolone. The following conversion factors were applied to non-prednisolone glucocorticoids as per the British National Formulary: betamethasone – 6.667, dexamethasone – 6.667, hydrocortisone – 0.25 (Joint Formulary Committee, 2022). As a result, the unit for glucocorticoid dosing in this study was 'daily prednisolone equivalents'. The index date for most participants was deemed likely to be close to the date of diagnosis when treatment with induction therapy would be ongoing. Induction treatments for AAV, such as cyclophosphamide and rituximab, are typically administered and supplied by hospital pharmacies, not primary care. Therefore, data documenting administration of such induction treatments was not available within the PIS dataset. Given that other immunosuppressive treatments, particularly induction treatment, would be important confounders and that glucocorticoid regimens that accompany induction treatment have already been explored in RCTs as described above, an *a priori* decision was made to examine the association between glucocorticoid exposure from six months after the index date. By this time point most individuals treated for AAV, including those treated with a standard glucocorticoid regimen, should be receiving a stable low dose such as 5 mg prednisolone daily (Walsh et al., 2020). Higher doses at this time point may represent ongoing disease activity or relapse. Disease activity was not possible to derive from available datasets as a confounding variable.

Various glucocorticoid dose groupings were used as exposures in separate models. The objective was to explore the relationship between different glucocorticoid doses and severe infection. The comparisons and associated models explored different aspects of glucocorticoid dosing, with rationales described below.

3.4.4.1 Glucocorticoid dose: 0 mg vs >0-10 mg vs >10 mg

This evaluation divided individuals into three glucocorticoid exposure groups. This aim of this model was to confirm whether there is a similar relationship between glucocorticoids and severe infection in the AAV population as has been established for specific glucocorticoid doses in other immunosuppressed populations. There are various reports of this relationship, which typically group patients into no glucocorticoid, low dose glucocorticoid and high dose

glucocorticoid. Schenfeld *et al* undertook several comparisons in an RA population. Low dose was defined as >0 to 7.5 mg and high dose as >7.5 mg per day. The low dose group had an adjusted rate ratio of 1.4 (95% CI 1.21–1.60) for severe infection and 2.8 (95% CI 2.32-3.34) for the high dose group, when compared to a group on no glucocorticoid (Schenfeld et al. 2017). A similar study from Strangfeld *et al* showed glucocorticoid dosing from 7.5 to 14 mg per day had an adjusted incident rate ratio (IRR) for severe infection of 2.1 (95% CI 1.4-3.2), compared to no glucocorticoid, while dosing of 15 mg per day or greater had IRR 4.7 (95% CI 2.4-9.4) (Strangfeld et al. 2011). Notably a previous iteration of the American College of Rheumatology guidelines for the treatment of RA described low dose glucocorticoid at >0-10 mg and high dose as >10 mg (Walsh et al., 2020).

3.4.4.2 Glucocorticoid dose: multiple dose thresholds

This evaluation divided glucocorticoid dosing into the following groups: zero mg, >0 to 5 mg, >5 to 10 mg, >10 to 20 mg and >20 mg. The aim of this model was to explore in greater detail whether there is an increasing risk of severe infection associated with increasing dose thresholds. This could be a a linear relationship; a non-linear, but smooth relationship; or a threshold effect, whereby at a specific dose threshold, there is a substantial increase in risk – potentially a logarithmic relationship. Stepwise increased risk associated with increased glucocorticoid exposure has been reported in other settings. Dixon *et al* reported a systematic review and meta-analyses of randomised controlled trials and observational studies describing the association between systemic glucocorticoid treatment and infections. As the dose category was restricted to higher thresholds, there was an increasing risk of infection: >5 mg prednisolone equivalents carried a relative risk (RR) of 2.46 (95% CI 2.08-2.92), >10 mg had RR 2.97 (95% CI 2.39-6.69) and >20 mg had RR 4.30 (95% CI 3.16-5.84) (Dixon et al., 2011). A similar relationship may exist in AAV, but this has not yet been evaluated.

3.4.4.3 Glucocorticoid dose: 0 mg vs 5-7.5 mg

It is common for individuals with AAV to be on a small dose of steroid for a prolonged time, such as 5 mg daily prednisolone equivalent. This is evidenced by

both regimens in the PEXIVAS trial having over six months at this dosage (Walsh et al., 2020). Following on from 12 months, there is limited data available to guide ongoing steroid dosing and there is no formal recommendation for clinicians. A previous meta-analysis by Walsh *et al* suggested that ongoing low dose glucocorticoid for greater than 12 months from diagnosis may be beneficial for prevention of relapse (Walsh et al. 2010). As a result, it is common practice for some clinicians to continue patients on low dose glucocorticoid beyond 12 months. Anecdotally, where there is perceived increased risk of disease relapse, perhaps relating to previous relapse or anti-PR3 antibody positivity, some patients continue on low-dose glucocorticoid for the long term. In the Walsh *et al* meta-analysis risk of infection was not quantified. The authors acknowledge that the benefit of relapse prevention at this stage is unclear in terms of impact on long-term outcomes and quality of life. They state that a substantial proportion of individuals are able to tolerate withdrawal of glucocorticoid (Walsh et al. 2010). At present it is not know the extent to which low-dose glucocorticoid is associated with the risk of severe infection, when compared to no glucocorticoid. This model aimed to address this question.

3.4.4.4 Glucocorticoid dose: continuous

The final model considers glucocorticoid dose as a continuous linear variable. This aimed to quantify the average increased risk of infection per milligram (mg) glucocorticoid dose. As described above, dichotomising the exposure variable was intended to explore the effect of dose thresholds. However, dichotomising loses information from the analysis and may reduce statistical power. Also dichotomising may result in increased residual confounding, whereby some of the variable's confounding effect can continue to be unadjusted for. However, this is unlikely to be an issue where the only variable being dichotomised is the main exposure variable and the effect of this on associations between other variables and the outcome are not within the scope of the study questions. Arguably a non-linear representation of continuous glucocorticoid dose would be most informative, for example by fitting a cubic spline or transformation of the variable with multiple fractional polynomials (Harrell 2015). However, these procedures typically require increased power, therefore it was decided to proceed with a standard linear description of glucocorticoid dose for this initial evaluation.

3.4.5 Covariates

Choice of covariates for inclusion in an epidemiological model depends on study objectives. This study aims to evaluate different steroid dose groups as prognostic markers of severe infection. It does not seek to identify a causal relationship between steroid dosing and severe infection, an important but separate scientific question. STROBE guidance advises specific covariates to be identified, namely confounders and effect modifiers, however specifying these covariates would be important in a study which seeks to identify a causal role for the exposure of interest and is not necessary in a predictive study.

Predictors to include as covariates in the survival model were selected based on recognised association with severe infection. This was explored in the published prognostic literature relating to infections in AAV and IMID, and though the clinical judgment of the study team (Fine et al., 1997; Shapiro et al., 2003; Hespanhol and Bárbara, 2020; Bahlis et al., 2021; Ye et al., 2023). Selected variables included age, sex, Scottish index of multiple deprivation in deciles (SIMD) as a continuous variable and comorbidities including cancer (localised or metastatic), cerebrovascular disease, chronic heart failure, chronic respiratory disease, diabetes, liver disease and renal disease. Derivation of these covariates is described below:

3.4.5.1 Age

Date of birth (DOB) was included in all the linked datasets described in Table 3-1. As a privacy preserving measure the date component was not supplied, meaning that for all individuals only the month and year of birth is supplied. “14” was imputed as the date of DOB for all individuals. Index date is the date of admission associated with the first care episode in SMR01 that includes a diagnostic code for AAV. The study team considered it likely that that this will approximate to the date of diagnosis for most patients. Age at the time of index date in years was derived as “index date – DOB” in unit “years”.

3.4.5.2 Sex

Sex was available in all the linked datasets, coded as ‘male’ or ‘female’.

3.4.5.3 Scottish Index of Multiple Deprivation

The Scottish Index of Multiple Deprivation (SIMD) is a rank assigned to 6,976 geographical 'data zones' across Scotland (Scottish Government, 2020). Its purpose is to quantify deprivation. It is derived from over 30 indicators, for example local educational attainment, proximity to primary health care services, crime and unemployment. These indicators are grouped in seven domains. These domains are then used to form an index which is used to rank each data zone. Data zones are then grouped in quintiles or deciles. When included in national administrative datasets, the SIMD quintile or decile can be used as an indication of the levels of disadvantage associated with the location where an individual resides. Notably SIMD is not a direct measure of the level of deprivation experienced by an individual. Deprivation has been identified on various analyses as being associated with occurrence of infection (Ye et al., 2023). SIMD in deciles was used to maximise granularity.

3.4.5.4 Comorbidities

In Scotland, trained clinical coders review medical documentation at the end of each care episode. In accordance with national standards, codes are assigned to describe the clinical events which occurred during the episode and comorbid conditions. SMR01 also includes data items such as demographics, date of admission and date of discharge, among other items. For the VOICES study each participant in the dataset had a five-year SMR01 'look-back' period, such that comorbidities could be identified. This approach has been shown to be valid, including in the setting of infection (Hwang et al. 2016). Comorbidities were ascertained from SMR01 look-back period episode codes as Charlson comorbidity items. The Charlson Comorbidity Index (CCI) is a weighted index of 17 comorbid conditions (Charlson et al., 1987). Quan *et al* developed coding algorithms to map ICD-10 codes to Charlson comorbidity items (Quan et al. 2005). The R programming package 'comorbidity' incorporates such coding algorithms, allowing programmatic conversion of lists of ICD-10 codes for an entire cohort of patients into a CCI for each patient, as well as the presence of individual CCI disease components (Gasparini 2018). This enabled efficient determination of important comorbidities for all patients in the study. Comorbidities were ascertained at the time of index date, as the linked dataset produced was

initially designed for the exposures to be ascertained at this point for other studies in this thesis.

3.4.6 Outcome

The outcome of interest was occurrence of first severe infection, defined as an admission to hospital documented within SMR01 associated with a diagnostic code for infection. A common definition of severe infection is requirement for intravenous antimicrobial therapy, requirement for an invasive intervention or requirement for hospital admission (National Institutes of Health. National Cancer Institute. 2017). A proxy of this definition often utilised in the epidemiology literature is infection-related hospital admission (Barber et al. 2013). This definition of severe infection was used in this thesis. In an national administrative database this can be described by using a hospital care episode associated with an ICD-10 code for infection. This study, and studies contained in subsequent thesis chapters, utilised a published code list by Inada-Kim *et al*. This code list was designed to detect infection related conditions which most closely identify patients with 'suspicion of sepsis'. The term 'suspicion' is used as Inada-Kim *et al* highlight that sepsis guidelines advise, and clinicians aim, to prevent conditions that represent a severe infection deteriorating into true sepsis. Therefore the group of patients that this code list aims to identify are individuals with a bacterial infection whose severity merits hospital admission (Inada-Kim et al., 2017). This aligns with the outcome of interest for this study. Inada-Kim et al used the 'primary diagnosis' code to determine if the episode represented 'suspicion of sepsis'. We extended this use by applying the code list to both the 'main diagnosis' in SMR01 and 'other conditions'. Other conditions are defined by the Scottish Clinical Coding Standards as either coexisting conditions, often longstanding and not identified as a major factor in the hospital admission or new active problems which required investigation and management during the hospital stay (Public Health Scotland: Data and Intelligence, 2014). Given that 'suspicion of sepsis' conditions are rarely, if ever, a longstanding problem, 'other conditions' were considered to be active issues if the code was contained in the Inada-Kim list.

Individuals who died before the end of the study period or for whom there was less than 365 days follow-up were censored. Death was extracted from the

linked dataset derived from the National Records of Scotland (NRS) Central Register. As a measure to preserve pseudonymisation, date of death is recorded within PHS linked datasets as month and year only. For survival time calculation, '14' was simply imputed as the day of the month.

3.4.7 Bias

Unequal ascertainment of events due to competing risks is a theoretical issue in this study. The competing risk for severe infection in this study was death. However, the vast majority of individuals with AAV survive to the end of the first year following diagnosis. Of those who died approximately 50% died of infection – this should be ascertained in administrative datasets as an infection if there was an associated admission to hospital with correct dates. If present, this source of bias would tend to reduce the apparent risk of severe infection. It is estimated that this would have a small effect.

Medical surveillance bias, also known as detection bias or ascertainment bias, occurs where the outcome is more likely to be detected in one group compared to another, due to the intensity of medical involvement (Vandenbroucke et al. 2007). It is not considered likely to have significantly impacted this study. Individuals on higher glucocorticoid doses may be more likely to receive more medical input as the higher dose may be representative of more active disease. In the current study, it could be possible to account for this by quantifying the intensity of medical follow up and adjusting for this in the statistical model. However, while medical surveillance bias may lead to increased detection of minor infections, it was not deemed likely to have substantially contributed to increased hospital admissions related to infection.

Inflation bias, sometimes referred to as 'p-hacking', on the part of the study team was minimised in this study. This is a practice where the investigator, often unconsciously, adopts practices which lead to more 'positive' results. Such practices include conduction of multiple analyses and selecting the most favourable or stratifying the data in multiple ways and choosing the version of a dataset that is the most flattering (Head et al., 2015). In this study only prespecified and planned analyses of the data were undertaken. Two of these were not reported as it was decided retrospectively that the questions posed by

these models were less clinically and scientifically relevant, irrespective of statistical results. This substantially limited the possibility of multiple analyses unintentionally leading to spurious results.

Selection bias, where the probability of inclusion is associated either positively or negatively with the exposure, is unlikely to be an issue for this study. To be included in this study an AAV patient had to have a hospital admission associated with an AAV code documented in the SMR01 database. As a result, the population included in this study is likely comprised of individuals more comorbid, frail and potentially susceptible to complications of AAV compared to the target population, namely all AAV patients in Scotland. This could potentially bias the study in two ways. Firstly, increased frailty could interact with higher glucocorticoid dose in a multiplicative way, resulting in an exaggeration phenomenon whereby the effect of higher glucocorticoid has a greater magnitude of effect on severe infection risk than is true. Alternatively, non-frail patients with AAV may never be admitted to hospital, including never having a significant infection, despite being treated with glucocorticoids. The impact on magnitude of effect is difficult to quantify in this case.

3.4.8 Study size

The maximum number of patients eligible for the study who were available in the VOICES dataset determined the study size. A formal sample size was not calculated. Sample size was however factored into the design of the study in terms of limiting the number of parameters included in statistical models. Events per parameter (EPP) was considered. For most of the models the number of events was 111 and for those models the number of parameters ranged from 11 to 14. This resulted in an EPP of 7.9 to 10. For the 5-7.5 mg comparison, the number of events was 25-30 (showed as a range for data protection purposes) and the number of parameters was 11. This resulted in an EPP 2.3 to 2.7, given that this model is more at risk for overfitting it should be considered more exploratory than the others. While it may have been preferable to determine the sample size by factoring in the number of proposed parameters and the number of events experienced by the population, a larger dataset was not available. However, though careful selection of a limited number of covariates, avoiding an unnecessarily low EPP decreased the risk of overfitting. Traditionally a ‘rule of

thumb' of EPP equal to 10 was considered reasonable to avoid overfitting. Other sources have suggested an EPP of 20 or 50 as more appropriate. Ultimately, sample size calculations incorporating other factors are the most appropriate mechanism to determine sample size or recommended maximum number of parameters (Riley et al., 2020). Given the rarity of AAV and the difficulty of curating large observational datasets, a lower EPP was considered acceptable for this study, the results of which were interpreted as exploratory.

3.4.9 Statistical methods

Baseline characteristics were reported. This information per glucocorticoid exposure groups as described in section 3.4.4. To protect against identification of participants, where a cell contained a low value (below five), the value was redacted and represented as '<5'. Where the value of a low cell count could be inferred from other cells, values were suppressed to a range (e.g. '15–20' as opposed to '16'). This was required by Public Health Scotland for extracting data from the National Safe Haven. Continuous variables were reported as median and interquartile range. Missing data was minimal and is noted in each baseline characteristics table. A complete case analysis was performed for each model.

Four separate Cox proportional hazards models were developed. The predictors and outcome described above were included in the model. Individuals were censored as described above in section 3.4.6. These models can therefore be considered cause-specific models for severe infection with non-infection related death being the competing event. For each version of the exposure, the full model was used, without variable selection strategies. Fitting the full model is one of a group of recommended strategies for an informative prediction model. Alternative approaches include using data reduction methods and penalised estimation to reduce number of covariates in the model or the impact of covariates. The 'full model' approach was selected for several reasons: it takes into account all potential confounding variables to reveal the predictive ability of the exposure of interest, it does not erroneously remove variables which can occur when penalisation methods are applied to insufficiently large data sets, it is not necessary to reduce variables in these models as only the exposure is of interest and the approach is commonly accepted and understood (Harrell 2015). Hazard ratios, 95% confidence intervals and p-values were reported for the

exposures. Results were considered statistically significant if the associated p value was less than 0.05. Adjusted hazard ratio plots were shown for the exposure. Unadjusted Kaplan-Meier (KM) plots were also produced to show the progression of the outcome over time. KM plots included a p-value derived using the log rank test. Testing for assumptions in the Cox model included: testing for proportional hazards, testing for influential observations and testing for non-linearity. The proportional hazards assumption was tested using Schoenfeld residuals (not significant for each covariate or the global test) and visual examination of scaled Schoenfeld residuals against transformed time (no pattern with time). This was done in the model which included glucocorticoid as a continuous variable. Influential observations were tested by plotting "dfbeta" – the estimated change in regression coefficients upon deleting each observation in turn. This confirmed that no single observation was overly influential. Plotting Martingale residuals against age confirmed there was not obvious non-linearity. SIMD was not tested for non-linearity due to significant number of zero values. All statistical analyses were done within the Scottish National Safe Haven (NSH). The thesis author is grateful for the support of the eDRIS team (Public Health Scotland) for their assistance with acquiring approvals, providing data, performing data linkage and facilitating the use of the secure analytical platform within the NSH. All analyses were done using R version 4.2.0 with packages including comorbidity, finalfit, survival and the tidyverse packages.

3.5 Results

Figure 3-1 shows the derivation of the AAV glucocorticoid exposure cohort from the VOICES cohort. It also included patients index dates that predated the availability of prescription data from PIS (i.e. before April 2009).

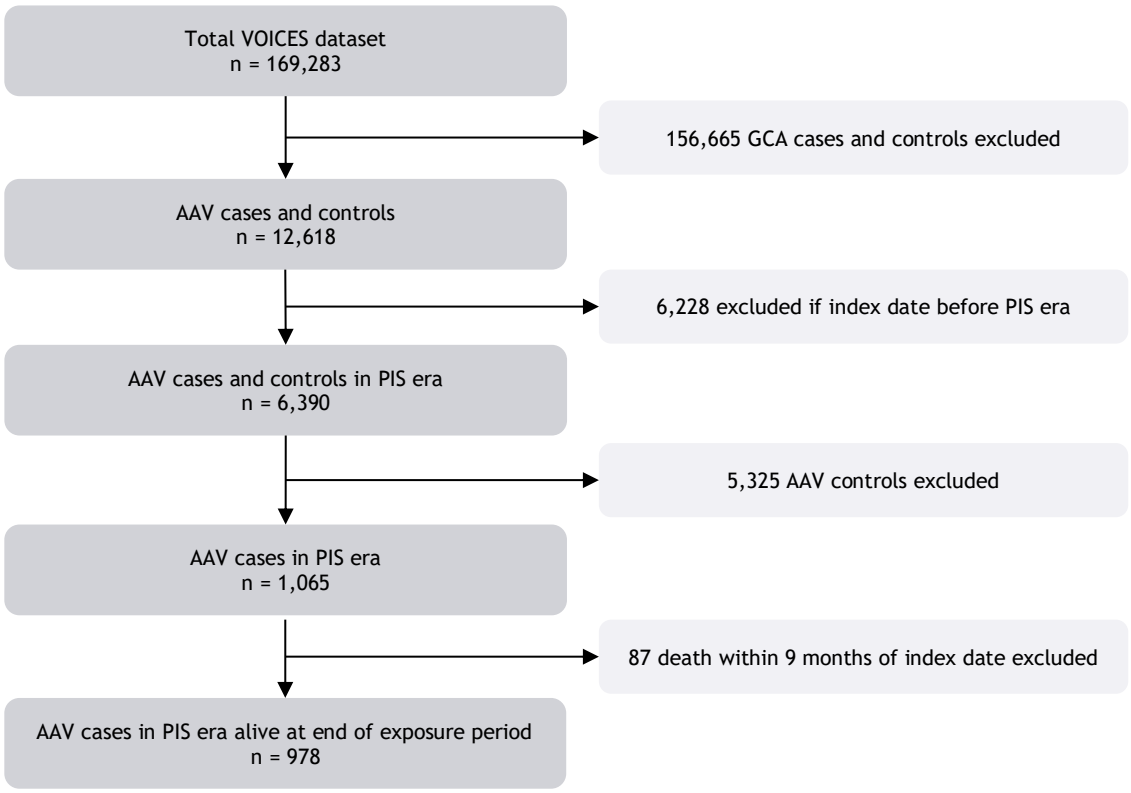


Figure 3-1 | Derivation of AAV glucocorticoid exposure cohort from VOICES cohort. The total VOICES dataset includes AAV cases and controls, as well as GCA cases and controls. Only AAV cases with sufficient prescription data and who were alive at the end of the exposure period were included.

3.5.1 Glucocorticoid dose: 0 mg vs >0-10 mg vs >10 mg

Table 3-2 shows the baseline characteristics for the comparison of those exposed to no glucocorticoids versus total daily prednisolone equivalent >0 mg to 10 mg versus >10 mg glucocorticoid per day. Table 9-1 in the appendices displays the full results of univariable and multivariable models. Glucocorticoid exposure equivalent to >0 mg to 10 mg prednisolone daily had a HR of 1.74 (95% CI 0.90 – 3.38, p = 0.102), while glucocorticoid exposure of >10 mg daily had HR 1.93 (95% CI 1.19 – 3.13, p = 0.008). This is displayed graphically in **Error! Reference source not found.** and **Error! Reference source not found.**.

Table 3-2 | Glucocorticoid 0 mg vs >0-10 mg vs >10 mg: baseline characteristics (see next page for legend)

	Missing N	Zero	>0 to 10 mg
Total N (%)		290 (29.7)	120 (12.3)
Age; median (IQR)	0	61.3 (46.5 to 71.3)	61.7 (48.8 to 72.2)
Female sex	0	166 (57.2)	61 (50.8)
SIMD (deciles); median (IQR)	6	5.0 (3.0 to 8.0)	5.5 (3.0 to 8.0)
Cardiovascular disease (all)	0	35-40 (12)	14 (11.7)
Atherosclerotic disease (all)	0	30-35 (11)	10-15 (9)
MI	0	5-10 (3)	8 (6.7)
CVD	0	20-25 (7)	<5
PVD	0	5-10 (2)	<5
Chronic heart failure	0	5-10 (2)	5-10 (6)
Hypertension	0	<5	<5
Diabetes	0	15-20 (6)	8 (6.7)
Renal disease	0	30-35 (11)	16 (13.3)
Dementia	0	<5	<5
Cancer (all)	0	15-20 (6)	<5
Cancer (localised)	0	16 (5.5)	<5
Cancer (metastatic)	0	<5	<5
Chronic respiratory disease	0	49 (16.9)	24 (20.0)
Rheumatic disease	0	20-25 (8)	5-10 (4)
Peptic ulcer disease	0	5 (1.7)	<5
Liver disease	0	5-10 (2)	<5
Previous infection	0	86 (29.7)	36 (30.0)
Charlson comorbidity index; median (IQR)	0	0.0 (0.0 to 1.0)	0.0 (0.0 to 1.0)
Severe infection (outcome)	0	23 (7.9)	10-15 (12)
Died	0	30-35 (11)	15-20 (15)
Follow-up (days)	0	365.0 (365.0 to 365.0)	365.0 (365.0 to 365.0)

Table 3-2 legend:
CVD = cerebrovascular disease, IQR = interquartile range, MI = myocardial infarction, PVD = peripheral vascular disease, SIMD = Scottish index of multiple deprivation

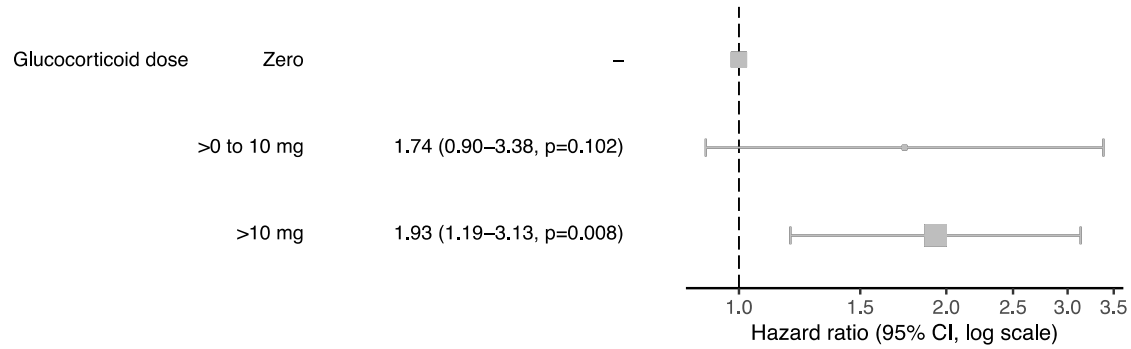


Figure 3-2 | Glucocorticoid 0 mg vs >0-10 mg vs >10 mg: HR plot. Adjusted hazard ratios for association with severe infection for glucocorticoid exposures. This is derived from a multivariable model including age, sex, SIMD, cancer, cerebrovascular disease, chronic heart failure, chronic respiratory disease, diabetes, liver disease and renal disease. See appendix for full model.

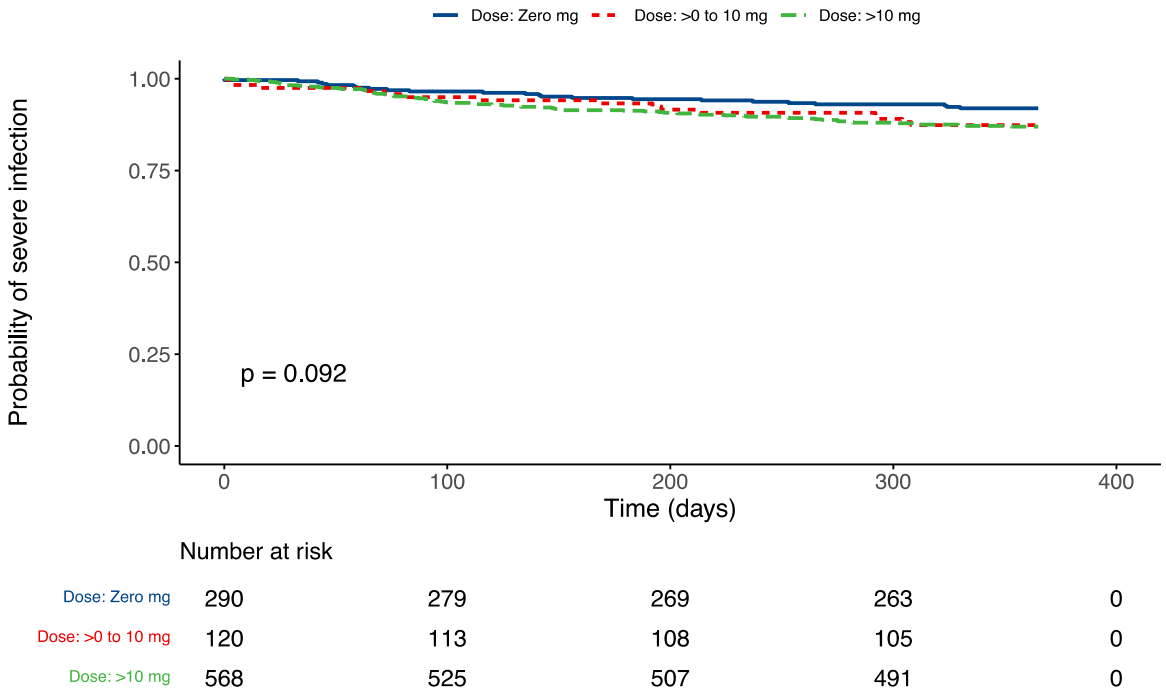


Figure 3-3 | Glucocorticoid 0 mg vs >0-10 mg vs >10 mg: KM plot. This Kaplan-Meier plot shows the probability of first severe infection over time for glucocorticoid exposure groups. A log-rank test p-value is displayed.

3.5.2 Glucocorticoid dose: multiple dose thresholds

Table 3-3i and Table 3-3ii show the baseline characteristics for the comparison of several groups based on multiple differing dose thresholds – the data is split across two tables for size purposes. Table 9-1 in the appendices shows the full univariable and multivariable models for severe infection within the first year following the index date for the glucocorticoid multiple dose model. Individuals with total daily glucocorticoid exposure of >0 mg to 5 mg prednisolone equivalent had a hazard ratio (HR) of 0.72 (95% CI 0.17 – 3.09, p = 0.659) for severe infection compared to the reference group who had no documented glucocorticoid exposure. Individuals with >5 to 10 mg prednisolone equivalent glucocorticoid exposure had HR 2.22 (95% CI 1.11 – 4.46, p = 0.025). The >10 to 20 mg group and the >20 mg daily prednisolone equivalent group had HR 1.73 (95% CI 0.97 – 3.09, p = 0.065) and HR 2.05 (95% CI 1.23 – 3.43, p = 0.015), respectively. This is displayed graphically in Figure 3-4 and Figure 3-5.

Table 3-3i | Glucocorticoid multiple doses: baseline characteristics (missing data, zero mg, >0 to 5 mg; see page 35 for table legend)

	Missing N	Zero	>0 to 5 mg
Total N (%)		290 (29.7)	38 (3.9)
Age; median (IQR)	0	61.3 (46.5 to 71.3)	61.0 (48.3 to 71
Female sex	0	166 (57.2)	16 (42.1)
SIMD (deciles); median (IQR)	6	5.0 (3.0 to 8.0)	7.0 (3.0 to 8.0)
Cardiovascular disease (all)	0	35-40 (12)	5 (13.2)
Atherosclerotic disease (all)	0	30-35 (11)	<5
MI	0	5-10 (3)	<5
CVD	0	20-25 (7)	<5
PVD	0	5-10 (2)	<5
Chronic heart failure	0	5-10 (2)	<5
Hypertension	0	<5	<5
Diabetes	0	15-20 (6)	<5
Renal disease	0	30-35 (11)	5 (13.2)
Dementia	0	<5	<5
Cancer (all)	0	15-20 (6)	<5
Cancer (localised)	0	16 (5.5)	<5
Cancer (metastatic)	0	<5	<5
Chronic respiratory disease	0	49 (16.9)	6 (15.8)
Rheumatic disease	0	20-25 (8)	<5
Peptic ulcer disease	0	5 (1.7)	<5
Liver disease	0	5-10 (2)	<5
Previous infection	0	86 (29.7)	14 (36.8)
Charlson comorbidity index; median (IQR)	0	0.0 (0.0 to 1.0)	0.0 (0.0 to 1.0)
Severe infection (outcome)	0	23 (7.9)	<5
Died	0	30-35 (11)	<5
Follow-up (days)	0	365.0 (365.0 to 365.0)	365.0 (365.0 to

Table 1-3ii | Glucocorticoid multiple doses: baseline characteristics (>5 to 10 mg, >10 to 20 mg, >20 mg; see page 35 for table legend)

	>5 to 10 mg	>10 to 20 mg	>2
Total N (%)	82 (8.4)	198 (20.2)	37
Age; median (IQR)	62.6 (49.0 to 72.4)	63.0 (54.5 to 71.9)	60
Female sex	45 (54.9)	107 (54.0)	17
SIMD (deciles); median (IQR)	5.0 (3.0 to 8.0)	6.0 (3.0 to 8.0)	6.
Cardiovascular disease (all)	9 (11.0)	21 (10.6)	47
Atherosclerotic disease (all)	7 (8.5)	18 (9.1)	39
MI	5-10 (6)	6 (3.0)	18
CVD	<5	9 (4.5)	15
PVD	<5	5 (2.5)	10
Chronic heart failure	5 (6.1)	5-10 (3)	12
Hypertension	<5	<5	<5
Diabetes	<5	15 (7.6)	23
Renal disease	11 (13.4)	21 (10.6)	25
Dementia	<5	<5	<5
Cancer (all)	<5	13 (6.6)	15
Cancer (localised)	<5	5-10 (4)	15
Cancer (metastatic)	<5	<5	<5
Chronic respiratory disease	18 (22.0)	40 (20.2)	67
Rheumatic disease	<5	11 (5.6)	12
Peptic ulcer disease	<5	5 (2.5)	6
Liver disease	<5	<5	6
Previous infection	22 (26.8)	56 (28.3)	10
Charlson comorbidity index; median (IQR)	0.0 (0.0 to 1.0)	0.0 (0.0 to 1.0)	0.
Severe infection (outcome)	13 (15.9)	25 (12.6)	45
Died	15 (18.3)	30 (15.2)	55
Follow-up (days)	365.0 (365.0 to 365.0)	365.0 (365.0 to 365.0)	36

Table 1-3i and 1-3ii legend:
CVD = cerebrovascular disease, IQR = interquartile range, MI = myocardial infarction, PVD = peripheral vascular disease, SIMD = Scottish index of multiple deprivation

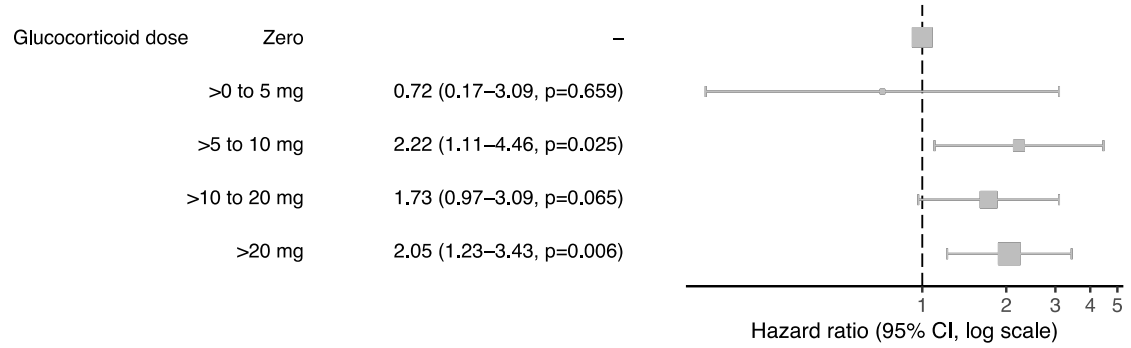


Figure 3-4 | Glucocorticoid multiple doses: HR plot. Adjusted hazard ratios for association with severe infection for glucocorticoid exposure groups divided by the zero mg, 5 mg, 10mg and 20 mg thresholds are displayed. This is derived from a multivariable model including age, sex, SIMD, cancer, cerebrovascular disease, chronic heart failure, chronic respiratory disease, diabetes, liver disease and renal disease. See appendix for full model.

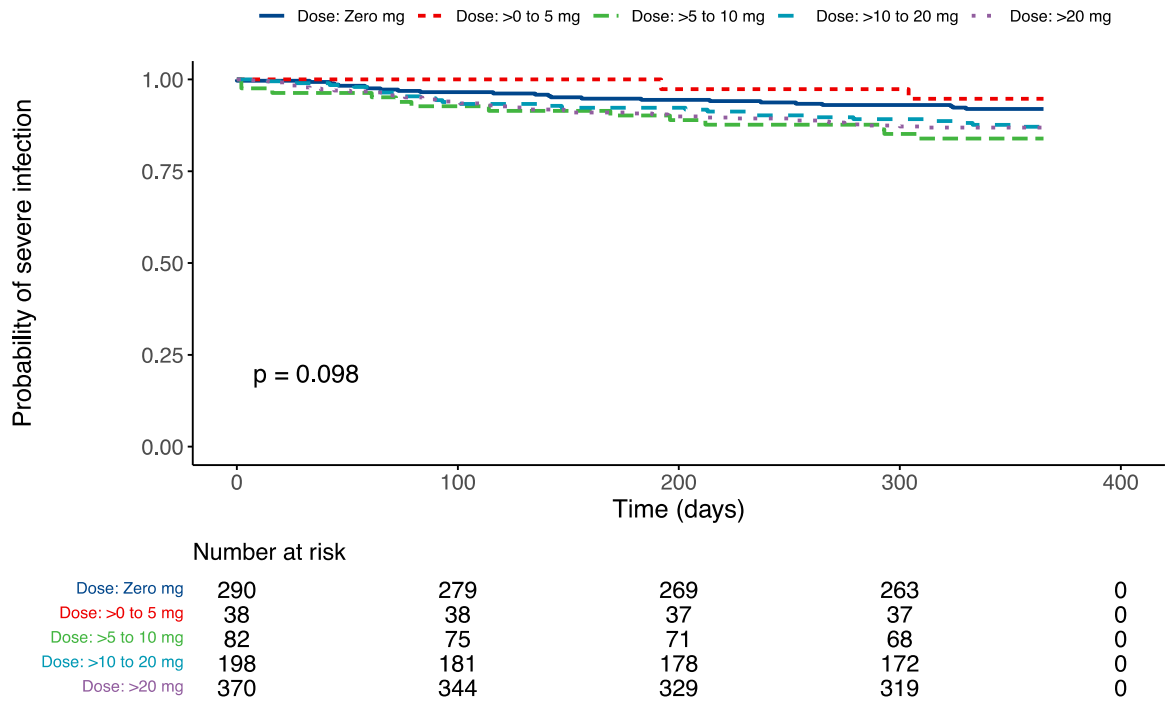


Figure 3-5 | Glucocorticoid multiple doses: KM plot. This Kaplan-Meier plot shows the probability of first severe infection over time for glucocorticoid exposure groups divided by the zero mg, 5 mg, 10 mg and 20 mg thresholds. A log-rank test p-value is displayed.

3.5.3 Glucocorticoid dose: 0 mg vs 5-7.5 mg

Table 3-4 shows baseline characteristics comparing individuals taking no glucocorticoid daily versus those taking daily prednisolone equivalents 5 mg to 7.5 mg daily. Table 9-2 in the appendix shows the full univariable and multivariable models. The 5 mg to 7.5 mg group had HR 2.55 (95% CI 0.72 – 8.94, p = 0.145) for association with severe infection. Risk associated with this glucocorticoid exposure is displayed graphically in Figures Figure 3-6 and Figure 3-7.

Table 3-4 | Glucocorticoid 0 mg vs 5-7.5 mg: baseline characteristics (see page 35 for table legend)

label	Missing N	Zero	5 to 7.5 mg
Total N (%)		290 (93.5)	20 (6.5)
Age; median (IQR)	0	61.3 (46.5 to 71.3)	64.2 (50.6 to 73
Female sex	0	166 (57.2)	14 (70.0)
SIMD (deciles); median (IQR)	<5	5.0 (3.0 to 8.0)	5.0 (2.8 to 8.0)
Cardiovascular disease (all)	0	35-40 (12)	<5
Atherosclerotic disease (all)	0	30-35 (11)	<5
MI	0	5-10 (3)	<5
CVD	0	20-25 (7)	<5
PVD	0	5-10 (2)	<5
Chronic heart failure	0	5-10 (2)	<5
Hypertension	0	<5	<5
Diabetes	0	15-20 (6)	<5
Renal disease	0	30-35 (11)	<5
Dementia	0	<5	<5
Cancer (all)	0	15-20 (6)	<5
Cancer (localised)	0	16 (5.5)	<5
Cancer (metastatic)	0	<5	<5
Chronic respiratory disease	0	49 (16.9)	6 (30.0)
Rheumatic disease	0	20-25 (8)	<5
Peptic ulcer disease	0	5 (1.7)	<5
Liver disease	0	5-10 (2)	<5
Previous infection	0	86 (29.7)	7 (35.0)
Charlson comorbidity index; median (IQR)	0	0.0 (0.0 to 1.0)	0.0 (0.0 to 1.0)
Severe infection (outcome)	0	23 (7.9)	<5
Died	0	30-35 (11)	<5
Follow-up (days)	0	365.0 (365.0 to 365.0)	365.0 (365.0 to

Table 3-4 legend
CVD = cerebrovascular disease, IQR = interquartile range, MI = myocardial infarction, PVD = peripheral vascular disease, SIMD = Scottish index of multiple deprivation

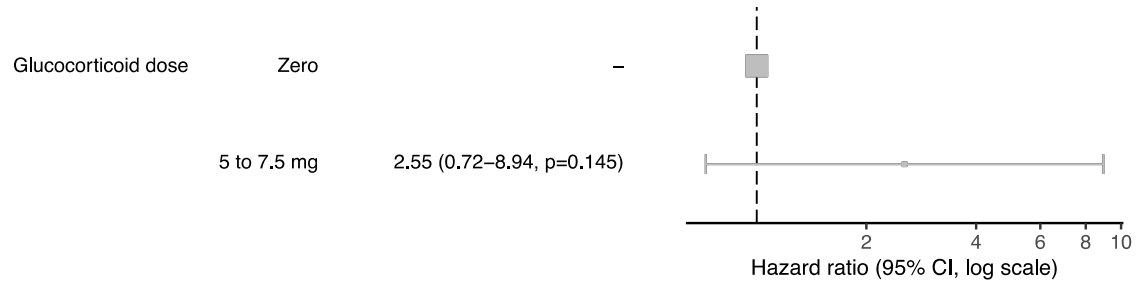


Figure 3-6 | Glucocorticoid 0 mg vs 5-7.5 mg: HR plot. The adjusted hazard ratio for association with severe infection for the 5 to 7.5 mg glucocorticoid exposure group is displayed. This is derived from a multivariable model including age, sex, SIMD, cancer, cerebrovascular disease, chronic heart failure, chronic respiratory disease, diabetes, liver disease and renal disease. See appendix for full model.

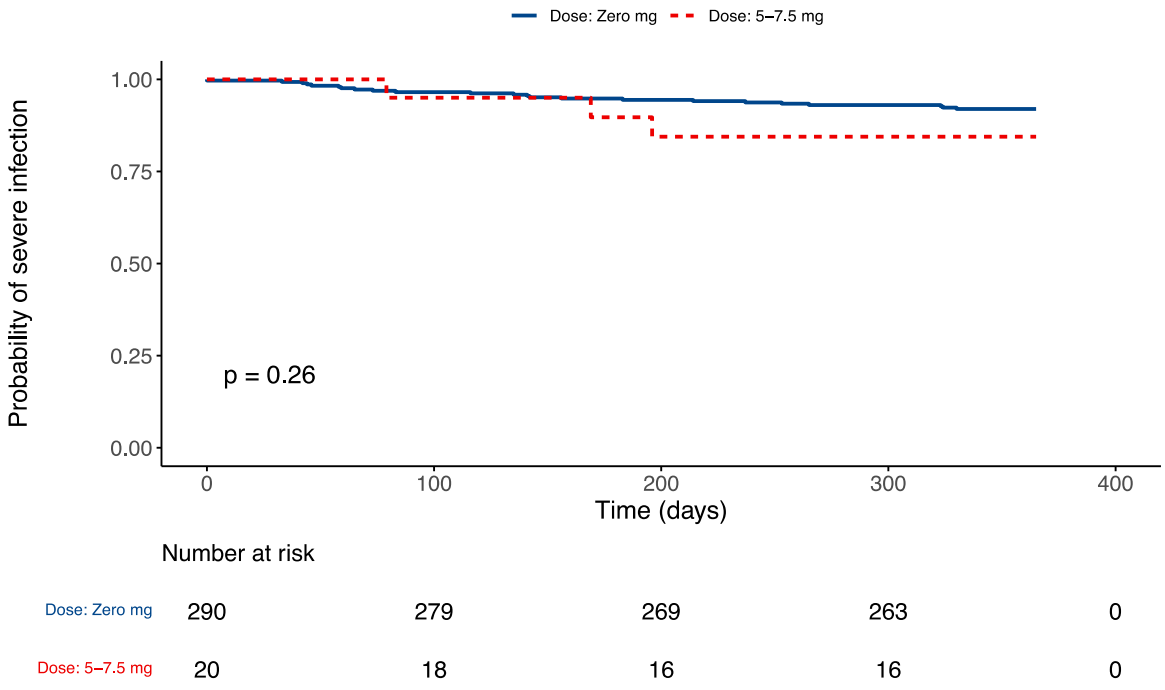


Figure 3-7 | Glucocorticoid 0 mg vs 5-7.5 mg: KM plot. This Kaplan-Meier plot shows the probability of first severe infection over time for the zero mg and the 5 to 7.5 mg glucocorticoid exposure groups. A log-rank test p-value is displayed.

3.5.4 Glucocorticoid dose: continuous

Table 9-4 in the appendix shows univariable and multivariable models incorporating glucocorticoid exposure as a continuous variable. Baseline characteristics for the components of the model are show in tables Table 3-2, Table 3-3i and Table 3-3ii. The multivariable model indicated that glucocorticoid dose in daily prednisolone equivalents had HR 1.01 (95% CI 1.00 – 1.02, p = 0.047), meaning that for every 1 mg increment in glucocorticoid dose there is a one percent increase in the hazard of severe infection over the subsequent year. This is displayed graphically in Figure 3-8.

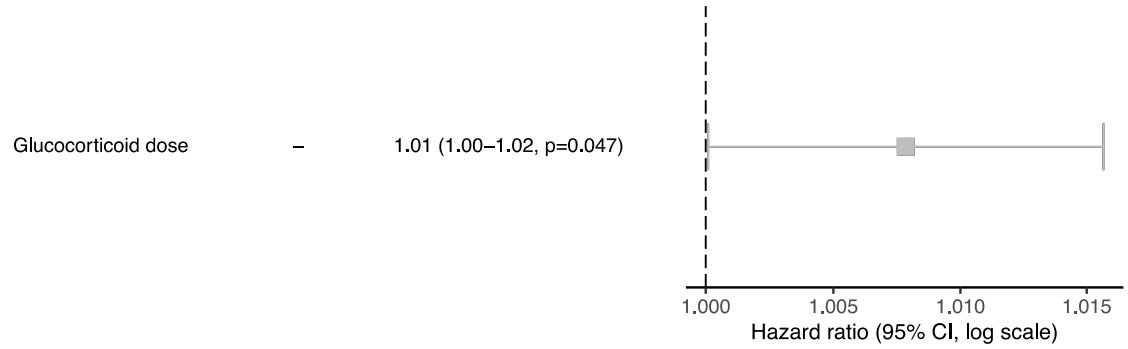


Figure 3-8 | Glucocorticoid as a continuous variable: HR plot. The adjusted hazard ratio for association with severe infection for every 1 mg increase in glucocorticoid exposure is displayed. This is derived from a multivariable model including age, sex, SIMD, cancer, cerebrovascular disease, chronic heart failure, chronic respiratory disease, diabetes, liver disease and renal disease. See appendix for full model.

3.6 Discussion

3.6.1 Key results

3.6.1.1 Glucocorticoid dose: 0 mg vs >0-10 mg vs >10 mg

Comparison of baseline characteristics of the exposure groups in this model did not reveal major differences in terms of demographics, comorbidities or overall CCI. An unadjusted log rank test comparing the exposure groups gave a p value = 0.092, but multivariable analysis showed there was a significant increased risk for severe infection for individuals taking greater than 10 mg glucocorticoid. Such individuals had a point estimate of 93% increased risk of suffering a severe infection over the course of one year (HR 1.93 (95% CI 1.19-3.13, p = 0.008)).

Due to a lack of power, it was not possible to make an estimate for individuals exposed to >0 to 10 mg glucocorticoid with a useful degree of precision. However, the point estimate and confidence intervals were compatible with the hypothesis that even lower doses of glucocorticoid confer increased risk for severe infection (HR 1.74 (95% CI 0.90-3.38, p = 0.102). This suggests that increased risk seen in other populations at similar dose thresholds also could occur in AAV (Walsh et al., 2020).

3.6.1.2 Glucocorticoid dose: multiple dose thresholds

Similarly, there were no substantial differences in terms of baseline characteristics across exposure groups in the multiple dose exposure groups. For the three exposure groups above 5 mg daily prednisolone equivalents, the point estimates for risk were relatively similar, suggesting that glucocorticoid at these dose ranges may predict a 73 to 122% increased risk of severe infection. The confidence intervals were wide however, therefore it was not possible to determine whether a clear dose-response relationship for glucocorticoids existed, though this remains possible. Point estimate risk for the >5 mg to 10 mg group was not lower than the higher exposure groups (HR 2.22 (95% CI 1.11 – 4.46, p = 0.025)). It may be the case that such low dose glucocorticoid predicts substantial increased risk. The degree of precision was low however, therefore the association may be less strong than these data suggest. Overall, these data support the case for glucocorticoid exposure to be considered as an important prognostic factor for severe infection in AAV. Where the aim is to develop predictive models, such a variable should be incorporated. While the main aim of this study was to determine prognostic associations with severe infection, it is possible that the association revealed may represent a causal effect. This is biologically plausible given the recognised immunosuppressant effect of glucocorticoids (Stahn and Buttgereit 2008). The >0 mg to 5 mg group was smaller than other groups and had a wide confidence interval that crossed the line of no effect (HR 0.72 (95% CI 0.17 – 3.09, p = 0.659)). The point estimate suggested a 28% lower risk of severe infection compared to no glucocorticoid. Due to the significant imprecision, all that can be stated given the current data is that the true relationship between different glucocorticoid dose groups and future severe infection remains unclear; both a linear dose-response relationship and a threshold effect relationship are possible given the current data. It does,

however, seem clear that high doses of glucocorticoid are associated with a substantial increase in severe infection risk.

3.6.1.3 Glucocorticoid dose: 0 mg vs 5-7.5 mg

As for the above glucocorticoid exposure groups, with respect to baseline characteristics, there were no major differences apparent between the 5-7.5 mg dose range group and the zero mg group. Like previous models, the confidence intervals were wide with HR 2.55 (95% CI 0.72 – 8.94, p = 0.145), which could confer a wide range of true associations. However, a true association for severe infection at this dose threshold seems plausible and would have implications for future prediction models. If this represented a causal effect, then there would be significant implications for patient management. Many patients are maintained on doses around 5 mg prednisolone for several months following the induction period of therapy in AAV, in line with international guidelines (Hellmich et al., 2023). Anecdotally many patients remain on such doses for prolonged periods. It may be that a 5 mg dose for the second half of the first year of AAV therapy confers more risk than has been recognised and that the benefit of preventing relapse and preserving organ function at this stage does not outweigh the risk of adverse events such as severe infection. In this scenario, there may be a role for a prediction tool: by factoring in other variables that contribute to risk of adverse events, a more individualised approach could be achieved. For an individual at low risk of severe infection, the benefit of ongoing low-dose glucocorticoid may outweigh the risks, whereas for a high-risk individual the converse may be true.

3.6.1.4 Glucocorticoid dose: continuous

This final model evaluated the exposure of glucocorticoid dose as a linear continuous variable. This showed that for every 1 mg dose increase in glucocorticoid there is an average associated one percent increase in the risk of severe infection over the subsequent year (HR 1.01 (95% CI 1.00 – 1.02, p = 0.047)). This observation provides further evidence for glucocorticoids being a prognostic factor for severe infection. Future models of this nature may benefit from non-linear modelling with cubic splines or multiple fractional polynomials. Such approaches would require a suitably powered dataset. This would inform

questions of whether increased glucocorticoid exposure is associated with the same increase in various patterns as described in section 3.3.4.

3.6.2 Strengths

Strengths and limitations of this study are assessed according to bias domains from the Quality in Prognosis Studies (QUIPS) Tool (Hayden et al., 2013), in addition to other important areas as identified by the study team.

3.6.2.1 Study participation

Given the real-world nature of the national administration health care data utilised in this study, study participation was not a significant source of bias – specifically selection bias. Aspects of study design may have contributed to an element of selection bias as participants were identified through hospital admissions data. This is not thought to have impacted the validity of the study and is discussed further in section 3.4.7. All theoretically eligible patients participated and the study sample was considered representative of a typical AAV population. Baseline characteristics were comprehensively described, though due to the nature of the data available, some potentially useful patient characteristics were not available for reporting or inclusion in the statistical models such as serum ANCA autoantibody positivity or serum immunoglobulins.

3.6.2.2 Study attrition

Attrition was not an issue due to study design. All data regarding hospital admissions subsequent to the index date and death data were captured at a national level. Only if events occurred outside Scotland would such data be missing. Such cases are likely to be minimal and would not affect the outcomes of this study.

3.6.2.3 Prognostic factor measurement

Glucocorticoid exposure, the prognostic factor (PF) of interest, was measured in a consistent manner for all study participants. Establishing medication exposure, including that of glucocorticoids, through large administrative sources of prescription data is an established methodology (Curtis et al. 2006; Grijalva et al. 2008; Sakellariou et al. 2022). Using a simple, intuitive formula it was

possible to determine mean glucocorticoid exposure in daily prednisolone equivalents over a three-month period. Cut points used for dichotomisation were designed to be clinically and/or biologically informative. Issues around the estimation of glucocorticoid exposure are described in section 3.4.4. Some PF studies measure multiple potential PFs which, where excessive, can lead to spurious results. Intentional or unintentional selective reporting due to publication bias can then lead to an over-representation of spurious PFs in the biomedical literature. Glucocorticoid exposure was the only prognostic factor measured and reported in this study, as a result selective reporting was not possible.

An important question for the design of this study was: to what extent does the duration of glucocorticoid exposure contribute to severe infection risk. There was no previously established ideal time window defined in the AAV literature with respect to this question. Methodology in other studies in AAV patients was reviewed, as described in section 3.3.3. As a suitable definition could not be obtained from the AAV literature, literature from RA, an adjacent IMID, was used to inform this consideration.

3.6.2.4 Outcome measurement

This study used a reliable and valid means of identifying the outcome. A clear, appropriate definition of severe infection was used. The outcome was measured in the same way for all individuals in the study. Differential measurement of outcome in different study exposure groups is unlikely to have introduced significant bias in the current study: there does not seem to be a plausible reason why individuals with differing glucocorticoid exposure should be more or less likely for the severe infection outcome to be documented as having occurred. Similarly to the exposure, severe infection was the only outcome measured for this study, thus avoiding selective reporting.

3.6.2.5 Statistical analysis and reporting

Statistical models used were appropriate for time-to-event data. The model building strategy took the approach of using the ‘full model’ as opposed to automated variable selection. Automated variable selection can be useful to make a model more simple for clinical application, but can result in an a less

accurate model (Harrell 2015). Only six statistical models were developed, four are presented and the decision for presentation was made based on judgement regarding the models' clinical and research relevance as opposed to model output. Each model was presented for a clear purpose. Various dose thresholds were evaluated to determine if 1) glucocorticoid exposure at different levels had comparable effects to other IMID populations, such as RA (zero mg versus >0-10 mg versus >10 mg); 2) to assess for a dose-response (multiple dose model); 3) to evaluate a common clinical scenario of prolonged low-dose glucocorticoid (zero versus 5-7.5 mg); and 4) a model maximising power by using glucocorticoid dose as a continuous variable, in order to determine the effect per mg and also the overall biological impact of this exposure.

3.6.2.6 Size

This study was one of the largest studies in AAV evaluating the relationship between glucocorticoids and severe infection. Other studies with a similar aim are described in detail in the introduction, Chapter 1. The large majority of these studies are less than half the size of the current study and have comparatively insufficient power to detect important relationships whilst minimising the likelihood of detecting spurious associations.

3.6.3 Limitations

3.6.3.1 Prognostic factor measurement

Whilst this study has considerable strengths with respect to prognostic factor measurement, there are some limitations. It seems a reasonable assumption that the calculated 90-day average glucocorticoid exposure according to the PIS prescribing dataset should approximate to the glucocorticoid exposure of an individual with AAV. The time window of assessment was specifically chosen to maximise likely reliability of the measure, as well as to limit confounding (see below): six to nine months following the index date is a time window when most AAV patients are on a stable dose of glucocorticoid. At this time glucocorticoid should be supplied from a community pharmacy and therefore should appear within the PIS dataset. Severe infections occur most frequently within the first year following AAV diagnosis. Therefore the follow-up period was not 'too late' in the natural history of AAV such that the frequency of the outcome would be

low. As a result, statistical power was maintained and the study evaluated a time-period that is highly clinically relevant for AAV patients. There may, however, be mechanisms by which inaccuracy of the exposure measurement may arise. AAV patients may be exposed to significant glucocorticoid out with that supplied by a community pharmacy. They may receive glucocorticoids from a hospital pharmacy, either as an inpatient or an outpatient. This may be more common if they were to suffer a relapse during the observation period. AAV patients may also keep a 'backup' supply of glucocorticoids in case they suffer a relapse and need immediate access to drug. Therefore, some patients may have been exposed to glucocorticoid, even if this was not evident based on their community prescriptions. Some patients may have been advised by their treating specialist to take a different dose of steroid over the course of the exposure window than what is represented by community prescriptions.

Potential issues may be revealed with the 5-7.5 mg dose exposure group. This was selected as a common dose that many AAV patients would typically be receiving at the chosen exposure time-window. According to international guidance most would be expected to be at or approaching 5 mg at six months post index (Hellmich et al., 2023). However only 120 (12.3%) patients in the overall cohort had glucocorticoid exposure of >0 to 10 mg daily prednisolone equivalents – the dose range most likely to contain individuals receiving 5 mg exposure, if the glucocorticoid exposure is correct as per the methodology of this study. Also more patients that would be expected at this time point appeared to be receiving zero glucocorticoid. Three reasons can potentially account for these issues. Firstly, guidelines aspire to the ideal glucocorticoid weaning pattern and the reality of clinical practice may result in this being achieved in fewer cases than expected. A poor response to treatment may result in a slower glucocorticoid taper. Secondly, the study index date may not represent the date of diagnosis. As the index date is derived from the first SMR01 episode associated with an AAV code, patients diagnosed as outpatients may have had established AAV for many years at the 'index date'. At the time of being entered into the current study they may have been admitted for a relapse or a non-vasculitis reason, but with a code for AAV representing this as a comorbidity. This may account for many individuals in the study not being on glucocorticoids according to PIS. These first two reasons should not have a

significant impact on the results of this study or the implications of the results for AAV patients: it was not an essential part of study design that the index date should represent the date of diagnosis. The third possible explanation is that the individuals in the 5-7.5 mg glucocorticoid exposure group are on a different dose for the some of the reasons described in the paragraph above.

The comparison of 5-7.5 mg to no glucocorticoids was not significant. In retrospect there would have been a rationale for extending the range downwards – perhaps to 4 mg. This may have improved the power of this model to detect an effect for this low-dose exposure.

3.6.3.2 Outcome measurement

The code list for severe infections, derived from a published algorithm for acute admissions to hospital, was a list of codes likely representative of conditions where there is the potential for sepsis to develop (Inada-Kim et al., 2017). The authors had a comprehensive methodology for identifying these codes. While this represents a systematic approach to using administrative data to identify severe infections, it was not possible in our study to confirm how accurate these codes were. This would require a nested case note review validation study. Such work has previously been carried out by a colleague from the same research group for various comorbidities, but not for severe infection.

We included not only the 'main condition' from hospital admissions episodes, but also 'other conditions' which can represent comorbid conditions or other current issues requiring management during the admission. We estimated that most infections listed as other conditions would likely be severe. Arguably this may result in some non-severe infections being including as outcomes, but it was considered that not including 'other conditions' for identifying infections would lead to greater inaccuracy, as this is the only way that hospital-acquired infections, which are often very severe, could be included in the analysis.

3.6.3.3 Confounding

Confounding was well addressed in this study, with variables in the models including age, sex, deprivation and multiple comorbid conditions. Confounding variables of interest, which may have value as potential prognostic factors

included age, chronic respiratory disease, renal disease and cardiovascular disease. These variables all have biological plausibility potentially supporting an aetiological role in relation to severe infection. The have also been identified as having predictive value in other studies of prognostic factors for infection in AAV, which are detailed in Chapter 1. Although important variables were included as potential confounders, the possibility of residual confounding remains. Two important potential sources of residual confounding include disease severity and frailty.

Confounding by disease severity may occur where individuals with worse disease are more likely to receive higher doses of glucocorticoid but are also more likely to experience severe infection due to severe vasculitis independent of glucocorticoid dose. At the six-to-nine month time point, higher dose glucocorticoid may represent ongoing disease activity or relapse. The increased risk of infection due to disease severity is incompletely understood but is likely related to the altered immunity of autoimmune disease and vasculitis damage disrupting aspects of innate immunity, such as mechanical barriers to pathogens in the respiratory tract. Receiving other immunosuppressive therapies at this time, such as cyclophosphamide and / or rituximab may contribute to increased infection risk. Frailty may also have contributed to residual confounding as a form of ‘confounding by contraindication’. This arises where individuals who are considered too frail for certain therapies, such as cyclophosphamide or rituximab, are instead treated, arguably inappropriately, with more aggressive doses of glucocorticoid. In this scenario, frailty is a confounder as it contributes both to the likelihood of receiving higher doses of glucocorticoid and the likelihood of the occurrence of severe infection. Therefore, glucocorticoids may be partially or fully confounded in this regard, making them a marker of infection risk as opposed to a causal factor. This is not necessarily an issue for predictive modelling, as a variable need not be causative. However, one can envisage issues in the future where glucocorticoid regimens may be replaced by non-glucocorticoid based regimens. If a prediction model were applied to such a population, not being on glucocorticoid may falsely be seen as a protective factor against severe infection. It was not possible to incorporate these potential confounders with the current data set, therefore future studies should consider this as part of study design.

Comorbidities for this study were ascertained based on data prior to the index date. This was because the data set utilised for the current study was derived from the cleaned data sets for the study in Chapter 4. Some patients may have acquired additional comorbidities during the six-month period after the index date. Basing the presence or absence of comorbidities on data that was possibly less 'up-to-date' may have introduced some noise into the study, making the results more difficult to interpret. A future iteration of this study should identify confounding variables at the time of the exposure period.

An important set of confounding variables not accounted for in this study were concomitant immunosuppressive therapies such as cyclophosphamide and rituximab. These medications are supplied by hospital pharmacies and therefore are not apparent in the community prescription database, PIS, that was used for this study. The study exposure period of six-to-nine months following the index date was partly selected to account for the lack of availability of this important set of confounding variables. Given that the index period will likely represent the approximate date of diagnosis for many patients, by the time of the exposure period, most individuals will not be receiving induction therapies, thus reducing the effect that these variables have on the outcome.

3.6.3.4 Selection bias

Participants for this study were identified via SMR01 – a routinely collected national admissions database. Therefore our study would not have identified patients with AAV who were not admitted to hospital during the VOICES study period. Individuals who were not admitted to hospital are likely to be younger, less frail and less sick with vasculitis than those admitted to hospital. Therefore the cohort identified for this study is likely sicker and frailer than the target population. If glucocorticoid use has a multiplicative relationship with frailty, then this selection bias may result in an exaggerated association between glucocorticoids and severe infection. This potential effect is difficult to quantify, but it is not thought to have substantially impacted the outcome of the study. It is also important to acknowledge the means of case ascertainment in this dataset. ICD-10 codes for AAV sub-diagnoses were sought in national, routinely-collected, healthcare administrative data. As discussed elsewhere in this thesis, each hospital admissions has different diagnoses coded as being

associated with the care episode. This includes the current main reason for hospital admission ("main condition") and up to five "other conditions" which may be other active issues during the hospital stay or pre-existing comorbid conditions. If AAV was an active issue during the care episode, then it seems highly likely that it would be coded in the database, though it may be challenging for clinical coders to identify AAV conditions, where variable nomenclature is used, for example eponymous names for GPA and EGPA. Where AAV is a pre-existing comorbid condition and the individual has multiple comorbidities, then AAV may not be coded. This may be exacerbated by clinical coding guidelines that described "higher priority" conditions such as cancer (Public Health Scotland: Data and Intelligence, 2014). Ultimately this may not be an issue that introduces significant systemic bias to the data set, presuming the individuals not identified due to lack of an appropriate AAV code are not systematically different to those who are identified. While the issues discussed may contribute to worse sensitivity, it is also necessary to consider specificity – in this setting this would be an individual being coded as having AAV in the national data, where in reality they do not have AAV. This may occur when an individual presents with a vasculitis mimic condition, that is initially thought to be AAV, but transpires to be another condition. Anecdotally, this clinical scenario generally seems uncommon, so is unlikely to have caused significant issues with case ascertainment in this thesis. Ultimately, the presence and impact of these theoretical issues with clinical coding of AAV in routinely-collected data are difficult to quantify without a nested data quality study, which was not possible for this thesis.

3.6.3.5 Statistical power

This study would have been more informative if estimates of precision were more accurate due to more statistical power. However, these data are still broadly compatible with the hypothesis that low doses of glucocorticoid are associated with subsequent severe infection. In the multiple dose threshold model, the >0 to 5 mg dose bracket had a point estimate suggesting lower risk than no steroids. If the point estimate were correct, this observation would not be compatible with the above-described hypothesis. However, as the confidence intervals for this comparison were wide (HR 0.72 (95% CI 0.17 – 3.09, p = 0.659)), a range of effects remains possible. Similarly for the multiple dose model,

although higher doses (i.e. 5 mg and above) generally appeared to be associated with increased risk, it was not possible to discern a pattern of changing risk with increased glucocorticoid dose due to the imprecision of estimates. Although the study was underpowered to demonstrate this, such a progression of risk remains possible.

3.6.3.6 Continuous glucocorticoid modelling

There are few completely linear phenomena in nature, therefore there is a strong case for exploring non-linear relationships between variables. For the continuous model which considered glucocorticoid dose as a linear variable, a non-linear transformation of the data as described in section 3.4.4.4 may have revealed interesting phenomena such as whether the magnitude of change in risk increases or decreases with increasing dose, or whether an inflection point in glucocorticoid dosing exists where risk sharply increases or, conversely, levels off. Non-linear exploration of dosing would require a larger dataset for increased power.

3.6.4 Interpretation

These studies demonstrate that glucocorticoids are strongly associated with severe infection in AAV patients. This effect was present across various dose thresholds examined including >5 to 10 mg, >10 mg and >20 mg. The >10 to 20 mg group did not have a statistically significant hazard ratio (HR 1.73 (95% CI 0.97-3.09, p = 0.065)), but given that the large majority of the confidence interval was located 'above 1' and that dose groups above and below were associated with severe infection, it seems plausible this effect is real. The >0 to 5 mg group had a wide confidence interval and the point estimate did not seem biologically plausible, therefore this exposure group was difficult to interpret without greater statistical power.

With respect to the 'multiple doses model', it was reasonable to have expected a dose response relationship whereby the risk of severe infection increased as the exposure to glucocorticoid increases. Possibly due to a lack of power, such a relationship was not clearly demonstrated.

Again underpowered, there is significant imprecision with respect to the HR assigned to the 5-7.5 mg dose group. However, most of the confidence interval sits above the line of null effect, suggesting a possibility that even this lower dose of glucocorticoid could be associated with substantially increased risk of severe infection. Should this be the case, and should there be further evidence that this effect is causal, this may have important clinical management implications for individuals with AAV. Similarly, the continuous variable model suggests that small differences in glucocorticoid dosing were associated with clinically important differences in severe infection rates, with a 1 mg higher glucocorticoid dose being associated with a one percent increase in severe infection risk. Ultimately it is clear that glucocorticoids were associated with increased infection risk at various levels of dosing, meaning that this variable should actively be considered for inclusion in risk prediction models relating to severe infection.

As discussed in detail in the introduction, section 1.13.3.3, a link between glucocorticoid exposure and subsequent severe infection has been apparent for several decades. This was particularly prominent in an early French RCT where glucocorticoid exposure in both treatment arms was higher than other regimens at the time and is considerably higher than modern regimens. At 3 months in this trial the average daily prednisone exposure was 55 mg daily. Over mean follow-up of approximately 28 months, 55% of participants had experienced an infection. The incidence of PCP was high at 20%, it was not stated whether antimicrobial prophylaxis was used. An important difference in this study population was disease severity – some participants in the trial were recruited from critical care, and 70% had renal involvement – both factors that would have increased propensity to infection (Guillevin et al., 1997). In the modern era, PEXIVAS provided compelling evidence that reduced glucocorticoid exposure was associated with fewer severe infections, as detailed above, with incident rate ratio 0.69 (Walsh et al., 2020). The smaller LoVAS RCT, performed in Japan, was consistent with this (Furuta et al., 2021). Apart from in the current study, lower doses in the maintenance phase have not been evaluated in AAV. Low dose glucocorticoid exposure has been explored in rheumatoid arthritis (RA), an adjacent IMID. George and colleagues found that the one-year cumulative incidence of severe infection was higher in those who received glucocorticoid at

less than 5 mg daily prednisolone equivalents, compared to those who were not treated with glucocorticoids (George et al., 2020).

3.6.5 Generalisability

As this study utilised routinely collected, real world, healthcare administrative data, its results are generalisable. Coverage of the Scottish population is effectively complete. The only feature of this study's design which may lead to lower generalisability is the selection bias described above in section 3.6.3.4, whereby individuals were included in the study if they had an AAV code associated with a hospital care episode. Therefore, this data may not be generalisable to individuals who have never been admitted to hospital.

3.7 Summary

In this chapter, through utilising one of the largest known observational datasets of individuals with AAV, several glucocorticoid exposures were evaluated in cause-specific Cox models for association with severe infection. Particularly at higher doses, it was clear that glucocorticoids were associated with severe infection. This supports findings in the medical literature indicating that glucocorticoid exposure should be considered a prognostic factor for severe infection in IMID. This supports the inclusion of glucocorticoids as a prognostic factor in multivariable models seeking to predict the occurrence of severe infection. In the next two chapters, we will seek to build on the contribution made to the prognosis research literature in AAV by developing prognostic models relating to severe infection in AAV.

4 Development and internal validation of a multivariable model to predict first severe infection in individuals with ANCA-associated vasculitis (AAV)

4.1 Overview

In the previous chapter, a series of prognostic factors based on glucocorticoid exposure were evaluated for their ability to predict severe infection in AAV patients. Prognostic factor research is crucial in any clinical domain, but the predictive characteristics of a single prognostic factor are typically lower than when combined with other factors in a prognostic model. The next two chapters will describe two such models, developed and internally validated using modern predictive statistical techniques. This chapter will build a model to predict severe infection events, while the model in chapter 5 will aim to predict early mortality after a diagnosis with severe infection. Following external validation and evaluation of clinical impact, such models could be used as both clinical and research tools to identify those at the highest risk severe infection and associated mortality. Such individuals could then participate in randomised controlled trials of novel strategies and therapies, leading to a personalised medicine approach to targeting a severe complication in this vulnerable population.

4.2 Abstract

4.2.1 Background

Severe infection is the most common cause of early mortality and overall excess mortality in AAV patients. Prediction of severe infection events would be of high utility to vasculitis clinicians and researchers. No prognostic models that aim to predict severe infection over the first year following AAV diagnosis have yet been developed. This chapter aims to develop and internally validate such a model.

4.2.2 Methods

A national Scottish dataset was developed using data linkage and routinely collected data. There was full coverage of the Scottish population. The study population was AAV patients identified through listed comorbidities associated with a hospital admission. Candidate predictors included demographics and comorbidities including a previous hospital admission with infection. The study outcome was occurrence of the first severe infection within one year, defined as a hospital admission associated with infection. A cause-specific Cox proportional hazards model was developed. The model incorporated a non-linear transformation of age, derived using fractional polynomials. Bootstrapping was used to determine optimism-adjusted performance measures. Pseudo-observations accounted for censoring when assessing model calibration.

4.2.3 Results

2,078 individuals were included in the dataset, 428 (20.6%) of whom experienced the severe infection outcome. The competing outcome of death occurred in 138 (6.6%) individuals. The final model included age (transformed as $(age / 100)^3$), Scottish Index of Multiple Deprivation (SIMD), renal disease and diabetes. On internal validation, optimism-adjusted model performance statistics included calibration slope 0.94, calibration intercept -0.01, calibration in the large 1.01 and concordance statistic 0.60. The scaled Brier score was 2.6%.

4.2.4 Conclusions

This prognostic model is an important initial step in predicting severe infection events in individuals with AAV.

4.3 Introduction

4.3.1 Background to this study

Individuals with ANCA-associated vasculitis (AAV) are at high-risk of severe complications including infection, malignancy and cardiovascular disease. The causes of these are multifactorial and can broadly be divided into disease-related and therapy-related factors. The potent immunosuppressive therapy required to induce remission of active disease has a marked contribution to infection risk. It is well recognised that infections have a considerable impact on patients with AAV. Severe infections are responsible for substantial morbidity and are recognised as the most common cause of early mortality (Little et al., 2010). Currently, clinicians and researchers do not have reliable means of predicting whether an individual with AAV will go on to suffer a complication such as severe infection. Such a tool could rely on a single biomarker, but it is more likely that a useful tool would incorporate multiple factors that are predictive of infection events. This likely related to the underlying multifactorial aetiology of clinical outcomes due to a complex interaction between biology and environment, and is evidenced by the multiple variables included in sophisticated prognostic modelling tools, such as those used in clinical practice (Wishart et al., 2010; Hippisley-Cox et al., 2017). This study will seek to develop such a predictive tool using national, routinely collected administrative data and modern clinical prediction model development methodology.

4.3.2 Potential impact

Predicting infections is an unmet clinical and research need in AAV patients. Offering clinicians and patients information regarding the likelihood of severe infection could assist with decisions around the timing and intensity of immunosuppression. Antimicrobial prophylaxis is widely used during the early stages of treatment of AAV. While this reduces risk of specific infections such as pneumocystis pneumonia, many individuals remain susceptible to other

infections. A predictive model could assist clinical researchers with selection of an at-risk clinical population for basic science and clinical studies. If patients who are at high risk for infection could be reliably identified then clinical trials could be designed which focus on this vulnerable group, either by allowing stratification by an *a priori* subgroup of interest or by requiring high infection risk, above a specific threshold, for inclusion. Through such an approach, individuals most likely to benefit from preventative therapy could be identified.

4.3.3 What is currently known

Prediction of infection is recognised as challenging in AAV. There are limited data available to assist with risk stratification for infection (Zeng et al. 2022). Many observation studies have been reported which describe predictive factors for severe infection in AAV. These are reviewed in detail in chapter 2. Most of these reports are retrospective. Many were underpowered to detect the true size of association of variables with outcome, given the number of variables relative to events under study. Very few studies had a stated objective of deriving a predictive model that would be testable in other populations and could theoretically be utilised in a clinical setting. They were typically underpowered and focused on a specific population not generalisable to most AAV patients. (Zhang et al., 2022; McClure et al., 2021)

4.3.4 Aim

The aim of this study was to develop a practical clinical prediction model on a real-world AAV population using routinely collected national administrative data, derived through a data linkage approach and using modern predictive analytical methods. The model sought to predict occurrence of first severe infection, starting from participant index date.

4.4 Methods

4.4.1 Study design and setting

Reporting followed the TRIPOD (Transparent Reporting of a multivariable prediction model for Individual Prognosis Or Diagnosis) guidelines (Collins et al. 2015). Vasculitis Outcomes in relation to Care Experiences (VOICES) is a services

mapping study of vasculitis care and outcomes with projects including both Scotland and the whole of the United Kingdom. Additional information regarding the study is available online (VOICES, University of Aberdeen 2022). VOICES includes a Scottish data linkage matched cohort study aiming to examine patterns of vasculitis care and outcomes. This dataset was utilised in the current study. A detailed description of the dataset is available in Chapter 3. The dataset identified all patients in Scotland with a relevant International Statistical Classification of Diseases and Related Health Problems 10th Revision (ICD-10) code for AAV and Giant Cell Arteritis (CGA) from the Scottish Morbidity Record admissions database (SMR01). SMR01 captures hospital admission data from all non-obstetric and non-psychiatric inpatient and day case care episodes. Ten controls were identified via SMR01, matched by age, sex and health board of residence. Patient identifiers were used to link to administrative health care data from multiple national datasets. These datasets are described in detail in chapter 3. An index date representing probable AAV diagnosis was defined as the earliest admission date associated with an AAV ICD-10 code. A look-back period of 5 years allowed comorbidities that occurred prior to the index date to be determined, according to established methodology (Quan et al. 2005). Data linkage was performed by PHS using a robust approach that has been demonstrated to result in highly accurate and complete data (Evans and MacDonald 1999; Scottish Public Health Observatory 2022).

4.4.2 Participants

The study period was defined from 1st April 1996, when ICD-10 was introduced, to the latest available record at the time of linkage, 31st October 2020. GCA patients and all matched controls were removed from the dataset to derive an AAV retrospective cohort. This cohort comprised all adults (aged 16 years or older) with an AAV code within SMR01. The following ICD-10 codes were used to identify patients: M31.1 (granulomatosis with polyangiitis), M31.7 (microscopic polyangiitis) and M30.1 (eosinophilic granulomatosis with polyangiitis). Patients were followed up from index date to death or the end of the study period, whichever occurred first. Baseline characteristics were reported including age, sex, SIMD, comorbidities, previous infection and Charlson comorbidity index for those who experienced the primary event, those who experienced the competing event, those did not experience either event and the total study population. To

protect against identification of participants, where a cell contained a low value (below five), the value was suppressed and represented as '<5'. Where the value of a low cell count could be inferred from other cells, values were suppressed to a range (e.g. '15–20' as opposed to '16'). Continuous variables were reported as median and interquartile range. P-values were derived using Kruskal Wallis test for continuous variables and chi-squared test for categorical variables.

4.4.3 Outcome

The outcome of interest was occurrence of first severe infection, defined as an admission to hospital documented within SMR01 associated with a diagnostic code for infection. Death was incorporated in the analysis as a competing event. Death was extracted from the linked dataset derived from the National Records of Scotland (NRS) Central Register. As a measure to preserve pseudonymisation, date of death is recorded within PHS linked datasets as month and year only. For survival time calculation, '14' was simply imputed as the day of the month. Where this resulted in a negative censor date, such individuals were removed from the dataset.

4.4.4 Independent predictor variables

Candidate predictors were selected based on published prognostic literature relating to infections in AAV and other autoimmune rheumatic disease, and clinical judgment of the study team. (Fine et al., 1997; Shapiro et al., 2003; Hespanhol and Bárbara, 2020; Bahlis et al., 2021) The selection of these predictors was carried out by the thesis author and presented to thesis supervisors, both academic clinicians, one nephrologist and one rheumatologist. One single demonstrator model was developed for illustrative purposes. Suggestions for inclusion of additional variables or exclusion of existing variables were incorporated. Following discussion there was no disagreement on variable selection but had this occurred it would have been resolved through further review of the published literature and consensus. Except for the single demonstrator model, variable selection was completed before the statistical modelling process began. Selected variables included age, sex, Scottish index of multiple deprivation in deciles (SIMD; as continuous variable), previous admission with infection and comorbidities including cancer (localised or metastatic),

cerebrovascular disease, chronic heart failure, chronic respiratory disease, diabetes, liver disease and renal disease. SIMD is a rank assigned to geographical 'data zones' and is derived from multiple indicators (Scottish Government, 2020).

To identify comorbidities, each participant in the dataset had a five year 'look back' period of SMR01 hospital admission data. Comorbidities were derived from SMR01 episode codes as Charlson comorbidity items (Quan et al. 2005), with the exception of severe infection which was derived based on a set of infection related ICD-10 previously reported (S. H. Sarica et al., 2020). ICD-10 codes for severe infection were derived from work by Inada-Kim and colleagues. This study aimed to define codes consistent with "suspicion of sepsis" in order to identify patients in National Health Service (NHS) Hospital Episode Statistics data across eight NHS trusts. Clinical consensus regarding the relevant codes was reached with input from multiple relevant specialist clinicians. The approach was successful in identifying the target population of interest (Inada-Kim et al., 2017).

Other models described in this thesis incorporate national prescribing data. This was not applicable to this study as predictive variables for the model were derived using index date as the starting time point. For most patients this will equate to the approximate time of AAV diagnosis. At this time point, most patients receive at least some of their initial supply of medications from hospital and not community dispensaries. Data from hospital prescriptions is not available in linked datasets therefore medications were not incorporated into the model. Therefore it was not possible to include potentially relevant therapies such as immunosuppressive treatment and antimicrobial prophylaxis.

4.4.5 Sample size

Sample size was determined in a pragmatic manner by using all the available data. This resulted in a significantly larger sample size compared to similar studies in AAV populations. A crucial factor to consider in relation to sample size is the number of parameters of candidate variables. Notably this refers not only to the number of variables but to each potential ß term in the eventual model. For example a variable with four categorical options would count as three

parameters for calculating sample size for a prediction model. Other important factors to consider are the total number of participants, the outcome incidence and the expected predictive performance of the model. Calculations have been developed to determine the minimal sample size for predictive models and associated statistical packages are available (Riley et al., 2019), but such tools are not currently available for models where competing risks are a consideration. There are also insufficient published models to provided indicative expected predictive model performance. As a guide, multiple sample size calculations were performed using a standard survival model approach to ensure that an appropriate number of candidate predictors were included in the model development step.

4.4.6 Missing data

Due to the administrative nature of the data, apparent missing data was minimal. SIMD was missing for a small number of individuals. A complete case analysis was carried out. Multiple imputation was not considered beneficial or necessary, as it was considered that deprivation would be difficult to ascertain based on other variables and impact on the eventual model was likely to be negligible. For the internal validation step of the model building process, it would have been necessary to include any imputation. This may have unnecessarily prolonged the compute time required for internal validation. This compute time was deemed to be more appropriately allocated to essential model building steps such as variable selection. Scottish administrative health data is recognised as having high data quality and minimal missing data (Public Health Scotland, 2023). This is difficult to quantify however, as distinguishing between data ‘not reported’ versus data ‘missing’ in a cohort study based on routinely collected administrative data is challenging, particularly where it is not possible to undertake a validation study.

4.4.7 Model rationale and development

When developing a model aiming to predict a non-fatal event, such as severe infection, it is usually appropriate to consider competing events. In this setting, the competing event in a population with AAV is death related to other causes. In this situation, prognostic models should describe absolute risk of the adverse

events, also described as the cumulative incidence. Cumulative incidence in this setting would be the risk of developing severe infection over one year, whilst acknowledging that individuals who die prior to a severe infection event can no longer experience that event. If one does not factor in a competing event during model building, then the cumulative incidence can be overestimated. The overestimation is larger where the risk of the competing event is greater. Additionally, where competing risks are not considered during the validation stage, model performance measures become inaccurate, particularly in relation of calibration. Survival models are able to take account of both censoring and competing risks, therefore such a model was selected for this study. A logistic regression model would not be able to account for these factors therefore it is not appropriate in this setting. Machine learning modelling techniques can be applied to competing risks data, however such approaches are less well described in the methodological literature and implementation can be complicated. When compared to more traditional regression methods, performance is not superior and machine learning models can be more poorly calibrated (Kantidakis et al. 2023).

There are few non-linear phenomena in nature, therefore non-linear modelling of continuous variables is encouraged in the prognostic modelling literature. Restricted cubic splines are a commonly used approach to modelling non-linear data and have been employed for many decades. An alternative approach is to use multivariable fractional polynomial (MFP) modelling - this more recent technique was used in this thesis. Polynomial functions can be used to the curved relationship that exists between many variables, but for lower order polynomials the variety of shapes is limited and for higher order polynomials there may be poor fit at the extremes of variables. Fractional polynomials are an extended group of curves, defined by a limited predefined set of power terms, which provide a vast array of different curved shapes. Restricted cubic splines would have been a reasonable approach to use, but the FP approach was used as FP terms require fewer degrees of freedom than splines and therefore allow inclusion of more candidate predictors in prognosis models with less risk of overfitting (Royston and Altman 1994). Use of FPs has shown to improve model fit and discrimination (Baneshi et al. 2013). From a practical purpose, the thesis author had also had previous training which utilised the MFP approach. Backward

elimination was used as a selection technique. This is preferred by statisticians as it starts with a plausible model (the full model). Combined with parameter-specific shrinkage, backward elimination often produces the most accurate models (Sauerbrei et al. 2020).

A non-linear FP term was fitted to age. This was performed by incorporating age as a solitary variable into a cause-specific Cox proportional hazards model transformed by FP functions with a maximum of four degrees of freedom. A backward elimination procedure was used to update FP functions. The variable selection level was set at 0.1. The resultant two suggested FP functions for age were then compared for plausible fit graphically and for parsimony (i.e. simplicity of fractional polynomial term). The full Cox proportional hazards model was then developed using a backward elimination process with the threshold for inclusion being $p < 0.1$. A cause-specific model was developed using first-severe-infection as the primary event, with all-cause mortality being the competing event. Estimated baseline survival at 1 year was reported. Model apparent performance measures were calculated including observed-to-expected (O/E) ratio, calibration intercept, calibration slope, concordance statistic (C-statistic), cumulative/dynamic area under the receiver operating characteristic curve (C/D AUC_t), Brier Score and scaled Brier score. A calibration plot was created. Definitions for these performance measures and background to calibrations plots are given below. Pseudo-observations were used as a proxy for primary event indicators to account for censored observations with respect to the calibration plot, calibration intercept and calibration slope (van Geloven et al. 2022).

4.4.8 Model performance measures

4.4.8.1 Calibration

Calibration assesses the agreement between predicted outcome proportions from the model and observed outcome proportions in data. Importantly, this is not just average calibration (“calibration in the large” or CITL), but also across the whole spectrum of predictions (van Geloven et al. 2022).

The **observed-to-expected ratio** (O/E ratio) is a simple measure of CITL or overall calibration, it indicates how close the overall estimated risk is to the overall actual outcomes of the population in question. It divides the proportion of the actual observed outcome by the expected proportion of the outcome based on the model. An O/E ratio of 1 demonstrates perfect CITL, greater than 1 suggests on average model predictions are too high, while less that 1 suggests they are too low (van Geloven et al. 2022).

The **calibration slope** and **calibration intercept** are both features of a best fit line fitted to the relationship between predicted risk from a model and the observed proportion that has the outcome. The calibration slope describes estimated risk based on the model. A value of 1 suggested ideal calibration. Less than 1 indicated that the model has overly extreme risk estimates: for those at low risk of an outcome, the model estimates that risk is even lower; while for those at high risk of an outcome, the model estimates risk is even higher. Models that give extreme predictions are said to be "overfitted". Overfitting is a common statistical problem. It results when a modelling approach is too complex for the available data, for example use of highly flexible machine learning algorithms, too many candidate predictors or variable selection based on statistical significance. The calibration intercept has an ideal value of 0. It is representative of CITL. Negative values suggest systematic overestimation of predicted risk, while positive values suggest systematic underestimation (Van Calster et al. 2019).

A **calibration plot** is a visual check of whether predicted risks for a population derived from the model match actual observed outcomes. Estimated risks and actual observed outcomes are plotted against each other. A typical method is to divide a population into ten equal groups based on estimated risk, each group is then plotted with predicted risk on the x-axis and outcome proportion on the y-axis. For example, a low-risk decile group might have a five percent severe infection estimated risk based on a model. One would expect the actual observed outcome proportion for this decile to be five percent. The plotted group will often have confidence intervals displayed. A smoothed line developed via a Loess regression is frequently added, further representing the relationship between estimated risk and observed outcomes. The smoothed line should only be displayed over the range of observed risks and not beyond. Deviations of the

decile groups or the smoothed line represent miscalibration – notably the diagonal line has intercept 0 and slope 1, equivalent to the calibration intercept and calibration slope. A significant challenge with survival data is how censored data and competing risks should be factored into calculating observed outcome proportions. One recommended method is to use pseudo-observations. These replace the primary event indicator with proxy observed event indicators for all patients, even those who were censored. Pseudo-observations are calculated as the weighted difference between the cumulative incidence estimate, at the chose time point, for the whole cohort and the same number but leaving out the individual in question. This permits straightforward calculation of observed outcome proportions, even where an observed outcome is unknown, as is the case with censored patients (Andersen and Perme 2010). Alternative techniques that deal with censoring include smoothing using a flexible regression model and inverse probability of censoring weighting (van Geloven et al. 2022). No one technique is preferred overall in the literature, therefore the pseudo-observations methods was employed, as this has been used in prior well conducted prognosis modelling research and code was available making it practical to use.

4.4.8.2 Discrimination

This is the extent to which the model can differentiate between higher and lower risk patients. Patients who experience an event earlier should have a higher predicted risk from the model than those who experience an event later or who are censored. Technically, poor discrimination is more concerning than poor calibration, because calibration can be addressed by model recalibration (Royston and Altman 2013).

The **c-statistic**, or concordance statistic (or c-index), assesses the ordering of predictions for all prediction pairs. It is a fraction where the numerator is the proportion of all possible pairs of patients where the model assigns appropriately lower or higher risk to each in the pair, while the denominator is the total number of possible pairs. The c-statistic ranges from 0.5, which indicates no discriminating ability (sometimes described as a “coin-toss” for assigning higher or lower risk to an individual) to 1.0, which indicates perfect discriminative ability. A model with perfect discrimination would always assign higher

predictive risk to the individual in a pair more likely to experience the outcome. In the setting of survival data, including competing risks, pairs can be compared such that if one individual has an event, that individual should be assigned higher risk than a paired individual that has an event later, or experiences a competing event (van Geloven et al. 2022). Notably, the c-statistic is mathematically the same as the area under the Receiver Operator Characteristic (ROC) curve. An ROC curve is a visual representation of the discriminative ability of a prognostic factor or model. It compares sensitivity on one axis to 1-specificity on the other.

The **cumulative/dynamic area under the receiver operator characteristic curve (C/D AUC_t)** is very similar to the c-statistic, but represents the model's ability to predict the event at particular time points. This can be determined for multiple time points and displayed graphically (van Geloven et al. 2022).

4.4.8.3 Overall prediction error

This characterises the overall predictive ability of the model, thus incorporating both calibration and discrimination.

The **Brier score** is the mean squared difference between observed survival at a given time point (either 1 or 0) and the predicted risk at that time point. This score ranges from 0 for a perfect model to 0.25 for a non-informative model, where the population in question has a 50% event rate at the time point in question. If the event rate is lower, then the score at which a model is non-informative becomes lower also. A scaled version has been developed to simplify interpretation across settings. The **scaled Brier score** is calculated as 1 – (model Brier Score / null model Brier score), the null model being one without covariates that predicts the same average risk for all individuals. A scaled Brier score of 100% suggests a perfect model, 0% an ineffective model and below 0% is a harmful model that is worse than the null model (van Geloven et al. 2022).

4.4.9 Model validation

Internal validation refers to using the dataset on which the model was developed to evaluate model performance. Different approaches are available, such as split-sample and cross-validation, but bootstrapping provides estimates of model

performance with low bias, low variability and efficient use of available data for model development (Steyerberg et al., 2001). The fundamental principal is "repeated sampling with replacement". A sample drawn with replacement means that a subject's data is randomly selected from the development data set. Further selection takes place from the data set, but the prior samples drawn are put back into the data set, such that they are available for random sampling again – thus replacement. For the current study, internal validation was performed using Harrell's bias correction, a bootstrap method (Harrell et al. 1996). This method has been evaluated against other bootstrap-based methods and it compared well (Iba et al. 2021). Bootstrapped samples were drawn with replacement from the original sample, until the bootstrapped dataset was the same size as the original dataset. Identical model building steps to the development of the initial model were then performed on the bootstrapped sample. Performance measures of this bootstrapped model were then calculated in the bootstrapped sample - such measures represent the apparent performance. Performance measures of the bootstrapped model were then calculated in the original sample - such measures represent the test performance. These steps were repeated with 1000 iterations. This process allows optimism to be considered. When a model's performance is tested in the dataset in which it was developed, performance measures tend to be overestimated – the extent to which this is overestimated is known as optimism. Performance measures calculated in the initial sample without any established internal validation approach yields so-called "apparent" performance measures. "Optimism-adjusted" measures are calculated by subtracting mean optimism from apparent performance (Steyerberg 2019). Mean optimism was calculated for each performance measure by subtracting mean test performance from mean apparent performance for all bootstrapped samples. Optimism-adjusted performance measures were calculated as above by subtracting optimism from the performance measures of the original model. Confidence intervals of optimism adjusted performance measures were calculated by the location-shifted bootstrap confidence interval method (Noma et al. 2021). R version 4.2.0 was used with packages including comorbidity, geepack, MASS, pec, riskRegression, survival and the tidyverse packages.

4.4.10 Patient and public involvement

Collaboration with the Aberdeen Centre for Arthritis and Musculoskeletal Health's user group and patients identified through national support groups (Vasculitis UK and the Lauren Currie Twilight Foundation) identified patient and carer priorities. This process informed the design and aims of the current study and the VOICES project overall.

4.5 Results

An AAV index cohort was derived from the VOICES cohort (Figure 4-1). The dataset comprised 2,078 individuals. Baseline characteristics are reported in Table 4-1. The developed cause-specific Cox model for first severe infection event prediction, for both the primary (severe infection) and secondary (non-infection related mortality) events, is shown in Table 4-2. Model coefficients, hazard ratios, 95% confidence intervals (95% CI), p values and the estimated baseline survival at one year are shown. Table 4-3 shows model performance measures, both apparent and optimism-adjusted using bootstrapping as a form of internal validation. Performance categories covered are calibration, discrimination and prediction error. Figure 4-2 is a calibration plot comparing estimated risks of the model to observed outcome proportions in the development cohort.

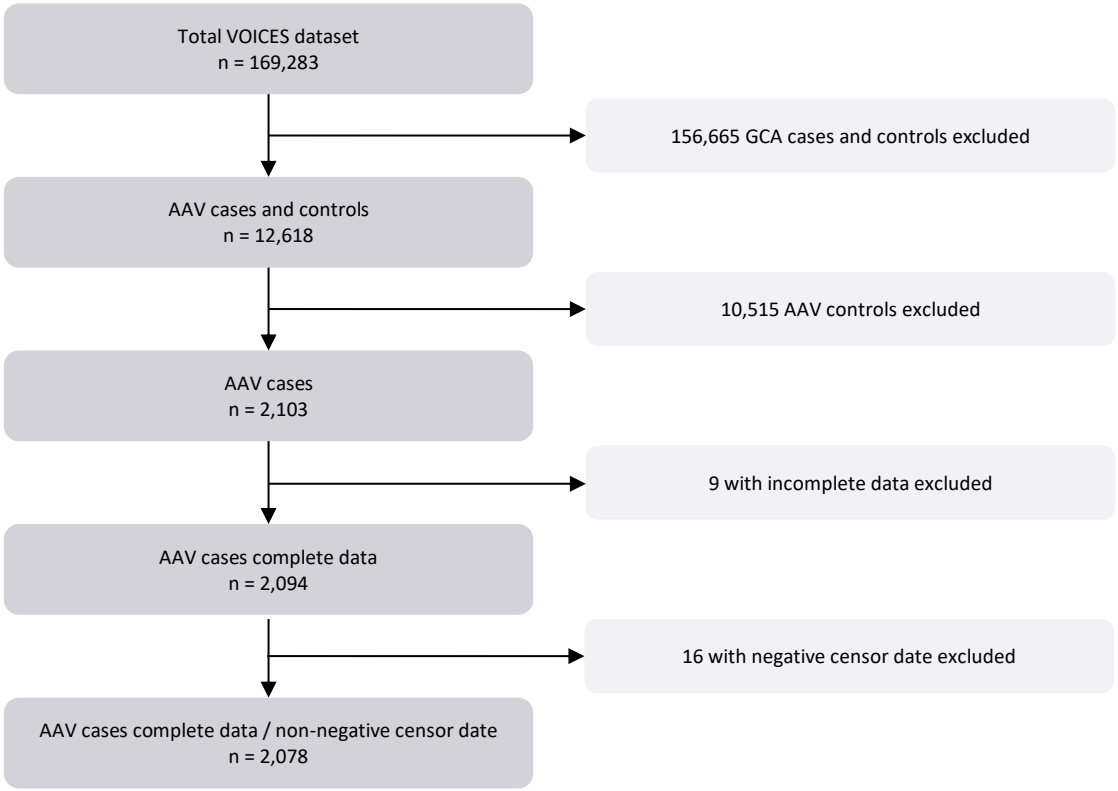


Figure 4-1 | Derivation of AAV index cohort from VOICES cohort

Table 4-1 | Baseline characteristics

Variable	No infection	Infection	Died
Total (%)	n = 1512 (72.8)	n = 428 (20.6)	n = 138 (6.6
Age; median (IQR)	58.5 (47.6 to 67.9)	65.9 (55.5 to 73.8)	71.0 (63.4 t
Female sex; n (%)	749 (49.5)	225 (52.6)	67 (48.6)
SIMD (deciles); median (IQR)	6.0 (4.0 to 8.0)	6.0 (3.0 to 8.0)	5.0 (3.0 to
Cardiovascular disease (all)	130 (8.6)	68 (15.9)	31 (22.5)
Atherosclerotic disease (all)	102 (6.7)	60 (14.0)	24 (17.4)
MI	42 (2.8)	21 (4.9)	12 (8.7)
CVD	49 (3.2)	26 (6.1)	9 (6.5)
PVD	17 (1.1)	22 (5.1)	7 (5.1)
Chronic heart failure	43 (2.8)	16 (3.7)	14 (10.1)
Hypertension	9 (0.6)	5 (1.2)	0 (0.0)
Diabetes	60-65(4)	38 (8.9)	5-10 (6)
Renal disease	100 (6.6)	66 (15.4)	22 (15.9)
Dementia	< 5*	< 5*	< 5*
Cancer (all)	59 (3.9)	20-25 (5)	10-15 (7)
Cancer (localised)	48 (3.2)	15-20 (44)	5-10 (5)
Cancer (metastatic)	11 (0.7)	< 5*	< 5*
Chronic respiratory disease	196 (13.0)	77 (18.0)	20 (14.5)
Rheumatic disease	56 (3.7)	15 (3.5)	5 (3.6)
Peptic ulcer disease	26 (1.7)	13 (3.0)	6 (4.3)
Liver disease	16 (1.1)	5-10 (2) *	< 5*
Previous infection	331 (21.9)	122 (28.5)	43 (31.2)
Charlson comorbidity index; median (IQR)	0.0 (0.0 to 0.0)	0.0 (0.0 to 1.0)	0.0 (0.0 to

CVD = cerebrovascular disease, IQR = interquartile range, MI = myocardial infarction, PVD = peripheral vascular disease, SIMD = Scottish Index of Multiple Deprivation.
***Low count cells (<5) have been suppressed, where a low count cell could be inferred from other cells these have also been suppressed and percentages rounded.**

Table 4-2 | First severe infection event prediction model

Variable	Coefficient	Hazard ratio (95% CI)	P
Severe infection (primary event)			
Age (FP term)*	2.07	7.96 (4.41 to 14.35)	<0.01
SIMD deciles	0.06	0.95 (0.91 to 0.98)	<0.01
Renal disease	0.52	1.69(1.29 to 2.21)	<0.01
Diabetes	0.47	1.59 (1.14 to 2.24)	<0.01
Non-infection related mortality (secondary event)			
Age (FP term)*	4.85	127.25 (47.99 to 337.43)	<0.01
SIMD deciles	-0.078	0.93 (0.87 to 0.98)	0.011
Renal disease	0.32	1.38 (0.87 to 2.20)	0.175
Diabetes	0.20	0.82 (0.40 to 1.69)	0.589
Estimated baseline survival at 1 year	0.807		

CI = confidence interval, FP = fractional polynomial, SIMD = Scottish Index of Multiple Deprivation

***Age (FP term) refers to the fractional polynomial transformation performed on age: (age / 100)^3**

Table 4-3 | First severe infection event prediction: model performance

Performance measure	Values (95% CI)
Apparent	
Calibration	
O/E ratio	1.01 (0.92 to 1.09)
Calibration intercept	0.01 (-0.09 to 0.11)
Calibration slope	1.05 (0.80 to 1.29)
Discrimination	
C statistic	0.61 (0.59 to 0.64)
C/D AUC_t at 1 year	0.63 (0.60 to 0.66)
Prediction error	
Brier score	0.157 (0.147 to 0.167)
Scaled Brier score (%)	3.8 (2.0 to 5.4)
Optimism adjusted	
Calibration	
O/E ratio	1.01 (0.92 to 1.09)
Calibration intercept	-0.01 (-0.10 to 0.09)
Calibration slope	0.94 (0.70 to 1.18)
Discrimination	
C statistic	0.60 (0.58 to 0.63)
C/D AUC_t at 1 year	0.62 (0.59 to 0.65)
Prediction error	
Brier score	0.159 (0.149 to 0.169)
Scaled Brier score (%)	2.6 (0.9 to 4.3)

C statistic = concordance statistic, C/D AUC_t at 1 year = cumulative over dynamic area under the receiving operator characteristic curve, CI = confidence interval, O/E ratio = observed-to-expected outcomes ratio.

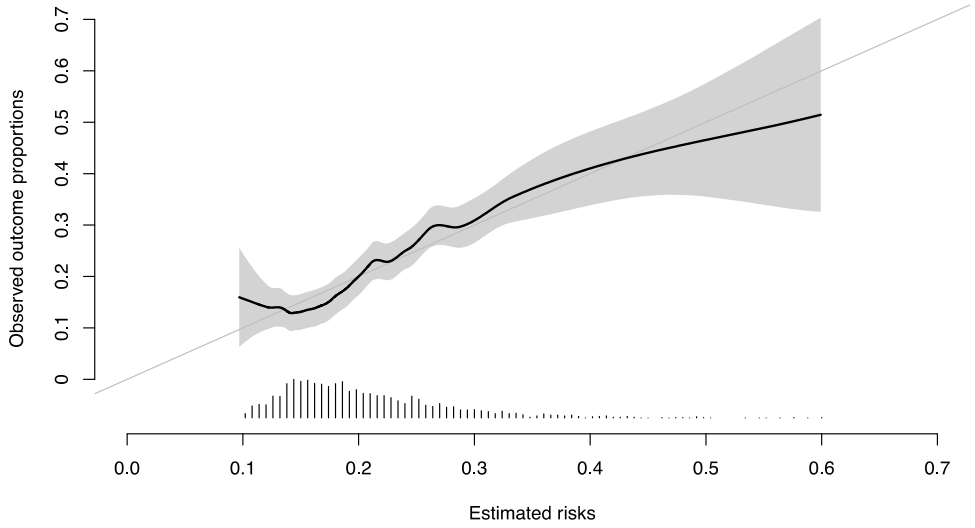


Figure 4-2 | First severe infection event prediction calibration plot

4.6 Discussion

4.6.1 Key results

This study is one of two infection prediction models in AAV reported in this thesis. The dataset is the largest in the medical literature to have been used to develop an infection clinical prediction model in AAV. It is one of the largest datasets that has been used to develop any prediction model in AAV. The data is real-world, national healthcare administrative data and theoretically has coverage of the whole Scottish population. Reported characteristics (Table 4-1) showed that this population had a high incidence of severe infection with 428 of 2078 individuals (20.6%) experiencing the primary event by one year. This demonstrates the vulnerability of this population to infection. It highlights the need to understand the epidemiology of severe infection in AAV and emphasises the importance of being able to predict its occurrence and downstream effects. On a univariable analysis presented in Table 4-1, individuals who were older, had cardiovascular disease, diabetes, renal disease and a history of prior infection were more likely to have an infection during the observation period, as opposed to not have an infection or die before the end of the observation period. Through the application of modern prediction modelling techniques, a parsimonious four component model was derived. This comprised a non-linear

transformation of age (age/100^3), a national deprivation index (SIMD in deciles), presence of prior renal disease and prior diabetes. These variables all have high levels of biological or socioeconomic plausibility for being associated with severe infection. As detailed in Chapter 1, age, diabetes and renal disease have been shown to be strong prognostic factors for severe infection across multiple observational studies in AAV. Known as immunosenescence, age-related changes in immune function lead to increase incidence and severity of infections in older people, as well as impaired efficacy of vaccination. Defects occur in the innate, cell-mediated and humoral immune systems (Weiskopf et al. 2009). For decades, diabetes has been recognised as leading to adverse effects on leukocyte biology (Robertson and Polk 1974). Vascular disease and neuropathy, in addition to attenuating an immune response, also lead to greater infection susceptibility in diabetes (Toniolo et al. 2019). Leukocyte dysfunction has also been identified in chronic kidney disease, with uraemia, renal anaemia and dialyser bioincompatibility being important causative factors (Vanholder and Ringoir 1993). Socioeconomic status has not been identified in AAV populations as being a prognostic factor for infections, but this has been reported in the general population. A large UK biobank study showed that lower socioeconomic status was associated with infections and that this may be mediated by lifestyle factors and chronic disease, such as cardiovascular disease (Ye et al., 2023). A systematic overview of reviews highlighted that lower socioeconomic status carries an associated risk of increased incidence of a number of communicable diseases (Ayorinde et al. 2023). Performance measures were calculated for the prediction model. These were adjusted for optimism using a bootstrapping approach. Performance measures showed fair discrimination, very good calibration and reasonable prediction error.

4.6.2 Novelty of findings

This is the first reported clinical prediction model developed in AAV on a large dataset of routinely collected data. It utilised modern clinical prediction methodology. This report established that it is possible to develop an effective predictive model in the domain of AAV and severe infection. This represents an important first stage in developing a clinically applicable model. With additional work, such as using other datasets to refine the model and to identify additional

important predictors, followed by comprehensive external validation, a promising clinical prediction tool could be deployed in practice.

4.6.3 Strengths

The PROBAST risk of bias and applicability tool informed evaluation of the study's strengths and limitations (Wolff et al., 2019).

Through utilising advanced data linkage capability in Scotland, it has been possible to take advantage of routinely collected datasets rarely available in other countries. This data is recognised to have a high level of completeness and overall data quality (Public Health Scotland, 2023). Routinely collected data has the advantage of including participants who would not normally be able to participate due to social circumstances or frailty. In this way, the data is highly representative of the population of interest and thus this study is at low risk of selection bias. Due to the nature of the data it is likely that inclusion criteria, exclusion criteria, predictors and outcomes were assessed in a similar way, regardless of the occurrence of the outcome. However, some underlying variability may be present due to, for example, different approaches to clinical coding across health boards in Scotland.

As the data was recorded prospectively, the recording of predictors could not have been influenced by a participant's outcome. All predictors were present at the time that the model would be intended to be used, ensuring applicability of the model. The outcome was reliably determined, with all admissions documented to be associated with an infection included. This outcome could have occurred at any Scottish hospital and was not restricted to a specific hospital or health board. There is a risk for an individual in the study, that the identified infection may not qualify as severe, as it may have occurred during an admission for another reason and may not have merited hospital admission in its own right. However it seems unlikely that this would vary across different risk categories and clinical coding guidance mandates that actively managed 'other conditions' are those which require 'significant investigation or management' – therefore it is likely that such infections do represent severe infections as defined in this study (NHS National Services Scotland 2017). Due to lack of hospital-administered medicines data, it was not possible to include intravenous

antimicrobials in the outcome data. Predictors were not included in the definition of the outcome and the outcome was determined without awareness of the presence or absence of predictors – factors which lower risk of bias.

Epidemiological studies in AAV often suffer from small sample sizes, making them susceptible to both detecting spurious associations and not detecting important true effects. As per the findings of the systematised review (Chapter 2) smaller studies in AAV typically have a sample size between 100 and 200 participants, with respect to investigations of the incidence of infection and infection prognostic factors. Only a few studies have a larger sample size, typically between 400 and 800 participants. It may be possible for such studies to detect important associations with some accuracy. However, they would be limited in the extent to which such effects could be rigorously examined, due to only being able to control for a limited number of variables in a statistical model. Spurious associations may be detected in such studies or important associations missed. Detected potential associations have wide confidence intervals, making it difficult to assess the relevance of a potential prognostic variable. Studies which have sought to develop clinical prediction models for infection in AAV have had a sample size between approximately 150 to 250 participants. (McClure et al., 2021; Zhang et al., 2022) The current study has a sample size of 2,078 participants, substantially larger than both comparable prediction studies and larger epidemiological studies. Sample size calculation in the domain of clinical prediction models is, however, a complicated subject. (Riley et al., 2020) Multiple factors need to be taken into consideration including the total number of participants, the number of events, the number of predictor parameters and the potential discriminative ability of the model. For this model, sample size was carefully considered. Due to a lack of sufficiently powered similar models and resultant lack of availability of potential model discriminative ability, it was not possible to perform a single informative sample size calculation. Therefore, a series of sample size calculations were carried out under a plausible range of scenarios with varied calculation inputs. This gave an indication of the number of model parameters that could reliably be included in the development of the model. As a result, this model was developed with minimal risk of overfitting. Out-with the rare disease setting, well conducted clinical prediction model studies typically have tens, if not hundreds, of

thousands of participants. Traditionally this would be challenging to achieve in a rare disease setting. Future models should seek to utilise large, federated sources of data, such as the FAIRVASC infrastructure, to develop models in a rare disease such as AAV with a larger selection of potential model parameters. (McGlinn et al., 2022)

Model development was performed using modern prediction methodology. Clinically relevant variables were carefully selected for this study. Studies identified for the systematised review (Chapter 1) were examined for important variables for inclusion, particularly with respect to comorbidity. This was because comorbidities were a readily available source of information given the study's data linkage design. Other literature was examined to identify important variables that are recognised as predictive of infection, both in immunosuppressed, autoimmune populations and more general populations. (Luna et al., 2016; Bahlis et al., 2021; Dixon et al., 2012) Finally, the clinical domain knowledge of the study team (thesis author and supervisors) was drawn upon to identify important variables for inclusion. Altogether, this process represents a comprehensive approach to identifying a limited set of appropriate variables for inclusion, thus maximising the potential predictive power of the eventual model, whilst minimising the risk of overfitting. Another factor which substantially limited overfitting was that, after a single demonstrator model was developed, the model was developed in a single iteration. Other approaches which maximised power and minimised overfitting were the use of continuous variables as opposed to dichotomised continuous variables and non-linear transformations with fractional polynomials of continuous variables. Variable reduction strategies are recognised as being useful for making a model parsimonious. Where an automatic variable selection approach is desirable, backward elimination is recognised as the preferred model building technique. One major advantage of backward elimination is that it starts with the full model and therefore is able to assess joint predictive ability (Chowdhury and Turin 2020). Internal validation was performed using bootstrapping. This approach is recognised as resulting in stable estimates with minimal bias (Steyerberg et al., 2001). That internal validation revealed low levels of model optimism was promising.

4.6.4 Limitations

As described above, this study is larger than any comparable studies in the AAV research literature, but significantly smaller than studies out-with the rare disease literature. As a result, confidence in the predictive ability of such a model would be lower than other predictive tools currently used in clinical practice. This is particularly the case prior to external validation of a model, which has not yet been carried out due to time constraints and lack of availability of a suitable dataset. Another theoretical limitation of the study, where it was technically hampered by sample size, was having to use a restricted set of initial potential predictive parameters. However, following the systematic approach taken to identify optimal predictive parameters, the study team was satisfied that appropriate variables were ultimately included in model development, given what variables were available from the national administrative linked datasets.

While a novel and efficient approach, an ideal data source for a prediction modelling study would be a prospective cohort with predetermined, domain specific criteria with respect to inclusion criteria, exclusion criteria and outcome definition. Validated means of prospectively identifying participants would likely be more reliable than relying on clinical coders deriving this information from a potentially incomplete clinical record. Inclusion criteria for this study included all individuals with an ICD-10 code for an AAV sub-diagnosis in the SMR-01 administrative database – a database of hospital care episodes. This may be prone to both false negatives and false positives. False negatives may occur where AAV is diagnosed in the outpatient setting and is not recorded as a significant “other condition” by clinical coders during subsequent inpatient admissions. Terminology is varied in AAV and clinical coders cannot be expected to identify all varieties of clinicians’ descriptions of AAV. False positives may occur where AAV was considered as a possible initial diagnosis, which was later found to be another condition. In Scotland there is a limit on the number of comorbidities that can be recorded for each hospital admission. A prospective design may be considered more likely to accurately identify outcomes, though prospective studies may also be limited in their capacity to identify all outcomes due to study design, lack of personnel, relying on participant self-reporting or not having access to clinical information sources. Prospective sources of data

also may be derived from randomised trials, which often comprise more homogenous populations and therefore have narrower predictor distributions with resultant models having worse discriminative ability. (Moons et al., 2019)

Data from the national datasets is recognised as having high quality and reliability, although it would have been desirable to have been able to perform a data validation study to confirm that codes describing acute hospital admissions do indeed reflect an individual's true comorbidities. Previous work of this nature performed for a similar study of individuals with AAV revealed that the accuracy of identified comorbidities was moderate to excellent, depending on the condition in question (Sarica 2018). A study of Singaporean administrative data in the setting of infection suggests that comorbidity ascertainment based on administrative data is as good as a medical chart review, although this study was based on a list of comorbidities obtained at the start of the hospital admission, whereas in Scotland administrative data for each hospital admission aims to identify the main reason for admission and up to five additional conditions which may be co-existing or may develop in hospital (NHS National Services Scotland 2017). Available variables represented a further limitation. Due to the nature of the administrative data, certain variables which may have utility for predicting infection were not available, for example vasculitis disease activity, detailed immunosuppressive medication information and biological characteristics such as immune system parameters. This likely substantially limited the potential prognostic power of the model. Various prognostic factors identified in Chapter 1 were not available for inclusion in this model. Induction and maintenance treatment regimens likely impact susceptibility to infection, but data on these therapies is not available in community prescription datasets, as these medicines are almost universally prescribed and administered in secondary care (Vassilopoulos et al. 2023). Biological parameters that are potentially prognostic of infection in AAV include the presence of leukopenia and hypogammaglobulinaemia (Lai et al., 2014; Morgan et al., 2016). Notably an important biological parameter, serum IgG level, was retained in the final infection prediction modelling in AAV by McClure and colleagues. It seems likely that such biological parameters would improve performance of the current model (McClure et al., 2021).

With respect to model development, coefficient shrinkage was not performed in this study. This may not be considered necessary due to the standard of optimism-adjusted model performance measures achieved but would be desirable in a future iteration of this study. There are two main modelling approaches for events where there are competing risks: cause-specific modelling and sub-distribution hazard approach. Cause-specific modelling was used in this study for practical reasons relating to availability to the thesis author of statistical programming code for modelling that was compatible with the approach to model performance evaluation. Both are considered acceptable, though the sub-distribution is considered preferable as it overestimates risk of events less frequently (Noordzij et al. 2013). Future work will ideally explore both modelling techniques. It was not possible to assess external validity or clinical utility due to a lack of an appropriate external dataset. These would be highly desirable studies to be performed in future.

4.6.5 Place in the current literature

Most reports in the published literature relating to identifying predictors of, or so-called 'risk factors' for, severe infection in AAV do not attempt to develop or validate a clinical prediction model. These reports are summarised in Chapter 1. Two recent reports do seek to develop prediction models: McClure *et al.* and Zhang *et al.*(McClure et al., 2021; Zhang et al., 2022)

McClure *et al.* published the first prediction model for infection in AAV. This was specifically in individuals who had been treated with a maintenance course of rituximab. Follow-up was 5.3 years, as opposed to 1 year for the current study. The first year is the most high-risk period with respect to infections. Restricting follow-up to this time period was an intentional component of the current study. The McClure *et al.* sample consisted of 147 participants from a single academic centre. The outcome of interest was time-to-first serious infection or third non-serious infection. The model had five predictors, developed using a backwards elimination procedure from a pool of 13 candidate parameters. Identified predictive variables were male sex, structural lung disease, diabetes, infections during therapy with rituximab and serum immunoglobulin G (IgG) level. There were 88 infection events. The optimism-adjusted C-statistic was 0.64, adjusted using a bootstrapping approach. Shrinkage was performed in this study by

multiplying model coefficients by a shrinkage factor, whereas shrinkage was not undertaken in the current report.

There were significant differences between the variables identified in the McClure *et al.* report compared to the current study. Diabetes was identified as predictive in both. Male sex was not identified in the current study, whereas age, renal disease and deprivation were. Previous infection was a candidate parameter in the current study but this did not have an effect significant enough to be included in the model. As discussed, a limitation of the current study was a lack of variables such as biological parameters – the McClure *et al.* model included such a parameter in the form of IgG level. They note that their model was not fully congruent with previously identified prognostic factors such as older age and presence of renal disease, both of which were included in the current model. They highlight events per variable (EPV) as a method to determine adequate sample size. They state their model meets a commonly accepted threshold of EPV > 10. Presumably their calculation resulted in an EPV of 17.6 (88 events to 5 variables). This is flawed for two reasons. In the modern prediction literature, this 'rule of thumb' EPV threshold is increasingly recognised as an inappropriate method for assessing sample size. Sample size calculations as described in the current study should instead be utilised. Furthermore, an accurate EPV must incorporate all *candidate* variables, not just those included in the final model. The concept of EPV is actually better represented as events per (candidate) parameter (EPP) (Riley et al., 2020). For the McClure *et al.* study, this would be 6.8 (88 events to 13 candidate parameters). As described in the above methods, notably the number of degrees of freedom for each variable should be included in the count of parameters, i.e. for a categorical variable with four possible options, this would count as three parameters. The McClure *et al.* study authors highlight that their study is likely underpowered and is not suitable for individual risk prediction, however it is more underpowered than the discussion of EPV alludes to. A further limitation is that age was dichotomised, rather than included as a continuous, potentially non-linearly transformed, variable. As a result, information was unnecessarily lost and model performance potentially reduced.

In comparison, relative strengths of the current study were the use of sample size calculations to carefully consider EPP, the relatively large sample size, use

of continuous variables with non-linear transformation and participants likely representative of a real-world population. Strengths of the McClure *et al.* study were excellent case ascertainment, use of shrinkage in the model development and wider availability of important biological variables. Ultimately the studies are complementary in that they sought to utilise similar, modern prediction methodology and both identify clinically and biologically plausible predictors.

The report from Zhang *et al.* sought to predict bacterial infection, however it seems that this model was concerned with identifying bacterial infection in a diagnostic, as opposed to a prognostic, sense. While neither the presence of possible vasculitis symptoms, nor the time to bacterial infection, are described in the report, it seems clear that the authors sought to use biological parameters to attempt early identification of bacterial infection, prior to microbial culture results being obtained, where the alternative diagnosis could be active vasculitis. Therefore, despite being an important question, a comparison of the current study to Zhang *et al.* was not considered instructive given the differing study objectives. (Zhang et al., 2022)

4.6.6 Future directions

This work represents an initial exploration of the development of prediction models in the domain of severe infection in individuals with AAV. It uses a novel approach by repurposing routinely collected healthcare administrative data for developing a clinical prediction model. The ultimate aim is to develop a model that could be deployed in clinical practice in an efficient manner for busy clinicians, potentially via a web application. Such a model would seek to guide clinical decision making, to inform patients about what to expect and to augment the selection of appropriate research participants.

Whilst representing a useful research development, model performance measures indicate that the reported model does not yet perform well enough to have a high level of clinical utility, even if the performance was confirmed in an external validation cohort. Adding additional clinical variables not available in the current dataset would likely improve model performance. Ideally such variables would have existing evidence supporting their ability to predict infection, biological plausibility for predicting infection or both. Additional

clinical variables which may improve the overall predictive ability of the model may be some of those which were found to be predictive in the McClure *et al*. report, such as structural lung disease and serum IgG level. Based on the wider literature explored in Chapter 1, it seems likely that age, structural lung disease, lymphopenia, renal function and smoking status may be predictive factors. Ideally this investigation would be undertaken in a large, well powered prospective cohort. Utilising different methodology such as the subdistribution hazard approach could be explored. External validation and assessment of clinical utility should be undertaken.

At present it remains unproven whether it is possible to develop a model that will predict severe infection in AAV patients with sufficient accuracy for use in routine clinical practice. However, this report gives a strong indication that it will be possible to develop such a model. Unresolved questions remain, including identification of the most important predictive variables. A focus of future research should not only be the development and refinement of prognostic models, but also the identification of prognostic factors – a fundamental but sometimes overlooked area of research in the prognostic research sphere (Riley et al., 2013).

4.7 Conclusion

This work represents the largest study yet undertaken to develop a clinical prediction model for severe infection in individuals with AAV. It is the only reported model focused on all individuals with AAV. It uses modern prediction methods and the resultant model had good predictive ability. Future work should seek to build on this model, by adding additional predictive variables, undertaking external validation and investigating clinical utility. The ultimate aim is that such a model will provide clear clinical and research benefit to AAV patients.

4.8 Summary

This chapter reported the development and internal validation of a prognostic model for severe infection events in individuals with AAV, over the course of the first year following diagnosis. This is the first time such a model has been

described. Upon further development, models such as this will enable clinicians and researchers to identify individuals at the highest risk of severe infection. This will facilitate clinical trials aimed at reduced morbidity and mortality from infection in this vulnerable population. The next chapter will continue to apply modern prediction methodology by developing a prognostic model for early mortality following severe infection in individuals with AAV.

5 Development and internal validation of a multivariable model to predict early mortality in individuals with ANCA-Associated Vasculitis (AAV) and severe infection

5.1 Overview

In the previous chapter, a prognostic model that predicts severe infection events in ANCA-associated vasculitis (AAV) was developed and internally validated. As has previously been established, many individuals with AAV will experience a severe infection. This chapter will consider the time point of a severe infection episode. It will seek to develop a similar model aiming to predict early mortality after a severe infection using modern prognostic modelling methodology. Such a model, following external validation and assessment of clinical utility, could be used in both the clinical and research settings to identify those at the highest risk of death. This high-risk group could be studied in randomised controlled trials to explore novel therapies with the aim of reducing mortality due to severe infection.

5.2 Abstract

5.2.1 Background

Individuals with AAV are at high risk of infections which can lead to early mortality. No predictive models for mortality in this setting exist. This chapter describes the development and internal validation of such a model.

5.2.2 Methods

The data source was a linked, routinely collected dataset with full coverage of the Scottish population. All participants were AAV patients and had at least one admission to hospital associated with an infection. Candidate predictors included demographics, comorbidities and community prescribed medications. The study outcome was death occurring prior to hospital discharge or within 30 days of discharge. Logistic regression, with a backwards selection procedure which fitted fractional polynomials to continuous variables, was used to develop the model. Elastic net penalised logistic regression was used to shrink model coefficients. Optimism-adjusted performance measures were estimated using bootstrapping.

5.2.3 Results

1,015 patients were included in the dataset, 157 (15.5%) of whom suffered the outcome. The final model included age, time since AAV diagnosis, presence of specific comorbidities (liver disease, metastatic cancer, renal disease and diabetes) and recent glucocorticoid exposure. On internal validation, optimism-adjusted model performance statistics included calibration slope 0.967, calibration in the large 0.004 and concordance statistic 0.713.

5.2.4 Conclusions

This predictive model represents an important step in developing a prediction model for the risk of death for individuals with AAV who experience a severe infection.

5.3 Introduction

Individuals with ANCA-associated vasculitis (AAV) require potent immunosuppressive therapy to prevent disease related morbidity and mortality.(Kitching *et al.*, 2020) As a result of this therapy, and disease related factors, severe infection is a frequent adverse clinical outcome.(Sarica *et al.*, 2018) This has a major impact on AAV patients: within the first year of therapy, infection is recognised as the leading cause of death.(Little *et al.*, 2010) Prognostic models for severe outcomes such as death have been developed in a wide range of conditions such as critical illness, cancer and cardiovascular disease.(Keuning et al., 2020; Phung, Tin Tin and Elwood, 2019; Damen et al., 2016) Such models aim to improve risk stratification and guide clinical decision making. Prognostic models for mortality have been developed in populations with infection such as severe sepsis and community acquired pneumonia (Vincent et al., 1996; Shapiro et al., 2003; Fine et al., 1997; Lim et al., 2003) but, to the authors' knowledge, no such prediction models have yet been developed in an AAV population. Such a prediction model would have important clinical and research applications. It could assist clinicians with management decisions such as whether to suspend or delay immunosuppressive therapy. Through quantifying risk of death, clinicians would be better able to communicate severity to patients and families. A predictive score could assist with risk stratification for clinical trials, enabling novel therapeutic strategies to be tested in those with the highest need.

There has been a strong focus in the recent predictive research methodology literature on formal methods for sample size calculation for model development.(Riley *et al.*, 2020) One issue that hampers advanced statistical modelling in AAV is sample size. Due the rarity of AAV, many cohorts on which previous epidemiological models have been developed have had an insufficient sample size to accurately quantify the association between prognostic variables and outcomes in multivariable models. The FAIRVASC project, described in Chapter 2, is an example of a large scale multinational collaboration aiming to pool data from multiple registries in order to achieve sufficient sample size for such analyses.(McGlinn *et al.*, 2022) Another approach is to undertake data linkage studies which utilise routinely collected data. Such studies are possible in Scotland utilising infrastructure provided by Public Health Scotland (PHS).

Our aim was to develop and internally validate a predictive model for mortality within 30 days of discharge of an admission associated with infection in patients with AAV, utilising routinely collected data from a large data linkage study.

5.4 Methods

5.4.1 Study design and setting

Reporting in the current study followed the TRIPOD (Transparent Reporting of a multivariable prediction model for Individual Prognosis Or Diagnosis) guidelines.(Collins *et al.*, 2015) Vasculitis Outcomes in relation to Care Experiences (VOICES) is a services mapping study of vasculitis care and outcomes with projects including both Scotland and the whole of the United Kingdom. Further information regarding the study can be found in Chapter 3 and online (VOICES, University of Aberdeen, 2022). VOICES includes a Scottish data linkage matched cohort study aiming to examine patterns of vasculitis care and outcomes. This dataset was utilised in the current study. The dataset identified all patients in Scotland with a relevant International Statistical Classification of Diseases and Related Health Problems 10th Revision (ICD-10) code for AAV and Giant Cell Arteritis (CGA) from the Scottish Morbidity Record admissions database (SMR01). SMR01 captures admission data from all non-obstetric and non-psychiatric inpatient and day case care episodes. Ten general population controls were identified as part of the VOICES study, this control data was not utilised in this current chapter. The controls were matched by age, sex and health board of residence. Community health index (CHI) numbers were used to link to administrative health care data from multiple national datasets. CHI numbers are unique identifiers allocated nationally to patients in Scotland (Public Health Scotland, 2024). An index date representing AAV diagnosis was defined as the earliest admission date associated with an AAV ICD-10 code. A look-back period of 5 years allowed comorbidities that occurred prior to the index date to be determined, according to established methodology.(Quan *et al.*, 2005) Data linkage was performed by PHS using a robust approach that has been demonstrated to result in highly accurate and complete data.(Evans and MacDonald, 1999; Scottish Public Health Observatory, 2022)

5.4.2 Participants

The study period was defined from 1st April 1996, when ICD-10 was introduced, to the latest available record at the time of linkage, 31st October 2020. GCA patients and all matched controls were removed from the dataset to derive an AAV retrospective cohort. This cohort comprised all adults (aged 16 years or older) with an AAV code within SMR01. The following ICD-10 codes were used to identify patients: M31.1 (granulomatosis with polyangiitis), M31.7 (microscopic polyangiitis) and M30.1 (eosinophilic granulomatosis with polyangiitis). Patients were followed up from index date to death or the end of the study period, whichever occurred first. Patients who had not experienced an admission with infection during the follow-up period were excluded.

5.4.3 Outcome

The sole outcome in this study was mortality, defined as occurring either in-hospital or within 30 days of discharge. As a measure to preserve pseudonymisation, date of death is recorded within PHS linked datasets as month and year only. As a result, for a small number of patients death could not be determined to be within the defined time frame. These patients were excluded from the final dataset.

5.4.4 Independent predictor variables

Candidate predictors were selected based on published prognostic literature on mortality in the setting of severe infection and clinical judgment of the study team.(Fine et al., 1997; Shapiro et al., 2003; Hespanhol and Bárbara, 2020; Bahlis et al., 2021) Variables included age, sex, Scottish Index of Multiple Deprivation in deciles (SIMD; as continuous variable – see below for discussion), time since diagnosis, prescribed glucocorticoids over prior 90 days to admission (prednisolone equivalents), cotrimoxazole taken in the prior 90 days, previous admission with infection and comorbidities including prior myocardial infarction, chronic heart failure, peripheral vascular disease, cerebrovascular disease, dementia, chronic respiratory disease, liver disease, diabetes, renal disease and metastatic cancer. SIMD is a rank assigned to small areas called data zones and derived from multiple indicators (Scottish Government, 2020). There are 6,976 data zones in Scotland, the average data zone population in 784 people.

Deprivation is assessed in each data zone across seven indicators: income, employment, education, health, access to services, housing and crime. Each data zone is ranked from most deprived (rank 1) to least deprived (rank 6,976). Notably this rank is assigned to an area and not every person who lives in this area will experience deprivation. For analysis purposes, SIMD is often divided into quintiles or deciles. This study utilised SIMD in deciles, an ordinal variable but one with many categories. Particularly if the underlying data is likely to be normally distributed, it is reasonable to analyse data with multiple ordinal categories as if it were continuous data (Verhulst and Neale, 2021). As a complex trait, deprivation is likely to be normally distributed. An advantage of analysing data as a continuous variable is that fewer candidate parameters are 'used up' in a prediction model. Entering a categorical variable with 10 categories into a prediction model represents nine candidate parameters – one for each degree of freedom. Entering the same variable into a model, but as a continuous variable only represents one candidate parameter. This effectively lower the events per candidate predictor (EPP), discussed below, thus enhancing the statistical power of a study or increasing the number of alternate candidate predictors that may be included. Time since diagnosis was defined as the difference between the index date and the date of admission. Prescription data was linked from the national Prescribing Information System.(Public Health Scotland, 2022) The following conversion factors were applied to non-prednisolone glucocorticoids as per the British National Formulary: betamethasone – 6.667, dexamethasone – 6.667, hydrocortisone – 0.25 (Joint Formulary Committee, 2022). Comorbidities were derived from SMR01 episode codes as Charlson comorbidity index (CCI) items (Quan *et al.*, 2005), with the exception of severe infection. The derivation of severe infection codes used in this thesis was from previously published work and is described in more detail in Chapter 3, Section 3.4.6 (Inada-Kim *et al.*, 2017).

5.4.5 Sample size

Sample size was calculated using recommended methodology (Riley *et al.*, 2020). Prediction models for similar populations were not available. Other models assessing mortality in the setting of infection report concordance statistics (c-statistics) typically between 0.7 and 0.9. Using an outcome prevalence in our data of 14.9%, we tested various scenarios with a range of c-

statistics from a conservative range of 0.7 to 0.8 and model parameters from 4 to 20. For a c-statistic of 0.7 and 7 parameters, the minimum required sample size was 940 individuals and 141 events, corresponding to an events per predictor (EPP) ratio of 20.01. For a c-statistic of 0.8 and 18 parameters, the minimum required sample size was 990 individuals and 148 events, corresponding to an events per predictor (EPP) ratio of 8.2. For the purposes of this exploratory analysis we selected 17 predictors, giving an EPP of 9.24 for our data.

5.4.6 Missing data

Due to the administrative nature of the data, there was no apparent missing data with the exception of SIMD for one individual. Carstairs index data was available, and SIMD was approximated based on this.

5.4.7 Model development

Multiple fractional polynomial (MFP) functions with a maximum of four degrees of freedom were fit to all continuous variables. MFP is a technique for modelling non-linear continuous variables and is described in greater detail in Chapter 4, Section 4.2.7. A backwards elimination procedure was used to update MFP functions and select variables. Variable selection level was set at 0.1. An elastic net logistic regression model was then fit on the variables selected with variables transformed as MFPs where appropriate. Elastic net regression is a form of linear regression, whereby penalty terms are used to “shrink” model coefficients. Coefficient shrinkage reduces the size of the coefficient and is an approach used to reduce model overfitting. It can reduce the coefficient of a variable to close to zero: effectively removing such a coefficient from, and thus simplifying, the model. This a form of feature selection. Such approaches are also known as regularisation techniques. Elastic net is a combination of two other forms of regularisation, least absolute shrinkage and selection operator (LASSO) and ridge regression, and has advantages of both. Parameters that require specification include Lambda, which determines the degree of shrinkage, and alpha, which determines where the formula sits between LASSO and ridge regression. Lambda can be determined using cross validation and for this study was selected as the minimum value following k-fold cross validation with 10

folds. Alpha can be prespecified, different values can be trialled or a cross-validation selection procedure can be used. For this study a practical approach was used, with alpha being prespecified at 0.3. C-statistic, calibration-in-the-large (CITL; calculated using logistic regression with the model linear predictor as an offset) and the c-slope (calculated as the coefficient of the linear predictor in a logistic regression model) were determined. A calibration curve was plotted, with the population divided into deciles of predicted probability and a LOESS curve fitted to all the data. A detailed discussion of prognosis model performance measures and calibration plots can be found in Chapter 4, section 4.4.8. All analyses were conducted in R version 4.2.0 with packages including pmsampsize, mfp, rms, glmnet and pROC (R Core Team, 2022).

5.4.8 Model validation

The model was then fit to bootstrapped samples with 1000 iterations. A discussion of bootstrapping and optimism adjusted performance can be found in Chapter 4, Section 4.4.9. Apparent performance of each bootstrap model with calculated in the bootstrap sample and test performance calculated in the original sample for the above predictive performance statistics. Optimism was calculated by subtracting the mean test performance from the mean apparent performance. Optimism adjusted performance was calculated by subtracting optimism from the original performance.

5.4.9 Bias

There are several potential sources of bias in a cohort study that is derived from routinely collected administrative data, such as the current study. These include ascertainment bias, selection bias, medical surveillance bias and inflation bias. Such sources of bias, and approaches to mitigation, are addressed in Chapter 3, section 3.4.7 and Chapter 4, section 4.6.

5.4.10 Patient and public involvement

Collaboration with the Aberdeen Centre for Arthritis and Musculoskeletal Health's user group and patients identified through national support groups (Vasculitis UK and the Lauren Currie Twilight Foundation) identified patient and carer priorities. This process informed the aims of the current study.

5.5 Results

The AAV infection cohort was derived from the VOICES cohort, as shown in Figure 5-1. The dataset comprised 1,105 patients with AAV who had at least one admission with an infection following the index date. 157 (15.5%) of patients died in-hospital or within 30 days of discharge. Baseline demographic and clinical features are shown in Table 5-1.

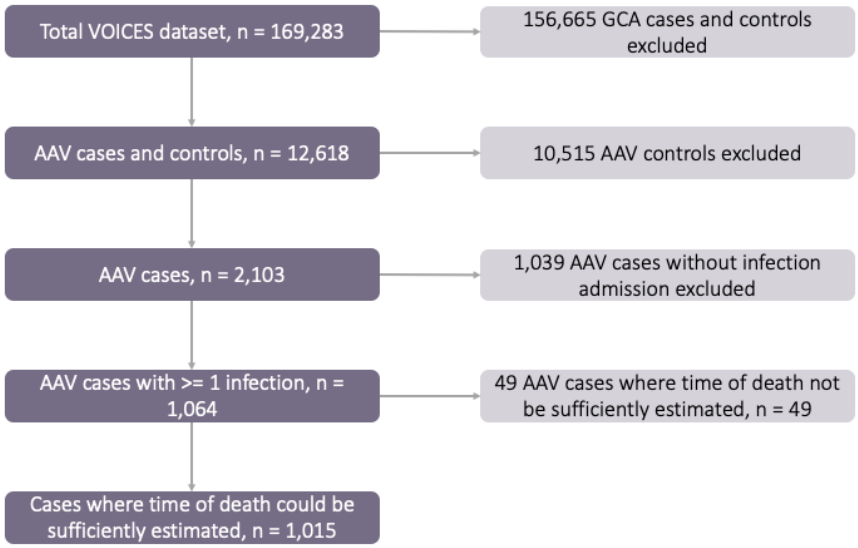


Figure 5-1 | Derivation of AAV infection cohort

Table 5-1 | Baseline characteristics of individuals who survived, who died and the whole cohort

Variable	Alive	Deceased	Total	p
Total N (%)	n = 858 (84.5)	n = 157 (15.5)	n = 1015	
Age (years); median (IQR)	67.0 (56.6 to 74.9)	73.9 (65.0 to 80.3)	68.1 (57.3 to 76.0)	<0.001
Female sex	434 (50.6)	72 (45.9)	506 (49.9)	0.317
SIMD (deciles); median (IQR)	6.0 (3.0 to 8.0)	5.0 (3.0 to 8.0)	6.0 (3.0 to 8.0)	0.661
Cardiovascular disease (all)	227 (26.5)	54 (34.4)	281 (27.7)	0.052
Atherosclerotic disease (all)	192 (22.4)	42 (26.8)	234 (23.1)	0.274
Myocardial infarction	76 (8.9)	15 (9.6)	91 (9.0)	0.897
Cerebrovascular disease	85 (9.9)	17 (10.8)	102 (10.0)	0.835
Peripheral vascular disease	61 (7.1)	18 (11.5)	79 (7.8)	0.087
Chronic heart failure	92 (10.7)	21 (13.4)	113 (11.1)	0.404
Hypertension*	20-25	<5	27 (2.7)	>0.9
Diabetes (all)	122 (14.2)	37 (23.6)	159 (15.7)	0.004
Diabetes (without complication)	112 (13.1)	30 (19.1)	142 (14.0)	0.059
Diabetes (with complication)	10 (1.2)	7 (4.5)	17 (1.7)	0.009
Renal disease	338 (39.4)	79 (50.3)	417 (41.1)	0.014
Dementia*	15-20	<5	20 (2.0)	>0.9
Cancer (all)	98 (11.4)	21 (13.4)	119 (11.7)	0.572
Cancer (localised)	85 (9.9)	14 (8.9)	99 (9.8)	0.812
Cancer (metastatic)	13 (1.5)	7 (4.5)	20 (2.0)	0.033
Chronic respiratory disease	268 (31.2)	46 (29.3)	314 (30.9)	0.698
Rheumatic disease	78 (9.1)	11 (7.0)	89 (8.8)	0.487
Peptic ulcer disease	39 (4.5)	8 (5.1)	47 (4.6)	0.924
Liver disease (all)	22 (2.6)	10 (6.4)	32 (3.2)	0.024
Liver disease (mild)	18 (2.1)	8 (5.1)	26 (2.6)	0.056
Liver disease (mod-severe)*	<5	<5	6 (0.6)	0.517
Charlson comorbidity index; median (IQR)	1.5 (1.6)	1.9 (1.9)	1.5 (1.6)	0.004
Previous infection	503 (58.6)	85 (54.1)	588 (57.9)	0.338
Time since AAV diagnosis (years); median (IQR)	5.3 (5.2)	4.7 (5.1)	5.2 (5.2)	0.184
GC exposure (mg daily); median (IQR))	5.0 (8.5)	5.3 (9.3)	5.0 (8.6)	0.701
Co-trimoxazole prophylaxis	96 (11.2)	22 (14.0)	118 (11.6)	0.379

*** low count cells (<5) have been suppressed, where a low count cell could be inferred from other cells these have also been suppressed**

GC = glucocorticoid, IQR = interquartile range, SIMD = Scottish Index of Multiple Deprivation
Glucocorticoid exposure was quantified in daily prednisolone equivalents

5.5.1 Model development and internal validation

We identified 17 candidate predictors as described in section 5.4.4. To reduce model parameters and the potential for over-fitting, SIMD deciles were considered as a continuous variable. Model predictors were selected by backwards elimination and flexible polynomials functions were fit to continuous variables. These variables were then entered into an elastic net penalised logistic regression model. The selected variables were age, liver disease (any severity), diabetes (with or without complications), renal disease and metastatic cancer. The final model is shown in Table 5-2. The model performance statistics described above are shown in Table 5-3. Figure 5-2 shows the model calibration plot which demonstrated minimal overfitting.

Table 5-2 | Final model derived by fitting of fractional polynomials and backwards selection, followed by penalised logistic regression (elastic net)

Model intercept and coefficients	Coefficient	Odds ratio
Intercept	-5.299	NA
Age (years) / 100	4.208	64.9
Liver disease (any severity)	0.925	2.5
Metastatic cancer	1.315	3.7
(Time since diagnosis (years) / 10) ^-2	0.000	1
(Time since diagnosis (years) / 10) ^-1	0.016	1
Renal disease	0.318	1.4
Diabetes (with or without complications)	0.528	1.7
Log((GC exposure + 1)/1000)	0.558	1.7
Log((GC exposure + 1)/1000)^2	0.092	1.1

GC = glucocorticoid. Glucocorticoid exposure was quantified in daily prednisolone equivalents.

Table 5-3 | Model performance statistics

Performance measure	Apparent	Optimism-adjusted	Optimism
Calibration slope	1.084 (0.835 - 1.345)	0.967 (0.727 - 1.217)	0.117
Calibration intercept (CITL)	0.000 (-0.180 – 0.174)	0.004 (-0.176 – 0.178)	-0.004
Discrimination: C-statistic	0.730 (0.686 – 0.773)	0.713 (0.668 – 0.757)	0.017

CITL = Calibration in the large

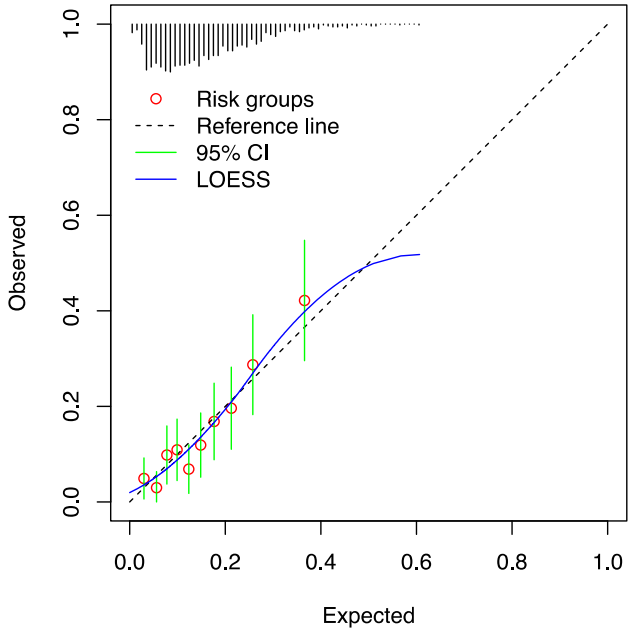


Figure 5-2 | Calibration plot
A graphical assessment of the degree to which predicted risks for a population derived from the model match actual observed outcomes. Expected (estimated) risks and actual observed outcomes are plotted against each other. The data was divided into ten equal groups based on estimated risk, with each group then plotted with predicted risk on the x-axis and outcome proportion on the y-access (red points, with green confidence intervals). A curved loess line was also applied to the same data.

5.6 Discussion

5.6.1 Key results

This study is the second of two prognostic models relating to severe infection reported in this thesis. This study is the second largest on such a topic known to the thesis author, while Chapter 4 presents a model based on the largest known. It is the first model that predicts mortality in AAV patients with an established severe infection, in this study this population was defined as experiencing a hospital admission associated with infection. A routinely collected, national dataset was utilised. Reported characteristics revealed that a substantial proportion of individuals in this study experience the outcome, with 157 (15.5%) of 1,015 dying in hospital or within 30 days of discharge. This provides further evidence of the unmet need of the population understudy including tools that would help clinicians, researchers and patients determine who is at risk of

death. A univariable analysis, presented in Table 5-1, highlighted factors with potential prognostic qualities that were associated with the outcome. These factors included age, cardiovascular disease, diabetes, renal disease, metastatic cancer and liver disease. CCI was also associated with mortality, but was not a predetermined candidate parameter, therefore was not included in model development. Sex, SIMD, time since diagnosis, glucocorticoid exposure and co-trimoxazole prophylaxis were predetermined candidate parameters, but were not associated with the outcome on univariable analysis. The final model, presented in Table 5-2, included seven parameters. Liver disease, metastatic cancer, diabetes and glucocorticoid exposure all had strong prognostic ability, however age appeared to be the strongest predictor by a substantial margin. Notably, the fractional polynomial method transforms variables in a variety of manners. Age was transformed in the model as age in years divided by 100 and carried an odds ratio of 64.9. If age were to be represented in a more clinically meaningful manner, the associated odds ratio would remain indicative of this variable being the strongest predictor. The model coefficients for time since diagnosis parameters were shrunk by the elastic net procedure to effectively zero, thus removing this variable from the model. Optimism-adjusted model performance statistics were derived through bootstrapping and reported in Table 5-3. These showed excellent calibration and good discrimination. A calibration plot also reflected excellent calibration.

5.6.2 Novelty of findings

Alongside Chapter 4, this study represents one of the first clinical prediction models developed in AAV using healthcare administrative data derived though data linkage. It utilised advanced prediction statistical methodology. While mature clinical prediction tools for mortality in the setting of infection have been developed in the general population, this is the first focus on AAV. There is a necessity to develop such models in AAV because the underlying risk, biology and clinical outcomes related to severe infection are different in this condition to other populations. Chapter 2 and other related literature demonstrated the high risk of severe infection, which is considerably greater than the general population (S. H. Sarica et al., 2020). The model described in this chapter is an important first step in realising a clinically applicable model. Further refinement of the model in other settings, including external validation and evaluation of

clinical utility using decision curve analysis, may allow a model such as that in the current study to be used for patient and research benefit at the bedside (Vickers, Van Calster and Steyerberg, 2016).

5.6.3 Strengths

This study had several strengths. These were similar to those of the Chapter 4 prognostic modelling study, but the following gives a brief overview. The PROBAST risk of bias and applicability tool was used to inform consideration of this study's strengths and limitations (Wolff et al., 2019).

Clinical studies in AAV are often hampered by sample size, while the present study was much larger by comparison. Few prognostic modelling studies exist in the AAV infection literature. The existing studies have had a sample size of approximately 150 to 250 individuals (McClure et al., 2021; Zhang et al., 2022). With a sample size of 1,015 participants, the current study is at least four-fold larger, which had a major impact with respect to model reliability. The result is a reduced propensity to overfitting and enhanced internal validity. This study is comparable in terms of size to studies of infection-related mortality in much larger populations. The Mortality in Emergency Department Sepsis (MEDS) score was developed using 2,070 patient visits, with some patients contributing more than one visit (Shapiro et al., 2003). The CURB-65 score, which is routinely used in clinical practice, was developed in a cohort of 718 individuals – substantially smaller than the current study (Lim et al., 2003).

Due to the use of routinely collected administrative data, in theory our dataset covers the entire Scottish population, resulting in the results being highly generalisable. The data linkage required to facilitate the creation of such a dataset is possible in Scotland though the service of PHS, capabilities that are not routinely available in other countries. The data linkage is highly robust and applies a deterministic method using CHI numbers, which are allocated nationally to all patients in Scotland (Dusetzina et al., 2014). Such a population-based study results in improved generalisability, as patients that may be excluded from traditional cohort studies are still represented. As a result, selection bias is minimised. PHS data is routinely audited and is recognised as

high quality (Public Health Scotland, 2023). Predictors and outcomes were therefore determined with high reliability.

Various important methodological recommendations from the modern prediction modelling literature were applied: sample size calculations were performed, continuous variables were not dichotomised thus maximising information, a non-linear approach was taken to modelling continuous variables, model parameters were restricted to reduce overfitting, internal validation was performed using bootstrapping and our approach to shrinkage applied elastic net penalisation. Elastic net regression has the advantage of the abilities to improve model fit and remove variables, resulting in a more parsimonious model and increased reliability when tested in other populations (Riley et al., 2021). The benefits of the other utilised aspects of prediction methodology are discussed in greater detail in chapter 4.

5.6.4 Limitations

Limitations of the current study include case ascertainment. Cases are identified though care episode ICD-10 codes. Whilst PHS data is known to have high standards of data quality, establishing that the identified cases had confirmed AAV would be reassuring. Sensitivity analyses could provide added confidence, for example by focusing on subpopulation of the AAV cohort with features consistent with having an AAV diagnosis, such as attendance at relevant speciality clinics and community prescription of appropriate medications. Unfortunately, such sub-studies were not feasible during time available. A nested data quality study would also have provided added reassurance regarding case ascertainment of AAV subjects. This was not possible given study permissions at the time of analysis. Ascertainment of comorbidities has similar issues, although is likely reliable due to current coding practices and quality checks (Public Health Scotland, 2023). A prospective cohort study may claim higher degrees of accuracy in measuring comorbidity. A more detailed discussion of case ascertainment issues and the potential impact can be found in Chapter 3, section 3.6.3.4.

Sample size assessment was challenging in the absence of similar studies. Recommended formulae for calculating sample size with respect to prognosis

modelling studies require the inclusion of performance measures from comparable studies (Riley et al., 2020). Indices from comparable studies in non-immunosuppressed populations were utilised and a conservative approach with respect to the number of included predictors was therefore adopted. There was still the potential that too many candidate predictors were included, as there was incomplete information to reliably estimate required sample size. Elastic net regression was employed to address this concern. While sample size was substantially larger than other prognostic modelling studies in the AAV literature and comparable to infection mortality studies in larger populations, the current study remains much smaller than some other datasets used to develop clinical prediction tools used in routine clinical practice. The PREDICT tool for survival following surgery for breast cancer was developed on a population of 5,694 individuals (Wishart et al., 2010). QRISK3, which predicts future cardiovascular disease, was derived from a population of 7.89 million patients (Hippisley-Cox et al., 2017). While a derivation cohort of several million would be impossible to achieve in AAV, it would be desirable to increase the size of future studies in AAV beyond that of the current study.

Due to the nature of the administrative data, potentially useful candidate predictors were not present. Physiological variables, such as blood pressure and conscious level, and blood test information, such as kidney function and inflammatory markers, were not available and potentially could have improved model performance. Immunosuppressive medications are additional potentially important candidate predictors that, beyond glucocorticoids, were not possible to include in the current study. Immunosuppressive therapy used for induction is effectively universally prescribed, and administered, in secondary care settings. As a result, the administration of these medications is not apparent in the community prescription data. Given that many individuals in the study will have recently been treated with rituximab or cyclophosphamide, and therefore may be at higher-than-average risk of infection-related death, while some individuals may have been several years post any immunosuppressive therapy, and at much lower risk of infection-related death, is a significant limitation that must be acknowledged. Therefore, it seems likely that such variables may be important prognostic factors and should be strongly considered for future prognostic modelling studies in this setting. To some extent, glucocorticoid dose may act as

a marker of recency of other immunosuppressant exposure, given recommended therapeutic approaches for AAV (Geetha et al., 2018). As described in Chapter 1, the burden of infection-related death clusters near the period closely following induction treatment. A further related factor is that immunosuppression is frequently de-escalated or paused in the setting of severe infection. This aspect of therapy was also unknown in the current study and therefore was not possible to include in model development. Such medication changes may impact survival, therefore this may represent an area where the model could be improved, should such data be available. Medication changes are, however, difficult to categorise, with consideration needed for medication type, dose, change of dose, timing of change and duration of change. This may make inclusion of such a variable in future models challenging. A preferred study population for a future study would be from a prospective cohort with predetermined design for prognostic modelling of infection-related mortality, similar to that described in Chapter 4, section 4.6.6.

5.6.5 Place in the current literature

Other well-established models have been developed in more general populations. The MEDS score enabled stratification with a c-statistic of 0.78 in a validation cohort.(Shapiro *et al.*, 2003) The CURB-65 score for mortality in the setting of community acquired pneumonia demonstrated strong positive predictive value for identifying individuals at low risk of death and has seen widespread clinical adoption.(Lim *et al.*, 2003) Models relating to infection in immunosuppressed populations are less readily available. One published model exists to predict the occurrence of infections in AAV patients treated with rituximab.(McClure *et al.*, 2021) A C-statistic of 0.64 was obtained for this model. This model was not aimed at predicting infection-related mortality. A similar model exists for SLE.(Tejera Segura *et al.*, 2019) The performance of existing infection mortality prediction scores, developed in more general populations, in an AAV population is currently unknown. However it is plausible that performance of these scores could be enhanced with AAV specific information, particularly in relation to immunosuppressive therapies and to comorbidities, which AAV patients are known to accumulate at a faster rate than the general population.(Shifa H. Sarica et al., 2020) A major gap that remains in the literature is the lack of models which predict infection-related mortality and

are specific to immunosuppressed populations. The present study aims to address this issue by developing an initial model through a data linkage approach, enabling comorbidities to be captured and factored into the prediction model. The same applies to glucocorticoids, but other immunosuppressive therapies were not available in the administrative dataset. The reported model performance measures, including a C-statistic of 0.713, can be classified as 'good', however achievable performance statistics vary depending on clinical scenarios. In some areas a c-statistic of 0.7 is difficult to achieve, whereas other clinical settings can achieve c-statistics of 0.9 in external validation. (Smolyansky et al., 2021; Strijker et al., 2019)

5.6.6 Future directions

The model described in this chapter demonstrated good discrimination and excellent calibration, but there is potential for enhancement. Future efforts should utilise larger and varied datasets, such as those from different geographical settings. Ideally prospective data with should be used. Studies should be designed to collect predictor variables that are specific to, and are likely to have prognostic value for, AAV patients. Such variables include clearly ascertained comorbidities, exposure to immunosuppressant therapies and biological parameters such as kidney function and immunoglobulin levels. External validation of models and assessment of clinical utility is essential (Steyerberg and Harrell, 2016; Vickers et al., 2016). If models with sufficient utility can be developed, application in clinical and research settings should follow. The models could be used to identify populations at high risk of mortality and clinical trials of approaches to reduce mortality could be applied. Such approaches may include adaptation to immunosuppressive therapy at the time of severe infection, different antimicrobial regimens with consideration for microbes that AAV patients are susceptible to, and novel anti-infection therapies. Such novel therapies may include use of genetically enhanced bacteriophages – viruses that infect bacteria – or removal of bacteria and bacterial toxins using extracorporeal techniques (Al-Shayeb et al., 2020; Didar et al., 2015). When alternative therapeutic strategies are available, prognostic models should incorporate these therapies as predictors. This would enable a stratified medicine approach whereby clinicians recommend individualised therapies to patients on the basis of prognosis research (Hingorani et al., 2013).

5.6.7 Conclusion

While this study demonstrates several strengths, due to its exploratory nature we would not recommend clinical application of the current model. Future studies should address external validity and clinical utility. They should incorporate a wider range of plausible candidate predictors, such as physiological and biochemical variables if available. The predictors identified in the current study could be incorporated using a model updating approach. Importantly, all future studies should include sample size calculation in their design. Routinely collected data and prospective cohort data both have respective strengths and both should be utilised to further refine predictive models in this setting. Ultimately, a prediction model to assess risk of mortality in AAV patients who experience a severe infection could help clinicians make more informed management decisions and enable high risk individuals to be identified for potential novel therapeutic strategies in clinical trials. This study is an important first step in achieving this important outcome for individuals with AAV, who remain at a much higher risk of severe infection, and related mortality, compared to the general population.(Sarica *et al.*, 2018)

5.7 Summary

This chapter described the development and internal validation of a prognostic model which aimed to predict early mortality following severe infection. The resulting model performed well on bootstrapped assessment of calibration and discrimination. Such models may assist with the identification of high-risk patients for inclusion in clinical trials of novel therapies for severe infection. Until this point, this thesis has examined severe infection as caused by all possible microbes, whether bacterial, viral or otherwise. The focus of the next two chapters will turn to a specific, novel infectious disease – Covid-19. Fundamental prognosis research and prognostic factor research on this disease will be reported.

6 Prognostic factors for severe outcome in individuals with systemic vasculitis and Covid-19

6.1 Overview

In the previous chapters, severe infection – a broad category of outcome in ANCA-associated vasculitis (AAV) – was evaluated. Incidence was reported, glucocorticoid exposure was assessed as a prognostic factor and prediction models were built. The remaining thesis chapters will focus on Covid-19, an infectious disease which caused a world changing pandemic. This will be done with continued focus on themes developed in earlier chapters of prognosis research methodology in AAV. As it transpired, Covid-19 was substantially more likely to cause severe disease in individuals with AAV and other systemic vasculitides. This chapter will summarise work undertaken early in the pandemic to report data on this group of special interest. An earlier iteration of this work was published in: Rutherford MA, Scott J, Karabayas M, *et al*. Risk Factors for Severe Outcomes in Patients with Systemic Vasculitis and COVID-19: A Binational, Registry-Based Cohort Study. *Arthritis and Rheumatology* 2021;73:1713–9.

6.2 Abstract

6.2.1 Background

Early in the Covid-19 pandemic, it was clear that the disease caused a wide range of severity, ranging from asymptomatic infection to death. Individuals with systemic vasculitis are highly vulnerable to severe infection, therefore understanding the impact of this novel virus in this population was essential. At the outset of the pandemic, there were no studies delineating the frequency of presenting clinical features, rate of complications or prognostic factors for severe disease. This study sought to report such data, including the prognostic properties of immunosuppressive therapies.

6.2.2 Methods

A multicentre registry-based cohort was established across two nations, through collaboration between the UK and Ireland Vasculitis Registry (UKIVAS) and the Ireland Rare Kidney Disease Registry (RKD). Patients who developed Covid-19 who had a pre-existing diagnosis of systemic vasculitis were included. Baseline clinical features and outcomes were reported. Individual logistic regression models were developed to identify prognostic factors for severe Covid-19 outcome. This endpoint was a composite of the need of advanced oxygen therapy, the need for invasive ventilation and mortality.

6.2.3 Results

Data for 105 patients was reported. Median age was 69 years and 43.8% were female. The majority of the cohort, 84 (80.0%) patients, had ANCA-associated vasculitis (AAV). Severe Covid-19 was experienced by 38 (36.2) individuals. Most patients required admission to hospital (84 of 105 [80.0%]), 15 (15.2%) were admitted to a critical care unit and 27 (25.7%) patients died. Existing treatment with any immunosuppressive agent was associated with severe outcome with adjusted odds ratio (aOR) 5.93 (95% confidence interval [CI] 1.76 – 27.71). Glucocorticoid exposure and cyclophosphamide exposure were also associated with severe outcome, with aOR 2.85 (95% CI 1.08 – 8.36) and aOR 3.45 (95% CI 1.08 – 11.98) respectively.

6.2.4 Conclusion

Exposure to any immunosuppression, glucocorticoid exposure and cyclophosphamide exposure were identified as prognostic factors for severe outcome in individuals with systemic vasculitis who developed Covid-19. These data may be able to inform prognostic models aiming to predict severe outcome in such a population.

6.3 Introduction

6.3.1 Background

At the end of 2019, reports emerged of a cluster of severe pneumonia cases of unknown cause linked to a seafood and animal wholesale market in Wuhan, a city in the Hubei Province in central China (Zhu et al., 2020). Subsequently identified as being caused by a coronavirus, the disease rapidly spread throughout the world. In February 2020 the disease was labelled coronavirus disease 2019 (Covid-19) by the World Health Organisation (WHO). Covid-19 resulted in a wide range of degrees of clinical severity, from asymptomatic infections to multiorgan failure and death (Zhou et al., 2020). In March 2020, WHO declared a global pandemic due to Covid-19. Widespread concern was evident within governments and reflected by the global media (WHO, 2020).

With the knowledge that individuals with AAV were already at high risk of severe infection of any cause, the necessity to understand the impact of this novel virus on the AAV population was apparent. Data relating to clinical features, natural history, rates of complications and prognostic factors for severe disease in this subset of patients were considered of immediate importance. The study team designed a cohort study to address these questions but also to deliver real-time updates to the UK and Ireland vasculitis clinical communities. These updates took the form of, initially weekly, short reports that were disseminated to interested clinicians.

Early reports described an excessive and dysregulated cytokine response, often described as a cytokine storm, in the setting of severe Covid-19 (Ye et al., 2020). Given that vasculitis patients are treated with potent levels of immunosuppression and with regimens that vary in their underlying mechanism of action, it was considered that interaction of Covid-19 with vasculitis patients would not only be important from a clinical perspective, but may also elucidate aspects of underlying immunobiology. It may have been the case that vasculitis patients would be more vulnerable to severe disease than the general population or that perhaps they may have a milder disease course due to immunosuppressive treatment dampening down overactive cytokine pathways.

6.3.2 Covid-19 biology

Coronaviruses are RNA viruses frequently encountered by humans and animals. Four coronaviruses are endemic in humans and typically result in symptoms of the common cold. Over the past 20 years, three zoonotic coronaviruses have crossed from animal reservoirs to cause transmissible infections in humans: severe acute respiratory syndrome coronavirus (SARS-CoV) in 2003, Middle East respiratory syndrome virus (MERS-CoV) in 2012 and SARS-CoV-2, the causative virus of Covid-19 in 2019 (Lamers and Haagmans, 2022).

The SARS-CoV-2 genome encodes structural proteins, such as spike protein (S) and nucleocapsid protein (N), non-structural proteins and accessory proteins. The structural proteins, together with host derived cell membrane, comprise the enveloped virion. Non-structural proteins are mostly involved in viral replication and transcription. Accessory proteins commonly have immunoevasive functions. The spike protein has two subunits: the S1 subunit which attaches to the angiotensin-converting enzyme 2 (ACE2) protein present on ciliated cells in the upper respiratory tract and alveolar type 2 cells in alveoli, the S2 subunit is then activated leading to fusion of virus and host bilipid layers. Viral RNA is then released into the host cell. Intracellular mechanisms can detect SARS-CoV-2 resulting in a signalling cascade leading to interferon transcription. Local epithelial and immune cells undergo activation of interferon-stimulated genes with resultant direct antiviral effects and recruitment of further immune cells. An important feature of SARS-CoV-2 pathogenicity, is that the virus prevents the infected cells from detecting viral RNA by hiding replication within “membrane-enclosed replication factories” within the cell. This can prevent a sufficient interferon response from occurring. Cytokine release facilitates the progression of adaptive B cell and T cell responses. Alveolar infection, where SARS-CoV-2 primarily infects AT2 cells, causes local inflammation and limits gas exchange. Importantly AT2 cells are responsible for surfactant production, which reduces surface tension and prevents collapse of alveoli (Lamers and Haagmans, 2022).

Transmission of the virus predominantly occurs via inhalation of droplets and aerosols containing virus (Comber et al., 2021). SARS-CoV-2 was highly transmissible compared to related human coronaviruses SARS-CoV and MERS-CoV. This is likely in part due to earlier peak viral titres in the upper respiratory tract

(Cevik et al., 2021). Sustained transmission in humans may have occurred due to the presence of the entry receptor (ACE2) in the upper respiratory tract epithelium. MERS was substantially less transmissible than SARS-CoV-2, this may be explained by the lack of expression of its respective entry receptor – dipeptidyl peptidase 4 (DPP4) – in the human upper respiratory tract (Meyerholz et al., 2016). (Lamers and Haagmans, 2022)

SARS-CoV-2 initially infects the upper respiratory tract. In mild cases, the initial immune response clears the virus. In more severe cases, it may be that virus produced in the upper respiratory tract is inhaled deeper into the lung to infect lower respiratory tract cells. Where severe Covid-19 occurs, histological examination reveals a pattern of lung injury known as diffuse alveolar damage. This pattern is characterised by features including alveolar oedema, microvascular thrombosis and pneumocyte death. Alveolar damage, whether caused directly by viral infection or local inflammation, leads to a “leaky state” of the alveolar epithelium and endothelium. This leaky state enhances inflammation and coagulation. Overactive coagulation is now well recognised as being present in severe Covid-19 and leads to systemic venous thromboembolism. Part of the underlying mechanism relates to neutrophil extracellular traps (NETs). In severe Covid-19 neutrophils express high levels of tissue factor and release NETs coated with tissue factor. NETs recruit platelets and in return platelets stimulate the formation of NETs – NETosis – this interaction further drives coagulation. In contrast to pneumonia caused by bacteria or other viruses, epithelial cell-derived IL-6 production has been identified as a phenomenon distinct to Covid-19. High levels of IL-6 is predictive of severe Covid-19 (Lamers and Haagmans, 2022). The RECOVERY platform trial showed the IL-6 blockade with tocilizumab reduced mortality (Abani et al., 2021). Data from RECOVERY also showed that glucocorticoid administration in the form of dexamethasone reduced mortality in severe Covid-19, presumably via pleotropic immunosuppressive effects (RECOVERY Collaborative Group et al., 2020). Overall, there is clear evidence that overactivation of inflammation, coagulation and the immune systems leads to severe Covid-19.

6.3.3 Covid-19 variants of concern

The initial months of the pandemic were characterised by less genetic adaptation and phenotypic variation compared to later (Walsh et al., 2020). The first major change, a non-synonymous mutation in the spike protein gene (D614G), has retrospectively been identified as present in specimens sampled in China in late January 2020. It was first identified in the UK at the end of February (Volz et al., 2021). Known as PANGO lineage B.1, this mutation conferred an approximate 20% growth advantage. Later in 2020 more highly mutated forms of Covid-19 emerged. Where such genetic changes caused sufficiently enhanced transmissibility, WHO designated such variants as variants of concern (VOC). There are several possible explanations for the origin of VOCs, with evolution within a chronically infected immunosuppressed human host or multiple hosts seeming most plausible.

Labelled as Alpha, Beta, Gamma, Delta and Omicron, these VOCs sequentially outperformed previous iterations of the virus and rapidly became dominant in the local, or global, population. Alpha and Delta VOCs respectively had 65% and 55% increased transmissibility over the variants they replaced. Omicron's evolutionary success can be attributed to more efficient viral entry, but also more efficient immune escape, resulting in effective infection of individuals with immunity to a previous variant. Disease severity was recognised as increased with Alpha, then further increased with Delta, but lessened with Omicron. Disease severity is, however, challenging to compare across variants due to variation in host factors such as increasing population immunity and individual severity factors such as comorbidities (Carabelli et al., 2023).

6.3.4 Covid-19 outcomes and prognostic factors

6.3.4.1 General population

Disease severity varies drastically and ranges from asymptomatic infection with SARS-CoV-2 through to severe Covid-19 associated with multiorgan failure and death. In February 2020, the Chinese Centre for Disease Control and Prevention published a case series of Covid-19 including 44,672, the largest at that time. The case-fatality rate (CFR), the proportion of individuals who die with confirmed infection, varied significantly depending on age. The overall CFR was

2.3%, but individuals aged 70 to 79 years had an 8.0% CFR, while those aged 80 years or over had a 14.8% CFR (Wu and McGoogan, 2020). However, many SARS-CoV-2 infections are mild and those with mild Covid-19 may not receive a diagnosis. Therefore, a more informative metric to estimate is the infection fatality ratio (IFR) – the probability of death of an infected individual. A large systematic analysis matched seroprevalence data to Covid-19 mortality rates across 190 countries and territories. The data used was up to the end of 2020 – before the vaccination era and the extensive emergence of VOCs. There was considerable variation in IFR according to age, time and geography. With respect to age, the modelling revealed a J-shaped relationship with the lowest IFR at age 7 years (0.0023%). There was then an exponential increase: age 30 years was associated with IFR 0.057%, age 60 years with IFR 1.00% and age 90 years with IFR 20.33% (COVID-19 Forecasting Team, 2022). As described above in section 6.3.3, different Covid-19 variants have varying disease severity, for example with the omicron VOC having a hazard ratio of 0.31 for death, when compared to delta (Nyberg et al., 2022).

The severity of Covid-19 is substantially influenced by a wide range of prognostic factors. As alluded to above, age has been recognised as one of the most important. Best represented by the infection fatality ratio (IFR; the probability of death of an infected individual), there is an exponential increase in the risk of death with increasing age such that the IFR doubles as age increases every 6-7 years. The strong influence of age is likely due to several factors associated with aging, with biological age and increased prevalence of comorbidities likely playing important roles (Zsichla and Müller, 2023).

Host genetics is recognised as contributing to Covid-19 severity. Relevant genes typically relate to aspects of Covid-19 pathogenesis: cell entry of SARS-CoV-2 (ACE2, TMPRSS2), respiratory surface barrier proteins (MUC1, LTZFL1) and immune system function (HLA, DPP9, TLR7). Other associated genes code for proteins involved in blood pressure regulation (ACE1), lipid metabolism (ApoE) and blood group (ABO). With the exception of some rare genetic variants, overall culprit genes mostly have a small effect on outcome (Zsichla and Müller, 2023).

Male sex increases the risk of severe Covid-19. There is some evidence however that post-menopausal women are at increased risk, independent of age. This

may be related to hormonal influence on viral entry into the host or host immunity. Pregnancy not only increases the risk of severe Covid-19 but also pregnancy-related conditions such as preeclampsia and adverse foetal outcomes such a intrauterine growth restriction, preterm birth and still birth (Zsichla and Müller, 2023).

Certain comorbidities are highly associated with severe Covid-19 including chronic obstructive pulmonary disease (COPD), hypertension, cardiovascular disease, chronic kidney disease (CKD), diabetes, obesity and cancer. Conditions where there is conflicting evidence include asthma, liver disease and certain mental health conditions. CKD is amongst the most strongly associated prognostic factors for Covid-19 hospitalisation and mortality. Frailty is a syndrome characterised by impaired exercise tolerance and reduced ability to withstand acute stressors. Age and multimorbidity both contribute to this syndrome. Large clinical studies demonstrate a prognostic effect of frailty for severe Covid-19 (Zsichla and Müller, 2023).

Various lifestyle factors appear to modify the effect of Covid-19. Regular physical activity and cardiorespiratory fitness are associated with attenuated disease severity. High levels of alcohol consumption are associated with severe Covid-19. The relationship between tobacco smoking and Covid-19 disease severity is complicated, with recent studies showing conflicting associations. The interaction between diet and Covid-19 is also complicated. Some studies show an association with a more benign course for the Mediterranean diet or plant-based diets (Zsichla and Müller, 2023).

Viral factors include the genetic variant, discussed above under Section 6.3.3. Various observations suggest that a high viral dose results in more severe disease (Zsichla and Müller, 2023).

Lower socioeconomic status has been associated with severe Covid-19. Various factors have been considered which may account for this including access to healthcare, nutrition and air pollution exposure. Some ethnic groups have been noted to have higher Covid-19 mortality. Most large studies have not shown an independent association for ethnicity with outcome, suggesting that variability in comorbidity prevalence and socioeconomic status may account for this

observation (Zsichla and Müller, 2023). Ultimately complex interactions will take place between many of the above-described prognostic factors.

6.3.4.2 Immune-mediated inflammatory disease

Individuals with immune-mediated inflammatory disease (IMID) represent a heterogeneous group with common factors resulting in vulnerability to severe infections. This includes susceptibility to viral infections including Covid-19. Underlying reasons which contribute to this risk include abnormalities intrinsic to the pathogenesis of such disorders, such as altered host immune defence, and immunosuppressive therapies frequently required to modulate disease activity. The risk of SARS-CoV-2 infection in certain IMIDs, notably rheumatoid arthritis (RA), was elevated in a large population-based study of almost 500,000 individuals with IMID from Ontario, Canada, while it was lower in others, namely inflammatory bowel disease and multiple sclerosis. This was despite an average 20% increase in testing across all IMID groups (Eder et al., 2023). Individuals with IMID were more likely to be hospitalised with Covid-19, with an adjusted odds ratio of 1.23 in one study (Eder et al., 2022). A matched cohort study found that individuals with RA had a higher risk of severe Covid-19 compared to controls (England et al., 2021). A data linkage study that utilised national health administrative data covering the whole of England showed that individuals with rare autoimmune rheumatic diseases had over twice the rate of Covid-19 related mortality compared to the general population (Rutter et al., 2022). An nationwide cohort study in England which utilised the OpenSAFELY data linkage platform found that having an IMID increased the risk of Covid-19 related mortality by 23% after adjustment for confounding (MacKenna et al., 2022).

Prognostic factors for severe Covid-19 in individuals with IMID reflect those described in the general population as described above in section 6.3.4.1. However important additional factors for this vulnerable population include immunosuppressive therapies such as disease-modifying antirheumatic drugs and glucocorticoids. The association with severe Covid-19 was only partially explained by comorbid conditions (Eder et al., 2022). Evidence from the COVID-19 Global Rheumatology Alliance (GRA) physician-reported registry evaluated 600 individuals with rheumatic disease and Covid-19 from 40 countries. This data showed that age and comorbidities, including hypertension or cardiovascular

disease, respiratory disease, diabetes and chronic kidney disease, were important prognostic factors for hospital admission. Certain immunosuppressive therapies were also associated with increased risk. Glucocorticoid at doses ≥10 milligrams (mg) per day prednisone equivalents showed an adjusted odds ratio of 2.05 for hospitalisation. Usefully it was reported that including disease activity in the model did not alter the direction or magnitude of this relationship in an important way. There was no association found for antimalarial therapy use or for non-steroidal anti-inflammatory drugs (NSAID). An inverse relationship was found for anti-TNF (tumour necrosis factor inhibitor) therapies, with an adjusted odds ratio of 0.40 (Gianfrancesco et al., 2020). Anti-TNF therapy with tocilizumab was also confirmed as a beneficial treatment for severe Covid-19 in the general population, with a reduction in the primary outcome of all-cause mortality, rate ratio 0.85 (Abani et al., 2021). In an analysis from the OpenSAFELY group, 'targeted therapies' were compared to 'standard systemic therapies' such as methotrexate, mycophenolate and ciclosporin in over a million individuals with IMID. They did not find evidence of differences in Covid-19 outcomes with most targeted immune-modifying treatments, but rituximab was associated with increased risk of Covid-19 related mortality (MacKenna et al., 2022).

6.3.4.3 Systemic vasculitis

Individuals with systemic vasculitides, such as ANCA-associated vasculitis (AAV) and giant cell arteritis, are vulnerable to severe infection and are considered even more susceptible relative to other individuals with IMID (Kitching et al., 2020). This is reflected in the data described in the thesis introduction. Reasons for this include altered host immunity, highly potent immunosuppressive therapy required to achieve control of disease activity, demographic factors and comorbid conditions. In the large Canadian population-based study of individuals with IMID above, individuals with vasculitis were noted to have the highest rate of hospitalisation with Covid-19 at 18 per 10,000 population. This corresponded to an odds ratio of 2.07 compared to matched controls (Eder et al., 2022).

One factor which may skew the interpretation of this data is testing. Testing was noted to be highest among individuals with vasculitis in another analysis of the Canadian population data of individuals with IMID (Eder et al., 2023), with an

odds ratio of 1.42 compared to non-IMID controls. This represents a form of selection bias whereby due to the increased risk posed by infection to vasculitis patients, they will be more likely to receive a test for Covid-19, likely driven by the desire of both patients and clinicians to achieve optimal and safe management. This may increase the denominator of metrics such as the CFR, falsely attenuating the association with severe disease. A further form of selection bias that likely increased apparent disease severity relates to the non-prospective nature of some reports, such as those from the Covid-19 Global Rheumatology Alliance (GRA) registry (Sattui et al., 2021). Patients were included in such datasets due to presenting to clinicians with significant symptoms, with the likelihood of patients presenting being associated with disease severity. For these reasons, case-series or registry data cannot give an accurate representation of a mortality metric such as IFR.

The GRA systemic vasculitis data showed that similar prognostic factors to other groups were important in systemic vasculitis, such as older age and comorbidities. More active disease and treatment with glucocorticoid ≥10 mg per day prednisolone equivalents were also identified as associated with more severe outcomes. Of particular importance for systemic vasculitis, evidence of a strong relationship between therapies typically used in AAV management was apparent. Rituximab and cyclophosphamide treatments were both associated with substantially increased levels of severe disease, with odds ratios of 2.2 and 4.3 respectively (Sattui et al., 2021). In the OpenSAFELY analysis, rituximab was associated with a 68% increased risk of death (MacKenna et al., 2022).

6.3.5 Rationale for this investigation

In early March 2020, there was widespread anxiety about the looming potential impact of the novel coronavirus. First reports of the virus spreading outside Asia were from the Lombardy region of Italy (Cereda et al., 2021). Health care systems in the region were overwhelmed. Concern was particularly apparent with clinicians who care for significantly immunosuppressed patients, such as those with systemic vasculitis. Our research group was well placed to collect data nationally across both the UK and Ireland. We recognised a need for rapid data collection, both to inform the practise of clinicians in real time using regular data reports and case vignettes, and to subsequently conduct analyses to

establish potential prognostic factors. By this time, the considerable immune system overactivation which drives severe Covid-19 was apparent (Ye et al., 2020). Due to the heterogeneous pattern of immunosuppression across the spectrum of systemic vasculitis, from interleukin-6 inhibition in GCA to B-cell depletion in AAV, we considered that prognostic factors identified could potentially also play a causal role in diminishing or augmenting Covid-19 disease activity. This may have informed the rapidly developing understanding of Covid-19 biology and may have provided signals regarding potential therapies.

One specific area of concern was the impact of the high doses of glucocorticoid commonly used to induce remission in vasculitis. The RECOVERY trial platform established that moderate-dose glucocorticoids had substantial benefit in patients from the general population admitted to hospital with Covid-19 and requiring oxygen therapy or mechanical ventilation, but showed potential deleterious effects when used in milder disease (RECOVERY Collaborative Group et al., 2020). However, glucocorticoids are also recognised as causing severe infections when used for more prolonged periods. This is most strongly evidenced in vasculitis in the PEXIVAS randomised controlled trial in AAV (Walsh et al., 2020). Taking into consideration these seemingly paradoxical effects, it was not known whether long-term glucocorticoid exposure would have a detrimental, neutral or protective effect in AAV patients with Covid-19.

6.3.6 Aims

The main aim of this study was to identify prognostic factors for Covid-19 disease severity in individuals with systemic vasculitis and confirmed Covid-19 in, what was at the time of initial analysis, the largest reported cohort of such patients (Rutherford et al., 2021). Additional aims were to describe the demographics of this cohort, the frequency of presenting symptoms, the frequency of complications and disease outcomes according to an international standard.

6.4 Methods

6.4.1 Data source, study design and setting

A registry-based multicentre cohort study was designed in March 2020. Clinical sites associated with the UK and Ireland Vasculitis Registry (UKIVAS; www.ukivas.org/) and the Irish Rare Kidney Disease Registry (RKD; www.tcd.ie/medicine/thkc/research/rare.php) were engaged. As of 2022, RKD was renamed the RITA-Ireland Vasculitis (RIV) Registry and Biobank after joining the Rare Immunodeficiency, AutoInflammatory and AutoImmune Disease (RITA) European Research Network. For consistency with the associated published article, it will continue to be referred to as RKD. UKIVAS had 89 participating centres while RKD had 8 participating centres. At the end of July 2020, there were approximately 7,400 individuals enrolled in the UKIVAS registry and 795 in the RKD registry. There was a small case overlap of four individuals across the registries. These figures are given to illustrate the scope of these registries, case ascertained for this study was not necessarily draw from the existing pool of subjects and cases could be submitted for inclusion without having previously been enrolled. Clinicians from these centres were asked to contribute cases. A vasculitis-specific Covid-19 digital case report form (CRF) was compiled. Collaborating clinicians returned the CRF by secure email to the study coordinators. A version of this document is attached as an appendix. Soon after this CRF was in use, new modules of the UKIVAS and RKD web-based data collection applications were designed to facilitate data capture.

The design of the CRF was underpinned by standardized biomedical ontologies such as SNOMED CT (Musen et al., 2012). A detailed discussion of the role of ontologies in medical research can be found in Chapter 2. The CRF was also designed to be interoperable with other international data sets which were emerging at that time, such as the such as the COVID- 19 GRA physician-reported registry (The COVID-19 Global Rheumatology Alliance, 2020). This would allow future data linkage.

For the UK, the Health Research Authority decision tool confirmed that ethical approval was not required for this study as the primary aim was to directly inform clinical care. The local sponsor determined the project represented

service evaluation (R&D reference no. GN20RH165). Ethical approval for the RKD registry had been previously confirmed by Tallaght University Hospital/St. James's Hospital Joint Research Ethics Committee (reference no. 2019- 08 List 29 [07]). All subjects in RKD provided informed consent for clinical data collection, therefore separate approvals were not required.

6.4.2 Participants

Subjects were eligible for inclusion if the individual had a clinician-confirmed diagnosis of systemic vasculitis and Covid-19. The vasculitis diagnosis was required to be consistent with the International Chapel Hill Consensus Conference Nomenclature of Vasculitides (CHCC2012) (Jennette et al., 2013). Covid-19 could be diagnosed by virology testing, radiologically or clinically. Recruitment began on 28 March 2020 and the last case was submitted in February 2021. The population sampling frame was made up of patients under the clinical care of centres represented in the UKIVAS and RKD registries.

6.4.3 Baseline characteristic data

A complete list of variables that were collected for this study is represented in the CFR in the appendix. With respect to baseline characteristic data, age in years at the time of Covid-19 diagnosis, sex and ethnicity data were gathered. Ethnicity was collected as subject-identified ethnicity according to standard NHS nomenclature, if available. Ethnicity was collapsed into broad categories – Asian, Black, White and Not stated. Smoking status was collected. The presence of various comorbid conditions was collected, detailed below in section 6.4.4. This was based on the Charlson Comorbidity Index (CCI) (Charlson et al., 1987). With respect to comorbid conditions, respiratory disease represents non-vasculitis related pulmonary conditions, however it was likely that some patients had additional pulmonary pathology caused by vasculitis.

Vasculitis specific data included a clinician impression of the presence or absence of active disease, this was on a four point scale with options 'remission', 'minimal or low disease activity', 'moderate disease activity', 'severe or high disease activity' or 'unknown'. Vasculitis disease duration in years was collected. Immunosuppressive therapy was ascertained as described in

the exposures section below. Data was collected exposure to angiotensin-converting enzyme (ACE) inhibitors, angiotensin II receptor blockers (ARB) and non-steroidal anti-inflammatory drugs.

Covid-19 data included clinical features at the time of diagnosis. The list of options was derived from the Covid-19 GRA case report form. Adaptations were made to also collect data on symptoms which could represent Covid-19 symptoms or vasculitis symptoms, such as haemoptysis. Laboratory data was gathered included creatinine, C-reactive protein (CRP) and lymphocytes at the time of Covid-19 diagnosis. Specific time windows for laboratory data were not set for simplicity of data collection. The methods for Covid-19 were collected, whether by polymerase chain reaction (PCR), radiological, clinical features or unknown.

6.4.4 Exposures

Variables selected as exposures for this study represented potential predictors of severe outcomes or already established predictors from other cohorts (Gianfrancesco et al., 2020; The OpenSAFELY Collaborative et al., 2020). Demographic factors incorporated included age and sex. Comorbid conditions investigated as exposures included: hypertension, cardiovascular disease (a collapsed variable consisting of atrial fibrillation, cerebrovascular disease, coronary heart disease, myocardial infarction, congestive cardiac failure and peripheral vascular disease), respiratory disease, diabetes, renal disease and end-stage kidney disease (ESKD).

Vasculitis diagnosis was examined as an exposure with respect to the most common types of AAV: granulomatosis with polyangiitis (GPA) and microscopic polyangiitis (MPA). For AAV, where CHCC2012 diagnosis was not specified, but ANCA autoantibody subtype was specified, this was collapsed as into two AAV categories: anti-proteinase-3 (PR3) antibody associated AAV was included with GPA and anti-myeloperoxidase (MPO) antibody associated AAV was included with MPA.

Detailed information on immunosuppressive therapy was collected. This included an overall immunosuppressive status with four categories: ‘Currently on

immunosuppression', 'Discontinuation of immunosuppression within 6 months prior to this encounter', 'Discontinuation of immunosuppression > 6 months prior to this encounter' and 'Treatment Naïve'. Presence of glucocorticoid was collected including dose in oral prednisolone equivalents. If individuals were exposed to glucocorticoid, any dose alteration at the time of the clinical encounter was collected. Other immunosuppressive treatments data included abatacept, cyclophosphamide (oral and intravenous), hydroxychloroquine, intravenous immunoglobulin, methotrexate, mycophenolate mofetil, rituximab, tocilizumab and calcineurin inhibitors such as ciclosporin and tacrolimus, and other immunosuppressive agents less commonly used in vasculitis. Immunosuppressants data collection was based on the GRA case report form with modifications to include typical immunosuppressants used in the treatment of systemic vasculitis. Intravenously administer immunosuppressants were labelled as "current" if the assessing clinician considered it likely that the drug was exerting a clinical effect at the time of Covid-19 diagnosis. For ease of data collection, specific guidance to estimate the clinical effect of IV immunosuppressants were not used. As for glucocorticoids, any dose change at Covid-19 diagnosis was sought. The immunosuppressive therapies that were ultimately evaluated for association with severe Covid-19 are shown in Table 6-3.

The referent for these exposures was typically the remainer of the cohort where the exposure was not present, for example the referent for the comorbid condition hypertension, was the group without reported hypertension. This is detailed in Table 6-3 and the rationale for this decision is elaborated on in the discussion.

6.4.5 Covariates

As described further in the statistical analysis section, age at Covid-19 diagnosis and sex were used as covariates in the models to determine association with severe Covid-19. Two covariates were chosen to limit the possibility of overfitting and therefore limit the chance of spurious associations. Notably age (in particular) and sex were emerging as important modifiers of the impact of Covid-19 at the time of study design.

6.4.6 Outcome

The main outcome was occurrence of severe Covid-19. This was derived from the seven-point GRA scale which is detailed in Table 6-2. This was a composite outcome: a binary version of this scale to represent severe Covid-19. The three most severe points were combined: requirement for non-invasive ventilation or high flow oxygen device, requirement for invasive ventilation or extracorporeal membrane oxygenation (ECMO), or death.

Occurrence of hospital admission and of intensive care unit (ICU) admission was collected. Potential complications collected in line with GRA data collection items included acute respiratory failure, acute kidney injury and disseminated intravascular coagulation. If admitted, date of hospital admission and discharge were collected. Length of stay was calculated.

6.4.7 Other variables

Other variables were collected that would inform the clinical phenotype of Covid-19 in vasculitis patients, that were possible outcomes of interest or that had the potential to inform future models.

Basic demographic and vasculitis diagnosis data, such as country of birth, date of vasculitis diagnosis, presence of vasculitis on biopsy and vasculitis diagnostic confidence were collected in the same manner as the UKIVAS main data collection tool. The categories selected for past vasculitis organ system involvement were based on the categories from the Birmingham Vasculitis Activity Score (BVAS, version 3) (Mukhtyar et al., 2009).

Covid-19 data collected included the date of Covid-19 symptom onset and the date of Covid-19 diagnosis. Data related to treatment of Covid-19 was based on the GRA data collection items. Data was collected relating to treatment of Covid-19 including antibiotics, antiviral therapy (such as remdesivir or neuraminidase inhibitors) or immunosuppressive agents (such as hydroxychloroquine or tocilizumab).

A optional second part of the CRF was labelled “Additional Information” and was visually distinct from the first, more pertinent section of the CRF. The option for

retrospectively collecting this data was highlighted. Information in this section included any additional databases that the case had been registered with, this was aimed at avoiding duplication of cases in the event of future merging the data with other projects. Information regarding employment history and educational status was sought, this was aimed at understanding the impact of Covid-19 on work and lifestyle, if future longitudinal data collection were possible.

Further data in the “Additional Information” section included vasculitis disease status at the time of Covid-19 diagnosis such as urinalysis results, last estimated glomerular filtration rate (eGFR) prior to diagnosis, weight and height. The formula used to calculate eGFR was not sought. Vasculitis disease activity was collected. This was recorded as the likelihood of relapse occurring at the time of diagnosis ranging from “high probability”, through “possibly” to not occurring. This was also intended to be adjudicated by a local senior clinician retrospectively, with options being “definite”, “high probability”, “possibly” and “not relapsing”. BVAS (version 3) and the vasculitis damage index (VDI) were requested, but were only expected to be returned if the completing clinician had undertaken training for these tools previously. Other data was gathered relating to Covid-19 diagnosis, laboratory investigations, secondary infections, requirement for acute dialysis and symptom resolution. These are detailed in the CRF in 9.4.

6.4.8 Bias

Ascertainment bias was recognised as an important consideration. Cases were identified by clinicians in secondary and tertiary care centres. Clinicians will have been more likely to encounter vasculitis patients with Covid-19 if the patient was unwell, given that the encounter may have taken place in hospital because the patient sought care due to their symptoms. Milder or asymptomatic cases may not have sought care and would have been less likely to be included. Therefore this study was does not provide an accurate indication of the frequency of severe Covid-19, either amongst diagnosed cases or all infected individuals. The outcomes are reported for the purpose of describing the study population, namely one with more severe disease.

Systemic vasculitis epidemiology is classically hampered due to the rarity of the conditions. This is more pronounced with a sub-population, such as individuals with both vasculitis and Covid-19. Spurious association of a potential exposure with the outcome may occur. This becomes less likely as sample size increases. To minimise spurious results the size of the cohort was maximised using multiple approaches. These included having three means of submitting data – manual completion of the CRF, digital completion of the CRF and submission of data to the UKIVAS web application. We publicised our project as much was deemed appropriate and feasible. We only undertook a preliminary analysis when the data set reached a reasonable size. Other issues relating to decisions around study size are discussed in section 6.4.9 below.

Confounding is where an observed relationship between variables, or a lack of relationship, may be explained by another common variable. We limited confounding due to age and sex by including these variables in the logistic regression models developed to assess potential prognostic factors. Two variables only were used to limit over fitting as described in section 6.4.11. Residual confounding may have occurred.

Due to the unknown biological interaction between systemic vasculitis and Covid-19, we may have failed to detect important findings purely due to not requesting certain data items. We attempted to mitigate against this by incorporating several options for inclusion of free text in the CRF.

We were conscious that we should make efforts to avoid non-completion or non-submission of cases. Specific effort was made to limit the number of data collection items and the complexity of data collection. This was intended to reduce the potential for bias where the busiest clinicians were unlikely to submit cases due to time pressure.

6.4.9 Study size

Formal sample size calculation was not undertaken for this study. Decision making with respect to required size for an initial analysis are explored in section 6.4.11.

6.4.10 Patient and public involvement

Specific patient and public involvement (PPI) work was not undertaken in advance for this project due to the urgent nature of the clinical issue and need for rapid data collection. The initial, and a subsequent, iteration of this study have been presented at UKIVAS and other international meetings. These meetings have included patients and patient representatives from organisations including Vasculitis UK, Vasculitis Ireland Awareness and Vasculitis International. The project received overwhelming support and positive feedback from vasculitis patients and patient representatives.

6.4.11 Statistical methods

At the time of study design, for the objective of compiling a formal report including statistical models, it was decided that when the study reached a suitable size for a basic adjusted model that the first analysis would be undertaken. When the dataset reached 65 cases, 25 (38%) of whom had experienced a severe outcome, this threshold was met. A pragmatic "rule of thumb" was applied, by which the ratio of events per variable (EPV) was greater than 10, meaning that for every 10 occurrences of severe Covid-19, one additional covariate can be added to the model. At the time of the first analysis of the data, the above EPV "rule of thumb" was approaching being met and due to the perceived clinical need for data around Covid-19 prognostic factors in rare immune-mediated inflammatory diseases (IMID), the first analysis was performed and subsequently published. The subsequent analysis for this chapter meets this criteria.

Continuous variables are reported as median and interquartile range (IQR). Discrete variables are reported as the number of individuals and percentage. Association between explanatory variables of interest and severe Covid-19 were calculated. Multiple unadjusted and age/sex-adjusted logistic regression models were used to determine the association for each explanatory variable. The output of these models was transformed to be reported as odds rations (ORs), *P* values and 95% confidence intervals (95% CIs). The adjusted odds ratios for age and sex were obtained from a single logistic regression model which included age and sex as the only explanatory variables. Potential interactions were

considered, if this may have accounted for a positive finding, then a logistic regression model including the explanatory and interacting variable was performed. When a subgroup may have had different overall findings, sensitivity analyses were performed in that group to check if results differed from the main findings. Missing data were acknowledged in the relevant tables. P values below 0.05 were considered statistically significant. Data cleaning and statistical analysis was performed with R (version 4.2.2) and packages including tidyverse and final fit.

6.5 Results

One hundred and five individuals were submitted as cases with a confirmed diagnosis of systemic vasculitis who developed Covid-19. Ninety-eight patients were registered as part of the UKIVAS registry and seven were registered as part of the RKD registry. There were no duplicate registrations.

6.5.1 Baseline characteristics

As described in Table 6-1, the median age of the study population was 69 years with interquartile range (IQR) 55 to 75. A minority of the cases were female at 43.8%. Most study subjects (84 of 105 – 80.0%) had anti-neutrophil cytoplasmic antibody associated vasculitis (AAV): of these, 40 (38.1) had GPA or PR3-associated AAV, 37 (35.2%) had MPA or MPO-associated AAV and 7 (6.7%) had eosinophilic granulomatosis with polyangiitis (EGPA). The baseline features of subjects with AAV were assessed and were similar to the full cohort (data not shown). Forty-seven subjects (50%) were considered by the submitting clinical team to have simultaneous active vasculitis at the time of Covid-19 infection. The median vasculitis disease duration was 2.7 years. Twenty-five subjects in the cohort had ESKD, this data was unknown/missing for 16 subjects (15.2%). Seventy-six subjects (72.4%) were diagnosed by polymerase chain reaction (PCR) or by serological methods. Five individuals (4.8%) did not have virologically confirmed diagnoses but instead had radiological confirmation of Covid-19. Four individuals (3.8%) were confirmed as having Covid-19 solely based on clinical features. Data relating to the means of diagnosis was unknown/missing for 20 individuals (19.0%). The baseline characteristics of study subjects who received a PCR based diagnosis were similar to the full cohort (data not shown).

Eighty-three individuals (79.0%) were receiving some form of immunosuppressive therapy at the time of Covid-19 diagnosis. Data relating to the presence or absence of immunosuppressive medications was missing in one individual (1.0%). The majority of subjects (71 of 105 [67.6%]) were treated with background glucocorticoid. Thirty subjects (28.6%) were treated with the equivalent of ≤5 mg prednisolone per day while 40 (38.1%) were treated with >5 mg per day. Data relating to the dose of glucocorticoid was unknown/missing in eight subjects (7.6%). Other baseline features including the presence of specific comorbid

conditions, other immunosuppressive therapies, other medications of interest (such as ACE inhibitors, ARB and NSAID) and laboratory tests at the time of diagnosis (such as serum creatinine, CRP and lymphocyte count) can be found in Table 6-1.

Table 6-1 | Baseline characteristics of study subjects (n = 105)

Characteristic	Value
Age (years), median (IQR)	69.0 (55.0 to 75.0)
Female sex	46 (43.8)
Ethnicity	
Asian	10 (9.5)
Black	3 (2.9)
White	73 (69.5)
Not stated	11 (10.5)
Missing data	8 (7.6)
Smoking	
Current	4 (4.6)
Former	19 (21.8)
Never	39 (44.8)
Unknown/missing data	25 (28.7)
Vasculitis diagnosis	
GPA (or PR3 AAV)	40 (38.1)
MPA (or MPO AAV)	37 (35.2)
EGPA	7 (6.7)
LVV	3 (2.9)
Behçet's	1 (1.0)
PAN	1 (1.0)
Other	9 (8.6)
Unknown/missing data	7 (6.7)
Comorbidities*	
Diabetes	18 (17.1)
Hypertension	38 (36.2)
Renal Disease	41 (39.0)
CV disease	28 (26.7)
Respiratory disease	27 (25.7)
End-stage kidney disease	
Yes	25 (23.8)
No	64 (61.0)
Unknown/missing data	16 (15.2)
Organ transplant	3 (2.9)

Characteristic
Vasculitis, active disea
Vasculitis disease dura
Unknown/mis
Current immunosuppr
Any immunosu
Any immunosu
Azathioprine
GCs (any dose
Predni
Predni
Unkno
Cyclophospha
Hydroxychloro
IVIG
Mycophenolat
Rituximab
Tacrolimus
Unknown/mis
Other medications
ACE inhibitor
ARB
NSAID
Unknown/mis
Laboratory tests
Creatinine
CRP
Lymphocytes
Method used for Covi
PCR/antibody
Radiological
Symptoms onl
Unknown/mis

AAV = ANCA-associated vasculitis (AAV), ACE = angiotensin converting enzyme, ARB = angiotensin receptor blocker, CRP = C-reactive protein, CV = cardiovascular, EGPA =

eosinophilic granulomatosis with polyangiitis, GCs = glucocorticoids, GPA = granulomatosis with polyangiitis, IQR = interquartile range, IVIG = intravenous immunoglobulin, LVV = large vessel vasculitis, mg = milligram, MPA = microscopic polyangiitis, NSAID = non-steroidal anti-inflammatory drug, PAN = polyarteritis nodosa, PCR = polymerase chain reaction.
Data is reported as number (%), unless specified.

6.5.2 Covid-19 symptoms

The frequency of Covid-19 symptoms in this cohort at presentation is displayed in Figure 6-1. Complete data in relation to symptoms was missing from five subjects (4.8%), missing data were removed from the calculation for symptom frequency. Dyspnoea was the most common presenting symptoms of Covid-19 in systemic vasculitis patients with 61 individuals (61.0%) reporting this symptom. This was followed by fever, at 53 individuals (53.0%) and cough at 50 individuals (50%). Haemoptysis occurred in six subjects (6.0%) and epistaxis occurred in three (3.0%). One patient had both haemoptysis and epistaxis. Notably some of the patients with haemoptysis had active disease at the time of Covid-19 diagnosis, as was the case for patients with epistaxis.

6.5.3 Covid-19 complications

The frequency of Covid-19 complications in this cohort is displayed in Figure 6-2. Complete data in relation to complications was missing from 10 subjects (9.5%). Respiratory failure was the most frequently reported complication in the cohort at 50 of 95 subjects (52.6%). The second most frequently reported complication was acute kidney injury at 16 of 95 (16.8%) subjects, followed by secondary infection at 14 of 95 patients (14.7%).

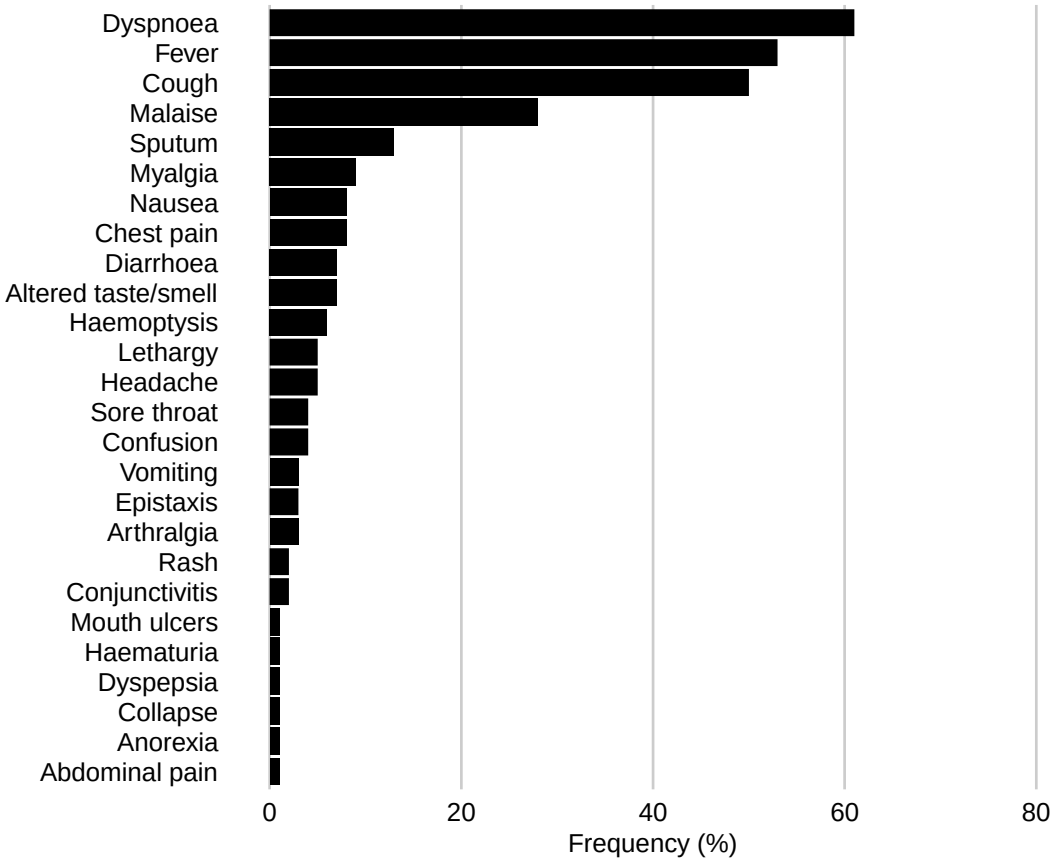


Figure 6-1 | Frequency of Covid-19 symptoms at first presentation

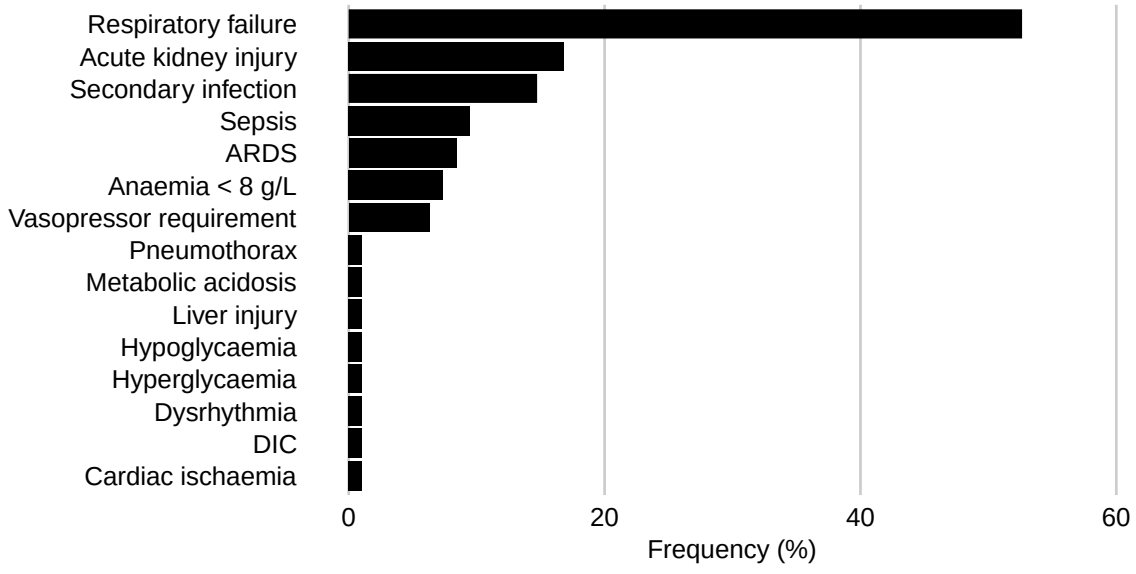


Figure 6-2 | Frequency of Covid-19 complications
ARDS = acute respiratory distress syndrome, DIC = disseminated intravascular coagulation.

6.5.4 Covid-19 outcomes

This cohort represents a severely affected group of patients, many of whom were likely identified once they had already become unwell and may have already been admitted to hospital. Full outcome data is summarised in Table 6-1, with unknown or missing data reported for each outcome. Most subjects were admitted to hospital (84 of 105 [80.0%]). Fifteen (15.2%) required admission to critical care. Twenty-seven (25.7%) subjects died. The median length of hospital stay was 11 days. The composite severe outcome was experienced by 38 of 105 subjects (36.2%).

Table 6-2 | Covid-19 outcomes in study subjects

Outcome	Value n (%)
Hospitalised	
Yes	84 (80.0)
No	18 (17.1)
Unknown/missing data	3 (2.9)
Critical care admission	
Yes	15 (15.2)
No	77 (77.8)
Unknown/missing data	7 (7.1)
Graded outcome (grade number)	
Not hospitalized, no limitations on activities (1)	9 (8.6)
Not hospitalized, limitation on activities (2)	7 (6.7)
Hospitalized, not requiring supplemental oxygen (3)	12 (11.4)
Hospitalized, requiring supplemental oxygen (4)	33 (31.4)
Hospitalized, on non-invasive ventilation or high flow oxygen devices (5)	6 (5.7)
Hospitalized, on invasive mechanical ventilation or ECMO (6)	5 (4.8)
Death (7)	27 (25.7)
Composite severe outcome	**38 (36.2)**
Unknown/missing data	6 (5.7)
Median length of hospital stay, days (IQR)	11 (5-22)
Length of hospital stay, unknown/missing data	74 (70.5)

6.5.5 Prognostic factors for severe outcome

Exposures examined are summarised in Table 6-3, with the number of severe outcomes for each exposure, along with unadjusted and age/sex adjusted models for severe outcome. Receiving any immunosuppressive treatment, when compared to receiving none, showed an association with severe outcome with adjusted odds ratio (aOR) 5.93 (95% CI 1.76 – 27.71). Glucocorticoid at any daily prednisolone dose equivalent carried aOR 2.85 (95% CI 1.08 -8.36), while glucocorticoid at daily prednisolone dose equivalent greater than 5 mg per day had aOR 4.66 (95% CI 1.47 – 17.30). There was not a significant association for lower dose glucocorticoid at daily prednisolone dose equivalent between 1 to 5 mg aOR 3.02 (95% CI 0.91 – 11.16). Treatment with cyclophosphamide was associated with severe outcome, aOR 3.45 (95% CI 1.08 – 11.98).

An associated for various exposures was not found. Age was not associated with severe disease in the adjusted model with aOR 1.02 (95% CI 1.00 - 1.06, p = 0.10). Significant associations were not found for sex, vasculitis diagnosis (whether GPA or MPA), any specific comorbid condition or smoking status. Other immunosuppressive therapies evaluated included azathioprine and rituximab, significant associations were not found with respect to severe disease for either of these therapies.

Table 6-3 | Unadjusted and age/sex adjusted ORs for association with severe Covid-19

	Number of severe cases / total cases (%)	Unadjusted OR (95% CI)
Sex		
Male (referent)	23/59 (39.0%)	-
Female	16/46 (34.8%)	0.88 (0.50 - 1.55)
Age	-	1.02 (1.00 - 1.06)
Vasculitis diagnosis		
GPA (referent: not GPA)	15/41 (36.6%)	0.96 (0.42 - 2.16)
MPA (referent: not MPA)	13/35 (37.1%)	1.00 (0.43 - 2.30)
Comorbidities (referent: individual comorbidity not present)		
Hypertension	18/38 (47.4%)	1.59 (0.89 - 2.86)
CV disease	13/28 (46.4%)	1.46 (0.78 - 2.72)
Respiratory disease	14/27 (51.9%)	2.28 (0.94 - 5.64)
Diabetes	9/18 (50%)	1.55 (0.75 - 3.24)
Renal disease	17/41 (41.5%)	1.22 (0.68 - 2.16)
End-stage kidney disease	11/25 (44%)	1.40 (0.54 - 3.59)
Smoking status		
Ever smoker (referent: never)	11/23 (47.8%)	2.33 (0.80 - 6.97)
Immunosuppressive therapy		
Any immunosuppressive therapy (referent: not receiving immunosuppressive therapy)	36/83 (43.4%)	4.85 (1.51 - 21.78)
GCs (referent: not receiving GCs)		
Prednisolone (any dose)	32/71 (45.1%)	2.58 (1.01 - 7.23)
Prednisolone 1.0-5.0 mg/day	13/30 (43.3%)	3.36 (1.05 - 12.22)
Prednisolone ≥5.1 mg /day	19/40 (47.5%)	3.98 (1.33 - 13.79)
Other immunosuppressive therapy		
Azathioprine (referent: not receiving azathioprine)	6/18 (33.3%)	0.80 (0.26 - 2.28)
Cyclophosphamide (referent: not receiving cyclophosphamide)	9/15 (60%)	2.95 (0.97 - 9.54)
Rituximab (referent: not receiving rituximab)	15/36 (41.7%)	1.31 (0.57 - 3.00)

GCs = glucocorticoids, CV = cardiovascular, GPA = granulomatosis with polyangiitis, mg = milligram, MPA = microscopic polyangiits, OR = odds ratio

6.6 Discussion

6.6.1 Key results

At the time of publication of the associated article, this cohort was the largest reported study of individuals with systemic vasculitis and Covid-19 (Rutherford et al., 2021). That report included 65 cases. This updated version of the same study reports 105 cases. A broad range of systemic vasculitides are represented, though with predominance of AAV – 79% of the reported cases had AAV. Twenty-five individuals, 23.8%, had ESKD at the time of Covid-19 diagnosis. A high proportion of the cohort had active disease at the time of reporting – 47 individuals, representing 50% of cases. A large majority, 79%, were actively treated with some form of immunosuppressive therapy. For most this included glucocorticoid, but other typical vasculitis therapies such as cyclophosphamide, rituximab, azathioprine and mycophenolate were also represented. Most of the cohort, 72.4%, had Covid-19 confirmed virologically. For the vast majority this was PCR-based diagnosis.

The most common presenting symptoms were dyspnoea, fever and cough. Symptoms more specific to vasculitis such as haemoptysis and epistaxis were present at lower frequencies. The most common complications of Covid-19 in this cohort were respiratory failure, acute kidney injury and secondary infection. A large majority, 80%, of the cohort was hospitalised in the context of Covid-19, 15.2% required critical care admission. The composite severe outcome was reached in 38 individuals (36.2%). This outcome comprised: requirement for advanced oxygen therapy, requirement for invasive ventilation or ECMO, or death.

Multivariable analysis was conducted, with adjustment for age and sex, on exposures of interest. Age and sex were not statistically associated with the composite severe outcome, but our study was likely underpowered to detect these potential relationships. Similarly, the various comorbid conditions examined had positive point estimates for association with severe disease, but had wide confidence intervals, so a range of potential effects remain possible. When examined as a group, individuals receiving any immunosuppressive therapy appeared to have a higher risk of severe outcome when compared to those not

receiving any immunosuppressive therapy, with aOR 5.93 (95% CI 1.76 – 27.71). For those receiving glucocorticoid, the aOR for severe outcome was 2.85 (95% CI 1.08 – 8.36). With respect to glucocorticoid exposure within specific dose ranges, there was no significant association but a wide confidence interval for daily prednisolone equivalents 1 mg to 5 mg (aOR 3.02 [95% CI 0.91 - 11.16]) and ≥5.1 mg was associated with severe outcome (aOR 4.66 [95% CI 1.47 - 17.3095% CI). Cyclophosphamide was associated with severe outcome (aOR 3.45 [95% CI 1.08 - 11.98]). Rituximab has a point estimate that suggested association with severe outcome, but was not statistically significant (aOR 1.57 [95% CI 0.66 - 3.79]).

6.6.2 Strengths

This was a multicentre study across two nations. Despite the early stages of the Covid-19 pandemic representing an unprecedented level of pressure on clinicians across healthcare systems, health care professionals submitted detailed reports on systemic vasculitis patients with Covid-19. This resulted in high quality data with low levels of missingness, given the circumstances. Centres covered a large spectrum of UK and Ireland clinical services with good representation from academic institutions and district general hospitals. Coverage included both urban and rural facilities. The high-quality nature of the data and wide coverage was facilitated by decisions made by the coordinating team, the UKIVAS Covid-19 task force. Data could be reported by the CRF, whether completed manually or digitally, and via the standard web-based data collection tools of UKIVAS and RKD. Intentionally, the CRF left aspects of reporting to the discretion of the submitting clinician, for example we did not provide exhaustive criteria for the different comorbidities that had to be fulfilled to determine whether any specific condition was present or not. As well as an optional BVAS form, submitting clinicians were invited to submit an overall impression of the presence or absence of active disease, via four intuitive categories based on the likelihood of active disease. This was intended to minimise effort required on the part of the submitting clinician and to maximise submission and completion of CRFs. UK and Ireland vasculitis clinicians were engaged through, initially weekly, reports sent to individuals signed up to the UKIVAS mailing list. These reports included up-to-date statistics on the cohort as well as anonymised case vignettes with clinical reflections on how to manage Covid-19 in systemic vasculitis patients. Having more than one method of data submission was

pragmatic and the approach to keeping clinicians updated with useful clinical information was both valuable to clinicians and maintained a high level of engagement with case submission.

As discussed, at the time of writing the earlier published iteration of this study, this cohort was the largest known of individuals with systemic vasculitis and Covid-19. Systemic vasculitides are rare, representing a significant challenge for all forms of research including vasculitis epidemiology. We used strategies described above to maximise case submission and engagement, balancing this with being mindful of the pressures that clinicians were facing in the early stages of the pandemic. Aiming for as large a study as possible in this setting maximised the chances of detecting important prognostic associations whilst minimising the chance of spurious results. Whilst a success for a novel virus in a set of rare diseases, the study remains small in the context of other similar studies in the general population such as the International Severe Acute Respiratory and emerging Infections Consortium (ISARIC) WHO Clinical Characterisation Protocol UK (CCP-UK) study (Docherty et al., 2020). This is discussed further below. When developing multivariable models to evaluate for prognostic exposures, it is important to consider the appropriate number of covariates included in models to limit overfitting. The maximum number of parameters included in our model was three (the exposure – consistently a two-level categorical variable, age and sex) and the number of events was 38. This equates to an EPV of 12.7. Whilst more complex means of calculating the required number of events and overall size of a data set required for sound development of a multivariable prediction model exist, simulation studies suggest that an EPV of the magnitude used in our study should result in reliable estimates of prognostic ability (Riley et al., 2020; Austin and Steyerberg, 2017). Given the challenge of achieving a large sample size in rare disease research, meeting expectations of a reasonable EPV in this setting is a significant strength of this work. It also allowed the inclusion of additional parameters, such as disease activity, in sensitivity analyses to confirm that the detected associations were not confounded by such factors. Adding an additional two-level categorical variable resulted in a EPV of 9.5.

A wide variety of data points were available for most participants in this study. This advantage is derived from study design. Being able to prospectively design

the dataset enabled a wide range of variables to be included. This covered demographics, vasculitis diagnosis, comorbidities, smoking status, vasculitis disease activity, ANCA serological status and medications. This range of baseline characteristics would be difficult to obtain from a retrospective review of clinical records. It also would not have been possible for the study team to access clinical records retrospectively from other health care providers. There is no comparable data resource available from routinely collected data which might be accessible using a data linkage approach. We considered our approach optimal for achieving a rich data resource.

As discussed below, ascertainment bias will have played a role in this study, in that individuals with more severe Covid-19 will have been more likely to have been included as subjects than individuals with milder disease. This has both beneficial and adverse impacts on the utility of the study. Reasons for this having occurred include that individuals who are more unwell are more likely to seek medical attention and are therefore more likely to encounter a clinician who might submit to a study. In one sense this is useful in that it results in this study being more likely to have a high number of clinical outcomes and therefore likely to be adequately powered to detect meaningful associations. There is ultimately greater utility in studying individuals more likely to have severe Covid-19, as by definition, they are a group who are more likely to have greater unmet need. One of the negative impacts of such ascertainment bias is that this data cannot be used to give an indication of the incidence of Covid-19 outcomes for the whole systemic vasculitis population. For this reason it has been clearly specified that it would be incorrect to interpret this data in such a manner. If a study were to accurately quantify the incidence of severe Covid-19 in a population, it would need to have a means of, ideally prospectively, identifying a sample of all cases of clinically identified Covid-19 to determine, for example, the CFR, or a means of identifying a sample of all individuals screened as positive for SARS-CoV-2, to determine the IFR.

6.6.3 Limitations

This study had a large preponderance of small vessel vasculitis (SSV), especially AAV, relative to other vasculitides, such as large vessel vasculitides (LVV) including GCA. GCA is the most common idiopathic systemic vasculitis, an

American study indicated a 1% lifetime risk of acquiring the condition in women with 0.5% life time risk in men (Crowson et al., 2011). The incidence of AAV is far lower, with approximately 20 cases being diagnosed per million person-years (Kitching et al., 2020). Therefore, it may have been expected that the frequency of GCA patients in this study would closer reflect the real-world frequency. However, 80% of the cohort in this study had AAV, while only 2.9% had LVV. This reflects a strong effect of ascertainment bias, likely secondary to a variety of causes. The centres affiliated with the UKIVAS registry are predominantly nephrology centres, traditionally nephrologists are not involved in the care of GCA patients. AAV patients typically are under more intense immunosuppression, this may have made them more susceptible to SARS-CoV-2 infection and also more susceptible to having significant Covid-19 symptoms and severe disease – all these factors would make them more likely to present to, or to come to the attention of, a clinician participating in data collection for this study. The ultimate effect of this type of ascertainment bias is mixed. A predominance of AAV patients results in this data being more generalisable for that population, while the implications of the data are less clear for the highly heterogeneous populations of non-AAV vasculitides.

Regardless of any degree of ascertainment bias, the heterogeneous nature of this cohort remains a significant limitation. The vasculitides are a diverse range of conditions. They are challenging to define. Formal diagnostic criteria are lacking (Kronbichler et al., 2023). We did not provide strict criteria for inclusion, advising for vasculitides consistent with the CHCC to be included (Jennette et al., 2013). This was a pragmatic approach and due to the complexity of vasculitis diagnosis, it seems that adding extensive inclusion and exclusion criteria based on vasculitis diagnostic or classification criteria would have been unlikely to improve the correct identification of cases. While some common features are shared, such as a common immunosuppressant across most vasculitides being glucocorticoids, these diseases have varied pathogeneses and often affect completely different organ systems. The case can be made that collecting data on this heterogeneous cohort may have helped reveal specific “at risk” groups that would otherwise have gone unidentified. The study did indeed identify certain immunosuppressants associated with increased risk, but it was not possible to make this association for rarer vasculitides such as polyarteritis

nodosa (PAN) and Behçet's, due to the low numbers of patients with these diagnoses included in this study.

Lack of power is a recurrent challenge in the rare disease setting, including in systemic vasculitis research. We describe above strategies used to minimise the difficulty of case submission for contributing clinicians and to maximise engagement with the project, whilst being sensitive to the stress front-line clinicians were under at that time. Power was sufficient to allow pragmatic and statistically valid development of models to adjust for important confounders, namely age and sex, whilst aiming to minimise the risk of overfitting and spurious results. However due to the ultimate size of this cohort and the number of events, confidence intervals were typically wide, effect sizes may have been exaggerated and subtle associations may not have been detected.

The choice of referents in this study had both strengths and limitations. In this setting the referent is synonymous with a "control group" for the individual model. In epidemiological studies, control groups should typically satisfy two criteria: the presence of potential confounding variables should be the same in both the exposure and unexposed group and ascertainment of the presence of the exposure should be accurate in both the exposed and unexposed groups (Coggon et al., 2003). As far as is practically possible, it is the case that these requirements are met for the selected referent groups in this study, though there are important considerations which impact interpretation. With respect to vasculitis diagnosis and specific comorbid conditions, the referent was those who did not have the specific condition. Similarly the referent category for immunosuppressive therapies was those individuals not exposed to that specific therapy. While this will have maximised statistical power and would permit any one condition or therapy to "stand out" from the pool of predictors, arguably the referent categories should have been different. For vasculitis and comorbid conditions, one can pose different research questions. In this study we compare a specific condition to all others in the study, not to a comparable control without that condition. For example, for the vasculitis diagnosis GPA, the question we posed was to determine the risk of severe disease when compared to a mix of other vasculitides, including MPA and EGPA. Arguably a more important question would be to explore the risk of severe disease in GPA when compared to a general population control without any form of vasculitis.

Unfortunately due to the nature of our study not including such controls this was not possible. Out approach is still valid, but must be interpreted as risk relative to a mixed group of other vasculitides. A similar position should be adopted to comorbidity association interpretation. It may have been possible in our study to have taken this alternative approach with respect to comorbid conditions, for example, instead of comparing individuals with hypertension to all those without hypertension, we could have compared those with hypertension to individuals with vasculitis but no additional comorbid conditions. This would potentially have allowed the odds ratio to be more generalisable for the specific comorbidity and may be easier to interpret, but would likely have been significantly hampered by a lack of events leading to low power and very wide confidence intervals. Similarly for treatment, the appropriate referent category would potentially be those in the cohort not receiving any immunosuppression. In general for this issue, adopting an interpretation where by the risk reported for each exposure is relative to the rest of the cohort rather than relative to no exposure being present is important. For example, being on azathioprine is unlikely to be a protective predictive factor for severe Covid-19 in this population, but is associated with reduce risk relative to the rest of the cohort. This approach was taken to maximise statistical power and to have clear, simple statistical models, although the case can be made that the interpretation becomes more difficult.

6.6.4 Interpretation and generalisability

The symptom distribution described in this cohort broadly reflects that of the general population. In the current study the most frequent presenting symptoms were dyspnoea, fever and cough. This was similar to the ISARIC WHO CCP-UK study – the largest UK study of individuals from the general population hospitalised with Covid-19 (Docherty et al., 2020). Notably 6% of our cohort experienced haemoptysis at the time of Covid-19 presentation. None were considered likely to have diffuse alveolar haemorrhage, a potentially life-threatening feature of systemic vasculitis. Haemoptysis is a recognised feature of Covid-19 in the general population, with 3.5% of the ISARIC WHO CCP-UK experiencing it. It may be that the symptom was more common in our cohort due to pre-existing lung damage that was aggravated by SARS-CoV-2 infection or active pulmonary vasculitis that was not clinically apparent. Differentiating

Covid-19 related haemoptysis from active respiratory disease due to vasculitis is clinically challenging. It is also important to note that these presentations may coexist. Clinicians should ensure both causes are considered when assessing a patient with systemic vasculitis and possible, or confirmed, Covid-19.

A substantial proportion of the cohort (50%) had concurrent active vasculitis at the time of Covid-19 diagnosis. Many of these individuals were considered to have acquired SARS-CoV-2 from a health care facility, as reported by submitting clinicians. Crucially, vasculitis disease activity occurred before Covid-19 in essentially all cases. This study therefore did not provide an indication that vasculitis activity could be triggered by Covid-19. There are reports of cases of systemic vasculitis in adults considered to have been triggered by Covid-19 but establishing the true underlying relationship between infection and subsequent vasculitis is challenging. A clearer phenomenon was the emergence of multisystem inflammatory syndrome in children (MIS-C). This was typically described as highly similar to an incomplete form of Kawasaki disease (KD), though it tended to occur in school-age children, as opposed to classic KD which tends to affect children below the age of five years (Mv et al., 2023). The frequency of active disease in the current study was higher than expected. UK cohorts have previously shown typical disease activity of approximately 20% (Basu et al., 2014). Disease actively was not shown to be associated with severe Covid-19 in this study, but it may be that those individuals with active disease are more susceptible to infection with SARS-CoV-2. This may be due to the underlying biology of active vasculitis but it could also be the case that clinicians or patients may have reduced immunosuppressive therapy to attempt to minimise the impact of Covid-19. Due to the nature of this study, the current data cannot address this question.

When compared to studies of individuals with IMID described in the introduction to this chapter, the point estimates in the current student were consistent with respect to the prognostic associations for age, sex and comorbidities. The findings in our study were not statistically significant, but the direction and magnitude of the association was similar (Gianfrancesco et al., 2020). This study demonstrated that, when taken as a combined group, any immunosuppression was associated with substantially increased risk of severe Covid-19 compared to those not on immunosuppression. When compared to a similar study from Covid-

19 GRA of individuals with systemic vasculitis or polymyalgia rheumatica, our results were consistent – although they did not report findings for any immunosuppression as a combined group, they did find increased risk for a broad range of immunosuppressive agents including glucocorticoid equivalent to 10 mg prednisolone daily or more, rituximab and cyclophosphamide. Our data was also compatible with that study with respect to individual immunosuppressants: azathioprine was not clearly associated with severe Covid-19, rituximab was likely associated (statistically significant association in GRA data, consistent point estimate but not significant in the current study) and cyclophosphamide was most strongly associated with severe disease (aOR 4.30 [95% CI 1.10-16.75] in GRA, aOR 3.45 [95% CI 1.08-11.98] in the current study). Interestingly, in the GRA systemic vasculitis or polymyalgia rheumatica study, higher doses of glucocorticoid were associated with severe Covid-19 in GCA and AAV, but not PMR. This may reflect underlying biology or more intense immunosuppressive with additional agents frequently required in GCA and AAV (Sattui et al., 2021).

In the previous iteration of this study, published in 2021, comorbid respiratory disease and glucocorticoid use (at any dose) were found to be associated with severe Covid-19. In that earlier version of the study, which examined 65 cases, respiratory disease carried aOR 7.53 (95% CI 1.93-38.22) and glucocorticoid (at any dose) had aOR 3.66 (95% CI 1.09–14.9). With greater statistical power in the current study, the association for respiratory disease has been attenuated considerably to a non-statistically significant aOR 2.21 (95% CI 0.90-5.54) and glucocorticoid at any dose has been moderately attenuated to aOR 2.85 (95% CI 1.08-8.36). This represents the importance of increased power to limit the impact of spurious findings (Rutherford et al., 2021). With added power in the current study, it was possible to establish that a dose threshold for glucocorticoids likely exists: risk was not clearly present at doses up to 5 mg per day prednisolone equivalents, but was present above this, though the precise threshold at which risk increases and the shape of such a relationship remains to be determined.

For our study and others, it is important to view these findings as associations, which can also be considered prognostic. Determining causal associations was not a stated aim of the study, however it is possible that associations detected may represent causality. While these data may support this, studies with specific

causal inference methodology should be used to provide clearer evidence. For the immunosuppressants associated with increased risk, a clear potential confounder is disease activity, though including this in the models did not substantially change the direction or magnitude of any reported association. Glucocorticoids were reported in the current study as prognostic for severe disease. Glucocorticoids have a multitude of effects across the immune system and are considered to lead to infection susceptibility (Cain and Cidlowski, 2017). High dose glucocorticoids are reported to be associated with increased SARS-CoV-2 viral shedding duration, similar to other coronaviruses (Johnson and Vinetz, 2020; Li et al., 2020). The glucocorticoid finding may appear to be in conflict with the results of the RECOVERY platform trial of dexamethasone (RECOVERY Collaborative Group et al., 2020). The RECOVERY trial showed that low-dose dexamethasone conferred reduced mortality risk in unwell patients hospitalised with Covid-19. Importantly, however, the groups that derived benefit in this trial were those who needed supplemental oxygen therapy. The point estimate for individuals who did not require additional oxygen indicated that dexamethasone treatment potentially could result in increased mortality, though this was not a statistically significant finding. It may be that, during the earlier stages of Covid-19, before excess immune system activation, that glucocorticoid are harmful, as reported in the current study and other cohorts of individuals with IMID. There is limited literature that elaborates on potential mechanisms for the contribution of cyclophosphamide to severe Covid-19, but this effect is not unexpected given the well-recognised risk of infections with cyclophosphamide-based treatment regimens in AAV (Kronbichler et al., 2015).

These data are generalisable to individuals with systemic vasculitis, particularly AAV, and severe Covid-19. Important caveats include that the large majority of individuals in this study had AAV, so the implications of the data for those with non-AAV vasculitis are less clear. It is also necessary to take into consideration the severity of Covid-19 in this cohort. Many individuals, including some with systemic vasculitis, who experience Covid-19 will have a mild disease course. The severe disease experienced by this group is reflected in the high proportion of hospitalisation at 91%. Due to study design, it is not possible to determine the incidence of either Covid-19 in vasculitis patients or the incidence of severe disease following infection with SARS-CoV-2 in vasculitis patients.

6.7 Conclusion

This study was the first to systematically describe a group of patients with systemic vasculitis who contracted Covid-19. The presenting clinical features reflected those of the general population without IMID. Glucocorticoids and cyclophosphamide were associated with severe Covid-19, but the impact of other immunosuppressants was less clear. Our findings were consistent with a possible effect of rituximab, reported in other subsequent studies. Azathioprine appears to be less significant in its association with severe Covid-19, also reflected by subsequently published literature. During the Covid-19 pandemic, governments worldwide advised those considered most vulnerable to severe Covid-19 to adhere to stringent social isolation guidelines. This included those with systemic vasculitis. These data highlight that predictive variables exist, which in combination with other data, could better inform individual risk. In future, risk stratification should be considered to help guide the management of vulnerable individuals with respect to their individualised risk of severe infections, such as Covid-19.

6.8 Summary

This chapter characterised Covid-19 in a severely affected group of individuals with an established diagnosis of systemic vasculitis. Data was collected by dedicated clinicians across the UK and Ireland. Symptom and complication profiles were established. Potential prognostic factors were identified: any immunosuppressive therapy, glucocorticoids and cyclophosphamide. The next chapter will seek to explore the underlying biology of a highly important and novel prognostic factor – SARS-CoV-2 vaccination in individuals with AAV.

7 Immunological response to SARS-CoV-2 vaccination in individuals with ANCA-associated vasculitis treated with rituximab

7.1 Overview

In this thesis so far, it has been established that severe infections are common in individuals with AAV, certain parameters such as the presence of immune-suppressing medication such as glucocorticoids have the potential to be utilised as predictive factors for severe infection and that multivariable predictive scores can be developed to predict severe infection related outcomes. In the previous chapter, the focus shifted to Covid-19, a novel infection with severe implications for individuals with vasculitis. Potential predictive factors for severe disease were explored. In this chapter, the antibody and cellular immune response to SARS-CoV-2 vaccination will be explored – factors with potential predictive ability in the vaccination era.

7.2 Abstract

7.2.1 Background

SARS-CoV-2 vaccination has been shown to be highly effective with respect to reducing Covid-19 incidence, severe Covid-19 and Covid-19 related death in the general population. The effectiveness of vaccination in vulnerable populations, such as those exposed to immunosuppressive therapies for chronic conditions, is unknown. Rituximab is frequently used for treatment of AAV, but impaired humoral response following influenza vaccines has been reported with this therapy and therefore concerns exist with respect to immunogenicity to SARS-CoV-2 vaccines. In this study, the immune response to the ChAdOx1 (Astra Zeneca) vaccine in rituximab treated AAV patients was evaluated.

7.2.2 Methods

This study reports data from participants of the OCTAVE trial who had AAV, all of whom were exposed to rituximab. OCTAVE was a multicentre, multi-disease prospective cohort trial which included participants with various immunosuppressed diseases. All participants received a SARS-CoV-2 vaccination as part of routine NHS care. Blood was sampled for IgG response to SARS-CoV-2 spike antigen (anti-S) and IFNγ T cell responses to SARS-CoV-2 antigens at baseline (where possible), immediately before the second SARS-CoV-2 vaccine dose and 28 days post-second dose. A control group included participants from the UK PITCH (Protective Immunity from T cells in Healthcare workers) study.

7.2.3 Results

Of 2,686 cases recruited to OCTAVE, 30 had AAV and 225 were healthy controls. Median age of the AAV group was 55.3, 15 (50.0%) were female. After the second SARS-CoV-2 vaccine dose 27.6% (8/29) of AAV patients achieved anti-S seropositivity, compared to 98.7% (222/225) of healthy controls. Three (10.3%) individuals with AAV achieved a titre greater than that of the lowest decile of healthy controls. After the second dose, the median SARS-CoV-2 specific T cell response in AAV participants was 104 IFNγ secreting T cells / 10^6 peripheral blood mononuclear cells (PBMC). The median T cell response was numerically higher in the AAV group when compared to healthy controls, but was not

significantly different (z-score -1.039, p = 1.00). Time since rituximab did not significantly influence immune response. Over the subsequent 12 months, 10 (33.3%) individuals in the AAV group had a positive SARS-CoV-2 test.

7.2.4 Conclusion

Compared to a healthy population, participants in OCTAVE with AAV previously exposed to rituximab had a severely attenuated antibody response, but a comparable T cell response. Therefore, AAV patients have a partial immune response to vaccination. Enhanced vaccination schedules and additional self-protective measures may be of value to this population.

7.3 Introduction

7.3.1 SARS-CoV-2 vaccination

From the start of the Covid-19 pandemic, national efforts were made to rapidly design and deliver vaccines against SARS-CoV-2. By late 2020, landmark trials of SARS-CoV-2 vaccination, which enrolled tens of thousands of participants, were reported. These demonstrated clear efficacy at reducing symptomatic Covid-19 (Polack et al., 2020; Voysey et al., 2021). Subsequent national surveillance studies confirmed the success of SARS-CoV-2 vaccination as a mass public health intervention. Vaccination was shown to be highly effective in reducing the incidence of asymptomatic and symptomatic SARS-CoV-2 infection, severe Covid-19 and Covid-19 related death (Haas et al., 2021; Pritchard et al., 2021). These efforts transformed the global experience of the Covid-19 pandemic, however subjects in the original SARS-CoV-2 vaccine trials did not have known chronic conditions and were not exposure to immunosuppressive therapies. Therefore the efficacy and effectiveness of SARS-CoV-2 vaccination in individuals with immune-suppressive conditions was not clear.

7.3.2 Vaccination in AAV and other immune-suppressive conditions

It is well recognised that individuals with immunosuppressive states are not only more susceptible to severe infections but also have attenuated vaccine responses (Agarwal et al., 2012). This is particularly true with respect to AAV, who are typically more intensely immunosuppressed than in other immune-mediated inflammatory diseases (IMID). After the introduction of SARS-CoV-2 vaccination, reports emerged of reduced antibody response in immunosuppressed individuals. One report found that 26% of individuals with autoimmune rheumatic disease did not mount a detectable antibody response (Boyarsky et al., 2021). Latterly a systemic review was performed to inform guidelines from the European Alliance of Associations for Rheumatology (EULAR) relating to management of rheumatic conditions in the context of Covid-19. This review found that the median detectable antibody response in populations of

individuals with rheumatic and musculoskeletal diseases was 88%, compared to the median response of 100% in control populations (Kroon et al., 2022).

The importance of neutralising antibodies against SARS-CoV-2 for prevention of severe disease has become increasingly apparent (Khoury et al., 2021; Chvatal-Medina et al., 2021). Of particular concern to AAV clinicians and patients is that the intense immunosuppression required to prevent early mortality due to active vasculitis suppresses the humoral immune system's ability to produce antibodies. While this includes ANCA-autoantibodies which promote AAV disease activity and likely plays a role in controlling active vasculitis, it also included antibodies directed at fighting microbes, whether derived from natural infection or vaccination (Merino-Vico et al., 2021). AAV patients who have recently been treated with rituximab are likely most vulnerable. Rituximab is a chimeric monoclonal antibody which targets a key component of the humoral immune system – B-cells. Rituximab is directed against CD20, a cell surface protein present on immature and mature B-cells but not expressed on stem cells or fully differentiated plasma cells. Rituximab leads to substantial B-cell depletion through antibody dependent cell mediated cytotoxicity and complement dependent cytotoxicity, which result in CD20-positive B-cell lysis, and through augmentation of CD20-positive B-cell apoptosis (Shaw et al., 2003). Plasma cells are not directly targeted by rituximab, which can lead to maintenance of ANCA autoantibody production, however the mechanism by which rituximab ameliorates disease may relate to reducing the role of increased levels of B cell cytokines in AAV and the role of dysregulated B cell populations (Merino-Vico et al., 2021). Ultimately rituximab typically depletes ANCA autoantibodies substantially and there are data to suggest that other general antibodies are depleted to a much lesser extent (Ferraro et al., 2008). Given that neutralising antibodies are likely a crucial immune component in combating SARS-CoV-2, that rituximab depletes the humoral immune response was concerning for a diminished antibody response to SARS-CoV-2 vaccination. This phenomenon has been observed in rituximab treated patients with respect to other vaccines, such as influenza vaccines (Eisenberg et al., 2013). A more recent report of AAV patients previously treated with rituximab described immunological response to SARS-CoV-2 vaccination. This included 11 patients who remained B-cell depleted and 8 with recovered B-cell status. The authors found that none of the B-cell

depleted group mounted a humoral response, while all the B-cell recovered group did. Notably this study examined the immune response following the first vaccine dose only (Marty et al., 2022). One report of 15 AAV patients treated with rituximab which examined the response to booster doses after the initial vaccine course did not provide much additional reassurance. This showed that the majority (eight patients) did not mount an antibody response. Most of those individuals remained B-cell deplete, while most of the responders had B-cell reconstitution at the time of booster (Kant et al., 2022). Similarly, in the broader context of rheumatic and musculoskeletal diseases, the EULAR systematic review found consistent evidence across multiple studies that rituximab substantially impacted the antibody response to SARS-CoV-2 vaccination. Two studies also showed that an increased time interval since rituximab administration was associated with an improved antibody response (Kroon et al., 2022).

While evidence has emerged of reduced immunological response to SARS-CoV-2 vaccination in immunosuppressed groups, many of these studies were limited by cross-sectional design, lack of a non-disease comparator group, heterogeneity in sampling timepoints, variation in immunological assays and small sample size. The EULAR systematic review found that individuals with rheumatic musculoskeletal disease did have a reduced antibody response to vaccination, however it highlighted that most of the studies had a high or unclear risk of bias (Kroon et al., 2022).

7.3.3 Immunological response to SARS-CoV-2 vaccination

SARS-CoV-2 engages with the host cell via the ACE2 receptor (Jackson et al., 2022). Specifically, the receptor-binding domain (RBD) on the S1 subunit of the spike protein binds to ACE2 to gain entry (Wrapp et al., 2020). Antibodies directed at the RBD tend to be efficient with respect to viral neutralisation (Premkumar et al., 2020). SARS-CoV-2 vaccines aim to induce an immunological response to the whole spike protein, crucially including the RBD. Assays assessing anti-SARS-CoV-2 spike protein antibodies (anti-S) typically detect antibodies directed specifically at the RBD (Roche, 2023b).

Measuring immune response to specific individual SARS-CoV-2 antigens can determine whether the immune response is due to natural infection or vaccination. Natural Covid-19 infection results in an immune response to the main SARS-CoV-2 antigens including spike, nucleocapsid and membrane proteins. In immunocompetent hosts both a humoral and T cell response occur to these antigens. SARS-CoV-2 vaccines result in the recipient being exposed to spike protein, but not other SARS-CoV-2 antigens. Therefore, an immunological response to spike protein (either anti-spike antibodies (anti-S) or a spike specific T cell response), can be caused by either natural infection or vaccination. An immunological response to other antigens (whether antibody or T cell), can only be caused by natural infection.

7.3.4 Aim

Observational Cohort trial T cells, Antibodies and Vaccine Efficacy in SARS-CoV-2 (OCTAVE), was a prospective, multicentre study undertaken to determine the immunological response to SARS-CoV-2 vaccination in immunosuppressed groups, across a wide spectrum of clinical conditions. AAV patients who had recently received rituximab were one of those disease groups, recruited by the OCTAVE team at University of Glasgow. This study aims to utilise the AAV subset of the OCTAVE study to characterise the humoral and cellular immune response to SARS-CoV-2 vaccination in AAV patients recently treated with rituximab, with comparison to a healthy control group from OCTAVE. We sought to evaluate the impact on immune response of recency of rituximab and to determine if there was a relationship between the magnitude of humoral response and cellular response. A secondary objective was to determine the number of AAV patients who went on to have virologically confirmed SARS-CoV-2 infection and to identify potential predictive factors.

7.4 Methods

7.4.1 OCTAVE study design and ethics

This study is a sub-study of the OCTAVE trial. OCTAVE was a multicentre prospective cohort trial including participants from multiple disease groups, all of whom were immunosuppressed. It evaluated the immune response to SARS-CoV-2 vaccination in individuals who were receiving this intervention as part of

regular care. The study was jointly led by teams from the Universities of Glasgow, Birmingham, Oxford, Sheffield and Imperial College London. The sponsor was the Cancer Research UK Clinical Trials Unit (CRCTU) at the University of Birmingham. Approvals were granted by the UK Medicines and Healthcare Products Regulatory Agency (MHRA) and by the London and Chelsea Research Ethics Committee (REC ref.: 21/HRA/0489) in February 2021. Study registration details are available on the International Traditional Medicine Clinical Trials Registry at the following link: ISRCTN12821688. The trial protocol was amended on eight occasions, although except for the first amendment, none of the amendments affected the current sub-study. The first amendment reflected an alteration to inclusion criteria which allowed participants to be included in the deep immunophenotyping group (described below), after receiving the first Covid-19 vaccination dose but before the third (booster) dose, as opposed to only recruiting those who had not received any vaccination dose. More detailed information regarding amendments is available in the initial published OCTAVE study article (Barnes et al., 2023).

7.4.2 Study participants

All individuals taking part in the study provided written, informed consent. A variety of disease groups were included, all of whom were immunosuppressed. This sub-study focused on the participants with AAV, all of whom were recruited by the team based at University of Glasgow (UofG). Only patients with AAV who had been treated with rituximab within the previous 12 months to the time of Covid-19 vaccination were recruited for this disease group. A detailed description of the other disease groups recruited as part of OCTAVE is available in the main OCTAVE article (Barnes et al., 2023). Adult and paediatric patients were recruited to the main study, however only adult clinical sites recruited as part of the UofG team. AAV patients were recruited in mid-February 2021.

Inclusion criteria for the AAV disease group participating in OCTAVE were:

- Covid-19 vaccination was planned as part of routine National Health Service (NHS) care

- The second vaccine dose had not yet been administered

- Expected to live for at least a further six months

- Have a diagnosis of AAV according to international standards and had received treatment with intravenous rituximab within the previous 12 months

Exclusion criteria (for the deep immune phenotyping group) initially included having received an initial vaccination dose and had not had usable blood samples taken as part of another study available, however this was revised in line with the amendment described above in section 7.4.1.

Across all disease phenotypes, the study recruited 2,686 participants. The deep immune phenotyping group (described further below) included 674 of these participants. All AAV participants were in the deep immune phenotyping group. The other group was labelled the 'serology cohort'.

7.4.3 Description of the deep immune phenotyping group

All patients with AAV in the OCTAVE study were entered into the deep immune phenotyping group. This was characterised by more intensive sampling. The deep immune phenotyping group were sampled at the following time points: before the first vaccine dose (baseline – if logistically possibly), before the second vaccine dose, 28 days after the second vaccine dose (plus or minus 3 days), 6 months after the second vaccine dose and 12 months after the first vaccine dose (with closest feasible proximity to these time points if not otherwise specified). Samples were taken for interferon gamma (IFNγ) T cell response to a selection of SARS-CoV-2 antigens (T-cell response, Oxford Immunotec assay) as well as samples for anti-SARS-CoV-2 antibodies (Roche Elecsys®) – these were the same as those taken for the serology group and are described below. Additional samples including blood and saliva were taken for assays which were to be decided upon as more information regarding the Covid-19 immune response emerged. The serology group was sampled at 28 days following the second vaccine dose, 6 months after the second vaccine dose and 12 months after the first vaccine dose. Sampling was performed in NHS clinical areas and samples were stored and transferred in line with local clinical policies and Human Tissue Authority guidance.

7.4.4 Baseline characteristics

Data was collected locally on a paper format and then transcribed to a UofG clinical database held on a secure server in line with local data protection policies. The data was then submitted to the sponsor which captured the data using a REDCap (Research Electronic Data Capture) database held at the University of Birmingham CRCTU. Baseline data included age, sex, AAV diagnosis, ANCA autoantibody titre, AAV disease duration, previous or current renal involvement (defined as per the Birmingham Vasculitis Activity Score criteria), eGFR in mL/min/1.73 m^2 (using the CKD-EPI formula), medication history and previous Covid-19 infection. Specific medication history reported for AAV participants included time since the last administration of rituximab, glucocorticoid dose in daily prednisolone equivalents and the presence of concomitant immunosuppressive treatment. Baseline data for health controls (HC) were derived from the main OCTAVE article.

7.4.5 Vaccine

SARS-CoV-2 vaccines were administered in this study as part of routine NHS care and in accordance with UK national legislation and guidance. The timing of vaccination was in line with routine care as per national guidance. This was done both either by NHS clinical staff or OCTAVE study investigators. For the current study UofG OCTAVE study investigators administered the vaccine in an NHS-ran facility. OCTAVE was registered with the UK Medicines and Healthcare products Regulatory Agency (MHRA; UK MHRA clinical trial authorization number: 21761/0365/001) given that this healthcare intervention was being administered to new patient groups. Vaccines utilised in OCTAVE were BNT162b2 (Pfizer/BioNTech) or ChAdOx1 nCoV-19 (Oxford–AstraZeneca) vaccines. At UofG clinical study sites, only ChAdOx1 nCoV-19 vaccines were administered, therefore all participants in the current study received this vaccine.

7.4.6 Immunologic outcomes, sampling and assays

Outcomes for this study included the magnitude of immunological response to SARS-CoV-2 vaccination, both humoral and T cell mediated, as described below.

7.4.6.1 Anti-SARS-CoV-2 Antibodies

This included anti-SARS-CoV-2 IgG antibodies against the spike (anti-S) and the nucleocapsid (anti-N) antigens of SARS-CoV-2. The assay utilised for antibody testing was the Roche Elecsys® immunoassay. This electro-chemiluminescence immunoassay quantifies antibodies to SARS-CoV-2 spike protein RBD in serum or plasma. The assay employs a recombinant protein which mimics the antigen and utilises a double-antigen sandwich method. The range of detectability of the assay encompasses 0.4 – 250 U/mL. Less than 0.8 U/mL is considered non-reactive, greater than or equal to this threshold is considered reactive (Roche, 2023b). An anti-S sub-optimal response threshold was calculated as the upper bound of the lowest decile in a healthy control cohort as part of the main OCTAVE analysis, this threshold was 380 U/mL. Samples were collected in serum separator tubes. The methods relating to the anti-N assay are the same, though for result interpretation less than 1 U/mL is considered non-reactive, while greater than or equal to this threshold is considered reactive (Roche, 2023). For illustration purposes, a suboptimal response threshold for anti-N was estimated from published reports at 10 U/mL (Movsisyan et al., 2022). These assays were performed by the UK Health Security Agency laboratories at Porton Down.

7.4.6.2 SARS-CoV-2 T cell response

T cell response was quantified with the T-SPOT Discovery SARS-CoV-2 assay by Revvity (formerly Oxford Immunotec; https://www.revvity.com/). This assay is a modified ELISPOT assay. This chapter reports the T cell response to the spike protein (whole spike) and nucleocapsid protein. Other T-SPOT assays were performed but are not reported in detail in this study. The other assays performed included response to individual spike protein subunits (S1 and S2) and membrane protein. Initially peripheral blood mononuclear cells (PBMCs) were isolated from whole blood. Then a specific quantity of PBMCs was stimulated with a pool of SARS-CoV-2 peptides, to which the cells release interferon gamma (IFNγ) if they are responsive to antigen stimulation. Labelled anti-IFNγ antibodies were then applied which react with released IFNγ. A detection reagent was then added which reacts with the labelled antibody. This reaction produces spots representing IFNγ secreting T cells which can then be quantified (Wyllie et al., 2021). The result was expressed as spot forming units (SFU) per

10^6 PBMCs. Whole blood was collected for this assay in lithium heparin collection tubes.

7.4.7 Clinical outcomes

Death during follow-up was identified, this is reported alongside baseline characteristics, including stratification by time since rituximab. The occurrence of Covid-19 over the first year post vaccination was collated, defined as a positive SARS-CoV-2 test obtained as part of routine clinical care. The type of test for SARS-CoV-2 could include polymerase chain reaction (PCR) or point-of-care antigen testing. Patient self-test data was not collected.

7.4.8 Control group

It was not possible for specific control data to be made available from the main OCTAVE study for the current sub-study, therefore references to control data are derived from the main OCTAVE published article. In the main study, non-disease control data was obtained from the UK PITCH cohort and the UKHSA CONSENSUS cohort. This data was prospective, multi-centre and assessed immune response to both Covid-19 natural infection and SARS-CoV-2 vaccination. In PITCH, blood samples were obtained 28 days (plus or minus 7 days) after the second vaccine dose (Angyal et al., 2022). In CONSENSUS, blood samples were obtained at 0, 3, 6, 9, 12, 15 and 20 weeks following the first vaccine dose (Amirthalingam et al., 2021). OCTAVE participants were matched to controls by age, sex, previous Covid-19 and vaccine type. Matching was performed using a proportional matching method with four analysis groups created to match the following groups: the complete OCTAVE dataset, the deep immunophenotyping group, the serology group and participants recruited specifically with renal disease. Controls referred to in this study are from the deep immunophenotyping group controls, given that all AAV patients in OCTAVE were part of the deep immunophenotyping group.

7.4.9 Bias

Selection bias was an important consideration, as for any cohort study. The study protocol was such that patients were invited to receive the vaccination in a clinical facility that may have been more difficult for them to access. As a

result, some frail patients or patients with mobility issues may have been less inclined to participate. Efforts were made to minimise inconvenience to participants. Reimbursement was made for travel expense and transportation was arranged for participants where requested. Availability of reimbursement was clearly communicated to participants and included on study participant information documentation. Crucially, this study recruited only AAV patients who had recently (within the past year) been treated with rituximab. Therefore the results will not necessarily extrapolate to AAV patients not treated with rituximab.

Due to the rarity of AAV, recruitment to clinical studies is typically challenging. Small numbers in an arm of a study increase the probability of type one and type two errors (detecting a spurious association and failing to detect a true association respectively). Due to anticipated relatively small numbers of participants, only univariable statistical analyses or analyses comparing two groups within the AAV cohort, without statistical adjustment for additional variables, were planned. As well as being statistically appropriate, this was also practically required due to individual level control data not being available for this sub-study. This lack of statistical adjustment would potentially result in certain confounding variables impacting upon the direction or magnitude of results.

Information bias, where measurement of outcomes varies across groups, is potentially of relevance. As described above, there were differences with the timing of blood sampling for OCTAVE participants compared to controls. Some controls may have been tested earlier than OCTAVE participants, potentially resulting in a decrease in the magnitude of the control outcome.

Confounding is a potential source of bias. In the main OCTAVE study, participants were matched to controls as described in section 7.4.8. It was not possible to do specific matching for AAV participants in the current study, therefore confounding may impact upon the magnitude of difference between groups. Potential important confounders include age, previous infection and kidney function. Age is now recognised as a major determinant of antibody response in SARS-CoV-2 infection and SARS-CoV-2 vaccination, but unusually with different effect directions. A systematic review found that in natural infection,

age was positively correlated with neutralising antibody response, while the UK REACT study found that for both BNT162b2 and ChAdOx1 vaccines antibody positivity decreased with age (Chvatal-Medina et al., 2021; Ward et al., 2022). Previous SARS-CoV-2 infection increased antibody response to vaccination (Ward et al., 2022). In OCTAVE, matching was not performed for kidney function. Low kidney function is recognised as having an adverse impact upon immune response to SARS-CoV-2 vaccination and upon the likelihood of occurrence of Covid-19 and severe Covid-19 following vaccination (Carr, Kronbichler, et al., 2021; Barda et al., 2021).

The Hawthorne effect, whereby participant behaviour can be influenced by being observed as part of the study, may have impacted the SARS-CoV-2 infection outcome. OCTAVE participants were informed that the reason for their participation was that they had an underlying condition which resulted in susceptibility to increased infection severity. As a result, participants may have taken additional protective measures on a personal basis to avoid potential exposure to SARS-CoV-2. The healthy control group would not necessarily have changed their behaviour in the same way as immunosuppressed individuals in OCTAVE. Furthermore, many of the control group were health care workers who would have had substantial and unavoidable occupational exposure risk relating to SARS-CoV-2. For this reason, comparisons between the AAV group and healthy controls are not drawn for SARS-CoV-2 infection in this study. Due to the potential Hawthorne effect the incidence of SARS-CoV-2 infection in this study should be interpreted with caution as it may underestimate the true population incidence of SARS-CoV-2 in vaccinated AAV patients.

7.4.10 Study size

Study size was determined by the maximal number of participants with AAV who had received rituximab within the prior 12 months who could be recruited to the sub-study. Specific sample size calculations were not performed.

7.4.11 Patient and public involvement

In the context of the Covid-19 pandemic, there was reduced time available for study development and approvals before patients received vaccination as part of

routine care. This resulted in a short window for recruitment and therefore limited opportunity for patient and public involvement prior to the study commencing. However, the Trial Management Group at the University of Birmingham includes patient and public representatives, who were involved in the development of the protocol. Specific local patient and public involvement was not possible for the current sub-study.

7.4.12 Statistical methods

Baseline characteristics were presented as number and percentage for categorical data. Continuous baseline data was presented as median (IQR). Immunological result data was presented as both mean and median in tabular form. For anti-S data, the number of individuals who achieved a titre above the suboptimal response threshold (≥ 380 IU/mL, described above in section 7.4.6.1) was displayed along with percentage. Relevant plots were annotated with this threshold. Similarly, a suboptimal dose threshold annotation was added to relevant plots for anti-N (≥ 10 IU/mL, see section 7.4.6.1). For pre second dose anti-N titres, most values (24) were undetectable. If zero was used as the value, this would have resulted in a distorted plot on the transformed log scale (as log of zero is undefined). Therefore, for plots with zero values, the lowest titre achieved before or after booster (0.0694 IU/mL) was used to replace zero values, zero on the non-log scale was used for all other purposes. Missing data is displayed in the relevant tables, either within the table or the table caption. Fisher's exact test was used to compare proportions of categorical outcome between different groups. The Wilcoxon test was used to test for difference between two groups of continuous variable observations. Visual inspection, Quantile-Quantile (QQ) plots and Shapiro-Wilk tests were used to confirm that the data was not normally distributed. The Pearson correlation coefficient (PCC) and associated p-value was calculated to assess the relationship between two continuous variable observations. For plots comparing two groups of continuous variable observations, boxplots with overlying scatter plots were used. Two versions of each of these plots are presented – one with a jitter effect applied to the scatter points, this disperses the points laterally and allows all data points to be visualised. The second plot is not jittered, but instead has paired lines – this enables the reader to visualise the direction and magnitude of change for individual participant antibody titres before and after the second vaccine dose.

For survival data, Kaplan-Meier curves were used. Hazard ratios, confidence intervals and p-values were derived using univariable Cox proportional hazards models. The proportional hazards assumption was check using scaled Schoenfeld residuals. All analyses were conducted in R (R version 4.2.2, http://www.r-project.org) with packages including tidyverse, survival and finalfit.

7.5 Results

7.5.1 Baseline characteristics

The OCTAVE study recruited 2,686 individuals, 30 of whom had AAV and had also received rituximab therapy within 12 months prior to Covid-19 vaccination. Recruitment for the AAV group was very high, with very few individuals declining participation (fewer than five individuals declined). Reasons cited for not participating included frailty and lack of mobility. The baseline characteristics of the group, including stratification according to recency of rituximab is shown in Table 7-1. Rituximab recency is divided into two groups: those who received rituximab within six months prior to vaccination (RTX 6 months) and those who received rituximab within six to twelve months prior to vaccination (RTX 6-12 months). The median age of the whole AAV cohort was 55.3 years. Those who received rituximab more recently had a younger median age (46.4 versus 58.5). Half of the whole cohort were female. The group who received rituximab more recently had a lower percentage of females (38.5% versus 58.8%). In the whole cohort the majority of patients had a vasculitis diagnosis of GPA, the remainder were AAV not specified. Most (25 individuals, 83.3%) were anti-PR3 positive, three (10%) were anti-MPO positive, one (3.3%) was ANCA-autoantibody negative, while 1 (3.3%) had unknown ANCA autoantibody status. There was a large overlap between those who had GPA and those who were anti-PR3 positive (data not shown). The median disease duration was 3.1 years. Twelve individuals (40%) had renal involvement. The median time since rituximab was 175 days. The median glucocorticoid dose was 5 mg (daily prednisolone equivalents). Four patients (13.3%) were receiving concomitant immunosuppressive in addition to recent rituximab. Two patients (6.7%) died during follow-up, both of whom were in the group which received rituximab within 6 months prior to Covid-19 vaccination.

7.5.2 Antibody response

Antibody response was evaluated before and after the second Covid-19 vaccine dose. The mean anti-SARS-CoV-2 spike antigen antibody response (anti-S) before the second dose was 5.15 IU/mL, compared to 172.15 IU/mL after the second dose ($p=0.042$, Wilcoxon test). The median anti-S response was 0.4 IU/mL before

and after, with 0.4 IU/mL essentially representing a completely negative test. Both before and after vaccination, the majority of study subjects had negative test results (i.e. below 0.8 IU/mL, represented by the thick grey line in the relevant figures). Only three (10.3%) patients of the 29 for whom data was available achieved a titre greater than the healthy control lower bound (380 IU/mL = 2.58 on log scale in figures) after the second vaccine dose. None had achieved this titre before the second dose. These results are visualised in Figure 7-1 A. and B. and tabulated in Table 7-2. When compared to the healthy control group from the main OCTAVE study, 8 of 29 (27.6%) AAV patients achieved seropositivity compared to 222 of 225 (98.7%) of healthy controls ($p < 0.0001$, Fisher's exact test).

Antibody response to anti-SARS-CoV-2 nucleocapsid antigen antibody (anti-N) was also evaluated before and after the second Covid-19 vaccine dose. The mean anti-N response before the second dose was 0.71 IU/mL, compared to 0.76 IU/mL after ($p<0.001$, Wilcoxon test). The median anti-N before was 0.07 IU/mL and 0.09 IU/mL after. Notably, only six individuals had a detectable anti-N value prior to the second vaccine dose, five of whom were below the threshold for a positive test. The remainder of the cohort had a "not detected" value substituted with the lowest detected value in the study for anti-N to enable visualisation and hypothesis testing, this is discussed further in the methods and results sections. Crucially, while statistically significant these mean and median results are all negative meaning they are unlikely to be biologically or clinically significant. One patient had an anti-N response that was positive (greater than 1 IU/mL, zero on the log scale in the figures) and also greater than 10 IU/mL (1 on the log scale in figures), an approximate figure representing the lower bound of a typical positive anti-N response for non-immunosuppressed individuals, discussed further in the methods section. These results are visualised in Figure 7-1 C. and D. and tabulated in Table 7-3.

7.5.3 T-cell response

Mean IFNγ T cell response to full spike protein (T-cell response, Oxford Immunotec assay) was 118.13 T cells per 10^6 PBMCs before the second vaccine dose and 183.31 T cells per 10^6 PBMCs after the second vaccine dose ($p = 0.239$, Wilcoxon test). The median was 46 T cells per 10^6 PBMCs before and 104 T cells

per 10^6 PBMCs after. This is displayed graphically in Figure 7-2 A. and B. and tabulated in Table 7-4. The mean T-cell response to nucleocapsid was negative before and after the second vaccine dose in all participants (p = 0.188, Wilcoxon test). This is displayed graphically in Figure 7-2 C. and D. and tabulated in Table 7-5. The T cell responses to the membrane protein and spike subunits were consistent with the above data and are not reported here. Individual level T cell response data for healthy controls was not available for the purposes of this sub-study. In the main OCTAVE study, a z-score was calculated indicating that the AAV group had a numerically higher T-cell response compared to healthy controls, though this was not statistically significant (z-score -1.039, p = 1.00).

7.5.4 Immunological response split by time since rituximab

For the group which received rituximab within 0-6 months prior to Covid-19 vaccination (RTX 0-6 months), the mean post-second vaccine anti-S response was 6.7 IU/mL, while for the group which received rituximab 6-12 months prior to vaccination (RTX 6-12 months), the mean anti-S response was 289.0 IU/mL (p = 0.187, Wilcoxon test). The median response was 0.4 IU/mL for both groups. This is displayed graphically in Figure 7-3 A. and B. and tabulated in Table 7-6.

The mean full spike T-cell response for RTX 0-6 months was 252.3 T cells per 10^6 PBMCs, with the mean RTX 6-12 response was 134.6 T cells per 10^6 PBMCs (p = 0.138, Wilcoxon test). The median response was 126 T cells per 10^6 PBMCs for RTX 0-6 months and 76 T cells per 10^6 PBMCs for RTX 6-12 months. This is displayed graphically in Figure 7-3 C. and D. and tabulated in Table 7-7.

7.5.5 Correlation of antibody and T-cell response

No significant correlation was established between anti-S and full spike T-cell response for the whole cohort (R = -0.1, p = 0.59, Pearson correlation coefficient [PCC]). This was also not significant when the same relationship was examined for the RTX 0-6 months group (R = -0.17, p = 0.54, PCC) or the RTX 6-12 months group (R = 0.072, p = 0.81, PCC). This is displayed graphically in Figure 7-4.

7.5.6 SARS-CoV-2 positivity

In the cohort overall, 10 individuals (33.3%) had a positive test for SARS-CoV-2 taken as part of routine clinical care over one year. As described in Table 7-8, individuals who had a positive test were younger, tended to have better renal function and tended to have had rituximab more recently. They tended to have a poorer antibody response to vaccination, but a better T-cell response to vaccination. These differences were not necessarily statistically significant, univariate associations were determined by survival analysis in Table 7-9 and visualised in Figure 7-5 to Figure 7-9. Older people, namely those aged 50 years or older compared to those below the age of 50 years, were at lower risk of SARS-CoV-2 infection with univariable hazard ratio (HR) 0.16 (0.04-0.61, p=0.008). This was similarly reflected when age was considered as a continuous variable with HR 0.93 (0.89-0.97, p=0.001). Better renal function was associated with higher risk of contracting SARS-CoV-2. eGFR greater than or equal to 90 ml/min/1.73m^2 were carried a HR of 7.86 (1.66-37.24, p=0.009), with the referent being those with eGFR below 90 ml/min/1.73m^2. There were no significant relationships identified between SARS-CoV-2 infection and the other examined variables which included sex, time since rituximab, anti-S level and T-cell response. Both immunological variables were measured 28 days following the second vaccine dose.

Table 7-1 | Baseline characteristics and mortality stratified by time since rituximab

		RTX within 6 months	RTX wi
Total N (%)		13 (43.3)	17 (56.
Age in years; median (IQR)		47.4 (37.8 to 57.6)	58.5 (5
Female sex; number (%)		5 (38.5)	10 (58.
AAV diagnosis; number (%)	GPA	11 (84.6)	14 (82.
	AAV unspecified	2 (15.4)	3 (17.6
ANCA autoantibody; number (%)	MPO	1 (7.7)	2 (11.8
	PR3	11 (84.6)	14 (82.
	negative	0 (0)	1 (5.9)
	unknown	1 (7.7)	0 (0)
Disease duration; median (IQR)		5.3 (1.0 to 11.3)	3.0 (1.4
Renal involvement		5 (38.5)	7 (41.2
eGFR (CKD-EPI – mL/min/1.73 m^2); median (IQR)		91.2 (75.1 to 116.8)	77.2 (5
Time since RTX; median (IQR)		90.0 (60.0 to 96.0)	214.0 (
Glucocorticoid dose; median (IQR)*		5.0 (5.0 to 10.0)	5.0 (0.0
Concomitant immunosuppression; number (%)		3 (23.1)	1 (5.9)
Previous Covid-19 infection		2 (15.4)	0 (0)
Death occurred in follow-up; number (%)		2 (15.4)	0 (0)

AAV = ANCA-associated vasculitis, eGFR = estimate glomerular filtration rate, GPA = granulomatosis with polyangiitis, HC = healthy controls, IQR = interquartile range, MPO = anti-myeloperoxidase antibody, PR3 = anti-proteinase 3 antibody, RTX = rituximab,
***Data was missing for one patient with respect to glucocorticoid dose. There was no other missing baseline or mortality data.**
****Healthy control data was taken from the main OCTAVE manuscript (deep immunophenotyping control group), as described in the methods section. Age (years) was reported in categories: 15-44: 27 (13%), 45-64: 64 (30%), 66-74: 98 (47%), over 75: 21 (10%). eGFR data was not available for controls. There was no missing or unknown data for age, sex or previous Covid-19 infection.**

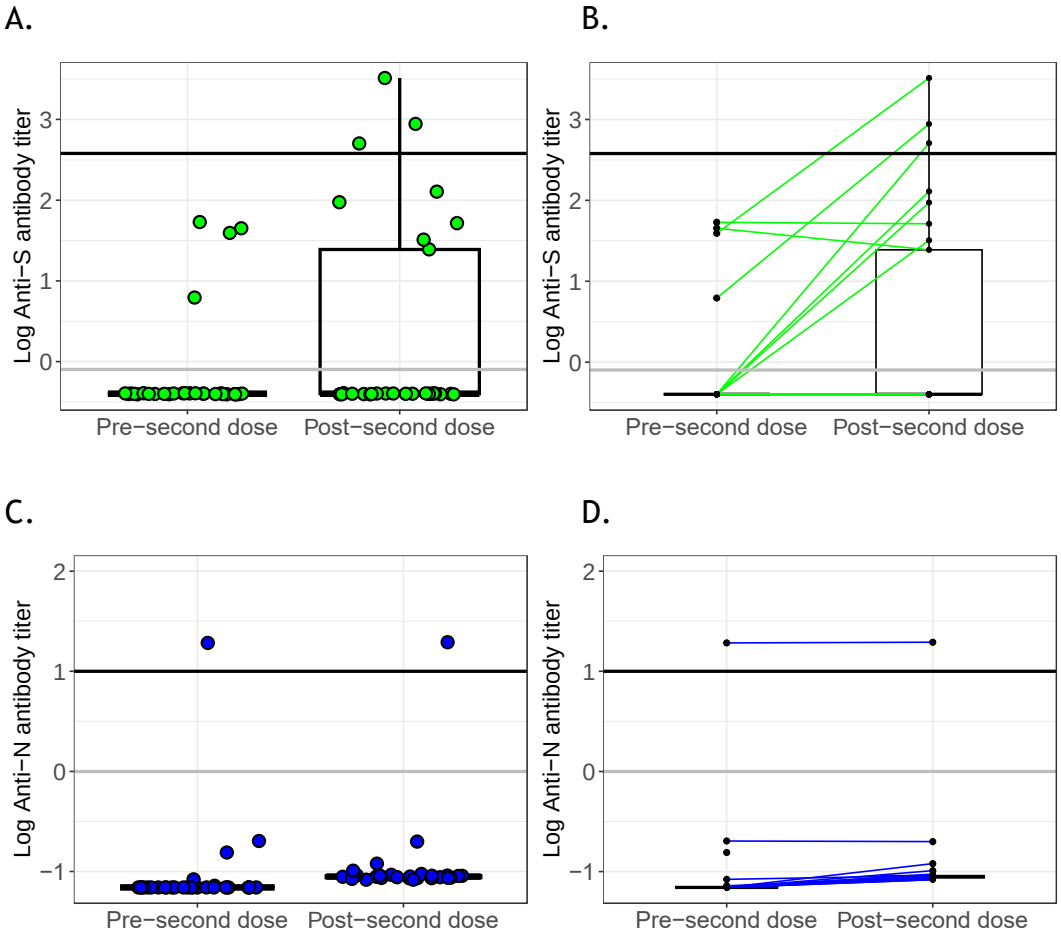


Figure 7-1 | Antibody response before and after second vaccine dose
A. Individual titre (IU/mL) for anti-spike (anti-S) antibody. Anti-S antibody represents the antibody response to vaccination or natural infection. On logarithmic scale for improved visualisation. Jitter function applied to separate data points horizontally ensuring no overlap, therefore all data points are visible. The thick grey line represents the threshold for a positive test (0.8 IU/mL = -0.1 on logarithmic scale), the thick black line represents the upper limit of the lowest decile of antibody response achieved in the healthy control comparator (380 IU/mL = 2.58 on logarithmic scale). p=0.042, Wilcoxon test.
B. As per Figure 7-1 A, but with paired lines to enable the change for individuals to be visualised. Jitter function not applied as this would distort the apparent magnitude of change.
C. As per Figure 7-1 A for anti-nucleocapsid (anti-N) antibody. Anti-N antibody represents the response to natural infection but does not become positive in response to vaccination alone in the absence of prior natural infection. The thick grey and black lines again represent the threshold for a positive test and a typical lower threshold that a non-immunosuppressed individual would achieve, respectively. The threshold for a positive test for anti-N is 1 IU/mL (0 on logarithmic scale)). P<0.001, Wilcoxon test (though importantly there is no clinically significant difference between these groups).
D. As per Figure 7-1 B for anti-N antibody.

Table 7-2 | Anti-spike antibody titre before and after second vaccine dose

Timing with vaccination	n	Missing	Mean Anti-S (IU/mL)*	Median Anti-S (IU/mL)	N (%), Titre >= 380 IU/mL
Pre-second dose	30	0	5.15	0.4	0 (0)
Post-second dose	30	1	172.15	0.4	3 (10.3)

***p=0.042, Wilcoxon test**

Table 7-3 | Anti-nucleocapsid antibody titre before and after second vaccine dose

Timing with vaccination	n	Missing	Mean Anti-N (IU/mL)*	Median Anti-N (IU/mL)
Pre-second dose	30	0	0.71	0.07
Post-second dose	30	1	0.76	0.09

***p<0.001, Wilcoxon test**

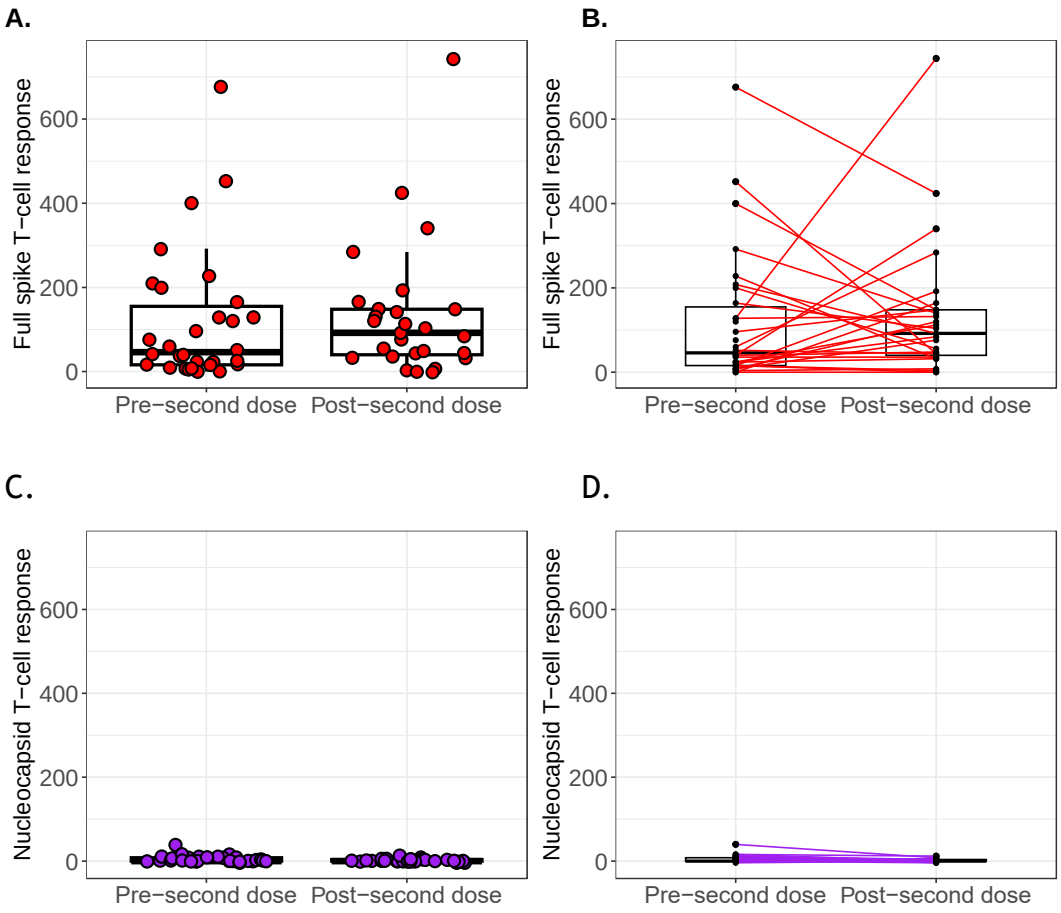


Figure 7-2 | T-cell response before and after second vaccine dose
A. IFNγ T cell response to full spike protein (Oxford Immunotec assay). T-cell response to full spike protein represents the T-cell response to natural infection or vaccination. Presented on a standard numeric (i.e. non-logarithmic) scale, units are spot-forming units (SFU) per 10^6 peripheral blood mononuclear cells (PBMCs). Jitter function applied to separate data points horizontally ensuring no overlap, therefore all data points are visible. p = 0.239, Wilcoxon test.
B. As per Figure 7-2 A, but with paired lines to enable the change for individuals to be visualised. Jitter function not applied as this would distort the apparent magnitude of change.
C. As per Figure 7-2 A for T-cell response to nucleocapsid protein. This represents the response to natural infection, but does not become positive in response to vaccination alone in the absence of prior natural infection.
D. As per Figure 7-2 B, but for T-cell response to nucleocapsid protein. p = 0.188, Wilcoxon test.

Table 7-4 | Full spike T-cell response before and after second vaccine dose

Timing with vaccination	n	Missing	Mean T cells per 10^6 PBMCs*	Median T cells per 10^6 PBMCs
Pre-second dose	30	0	118.13	46
Post-second dose	30	1	183.31	104

***p = 0.239, Wilcoxon test**

Table 7-5 | Nucleocapsid T-cell response before and after second vaccine dose

Timing with vaccination	n	Missing	Mean T cells per 10^6 PBMCs*	Median T cells per 10^6 PBMCs
Pre-second dose	30	0	5.2	0
Post-second dose	30	1	1.52	0

***p = 0.188, Wilcoxon test**

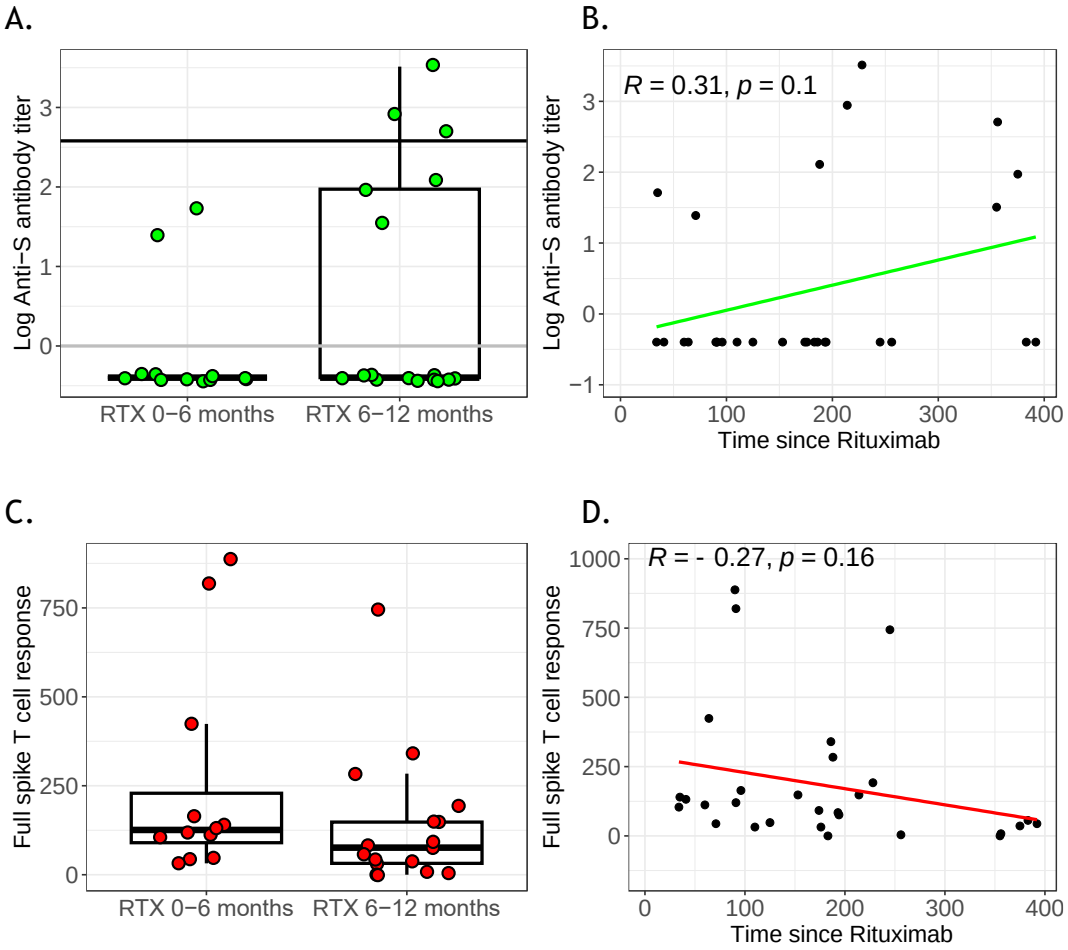


Figure 7-3 | Immunological response after second vaccination split by and correlated with time since rituximab

A. Individual titre (IU/mL) for anti-spike (anti-S) antibody, grouped according to rituximab within 0-6 months versus 6-12 months prior to vaccination. Anti-S antibody represents the antibody response to vaccination or natural infection. On logarithmic scale for improved visualisation. Jitter function applied to separate data points horizontally ensuring no overlap, therefore all data points are visible. The thick grey line represents the threshold for a positive test (0.8 IU/mL = -0.1 on logarithmic scale), the thick black line represents the upper limit of the lowest decile of antibody response achieved in the healthy control comparator (380 IU/mL = 2.58 on logarithmic scale). p = 0.187, Wilcoxon test.

B. Log anti-S antibody titre (IU/mL) correlated with time since rituximab (days).

C. IFNγ T cell response to full spike protein (Oxford Immunotec assay), grouped according to rituximab within 0-6 months versus 6-12 months prior to vaccination. T-cell response to full spike protein represents the T-cell response to natural infection or vaccination. Presented on a standard numeric (i.e. non-logarithmic) scale, units are spot-forming units (SFU) per 10^6 peripheral blood mononuclear cells (PBMCs). Jitter function applied to give horizontal movement to data points ensuring no overlap, therefore all data points are visible. p = 0.138, Wilcoxon test

D. T-cell response to full spike protein, as per Figure 7-3 C, correlated with time since rituximab (days).

R = Pearson correlation coeficient.

Table 7-6 | Anti-spike antibody titre split by time since rituximab

Time since RTX (months)	n	Missing	Mean anti-S, IU/mL*	Median anti-S, IU/mL	n (%), titre >= 380 IU/mL
0-6	13	1	6.66	0.4	0 (0)
6-12	17	0	288.96	0.4	3 (17.6)

***p = 0.187, Wilcoxon test**

Table 7-7 | Full spike T-cell response by time since rituximab

Time since RTX (months)	n	Missing	Mean T cells per 10^6 PBMCs*	Median T cells per 10^6 PBMCs
0-6	13	1	252.33	126
6-12	17	0	134.59	76

***p = 0.138, Wilcoxon test**

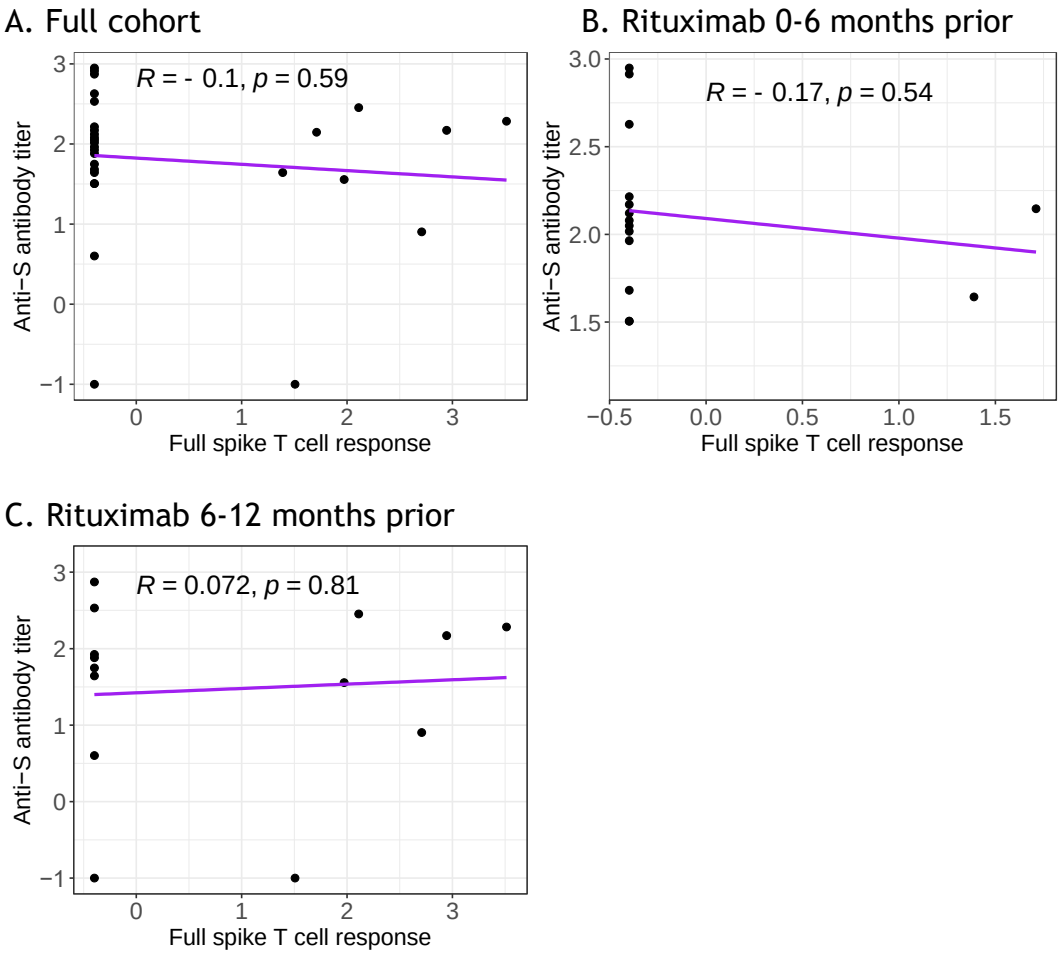


Figure 7-4 | Anti-spike antibody titre correlated with T cell response, split by time since rituximab
A. Log anti-spike (anti-S) antibody titre (IU/mL) correlated with log T-cell response for the full cohort. T cell response is IFNγ T cell response to full spike protein (Oxford Immunotec assay). Anti-S antibody represents the antibody response to vaccination or natural infection, units are spot-forming units (SFU) per 10^6 peripheral blood mononuclear cells (PBMCs). On logarithmic scale for improved visualisation.
B. As per Figure 7-4 A, but restricted to individuals who received rituximab within 0-6 months prior to vaccination.
C. As per Figure 7-4 A, but restricted to individuals who received rituximab within 6-12 months prior to vaccination.
R = Pearson correlation coeficient

Table 7-8 | Patient characteristics stratified by Covid-19 occurrence

		No	Yes
Total		20 (66.7%)	10 (33.3%)
Age (continuous)	Median (IQR)	60.7 (54.5 to 72.2)	37.4 (32.3 to 48.4)
Age (categorical)	< 50 years	3 (15.0)	7 (70.0)
	>= 50 years	17 (85.0)	3 (30.0)
Sex	Female	10 (50.0)	5 (50.0)
	Male	10 (50.0)	5 (50.0)
eGFR (CKD-EPI, ml/min/1.73m^2)	< 90	15 (78.9)	2 (20.0)
	≥ 90	4 (21.1)	8 (80.0)
Time since rituximab	< 6 months	9 (45.0)	7 (70.0)
	6-12 months	11 (55.0)	3 (30.0)
Anti-S (binary)	negative	13 (68.4)	8 (80.0)
	positive	6 (31.6)	2 (20.0)
Anti-S (value)	Median (IQR)	0.4 (0.4 to 63.0)	0.4 (0.4 to 0.4)
	Mean (SD)	258.6 (761.1)	7.9 (17.1)
T-cell response	Median (IQR)	92.0 (34.0 to 178.0)	116.0 (63.0 to 138.0)

Yes = SARS-CoV-2 positive test after one year, No = no positive test
Anti-S = anti-S antibody level measured 28 days after the second vaccine dose, eGFR = estimated glomerular filtration rate, IQR = interquartile range, SD = standard deviation

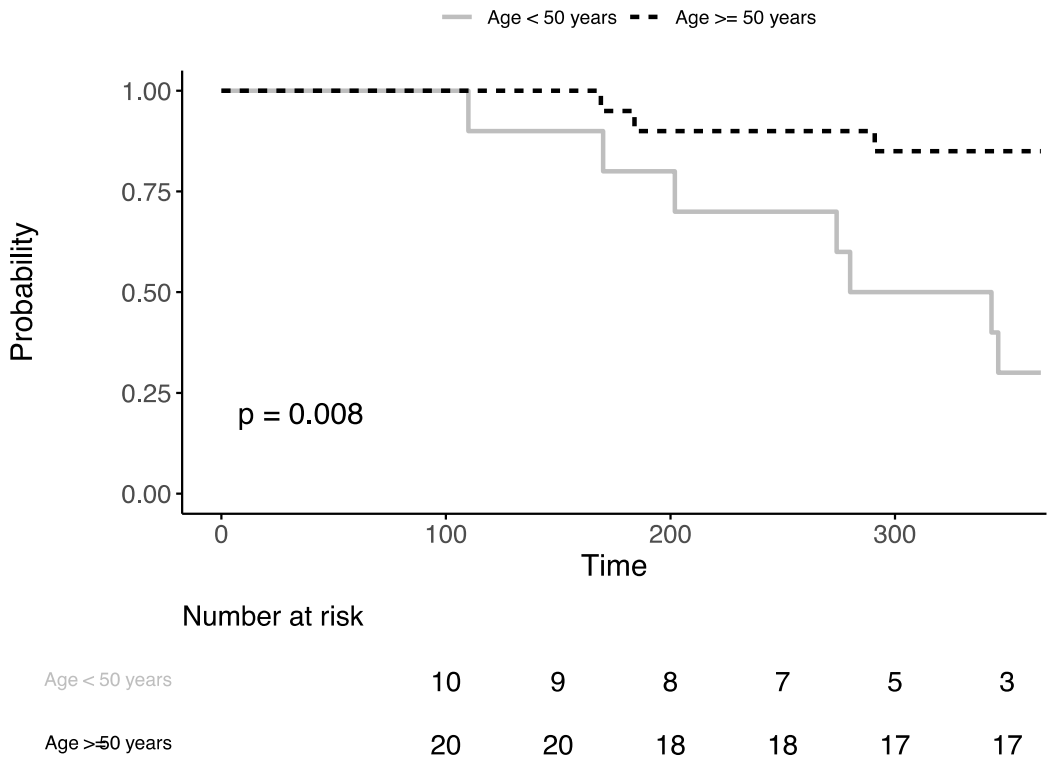


Figure 7-5 | SARS-CoV-2 positivity in first year by age
Test = univariate Cox proportional hazards model

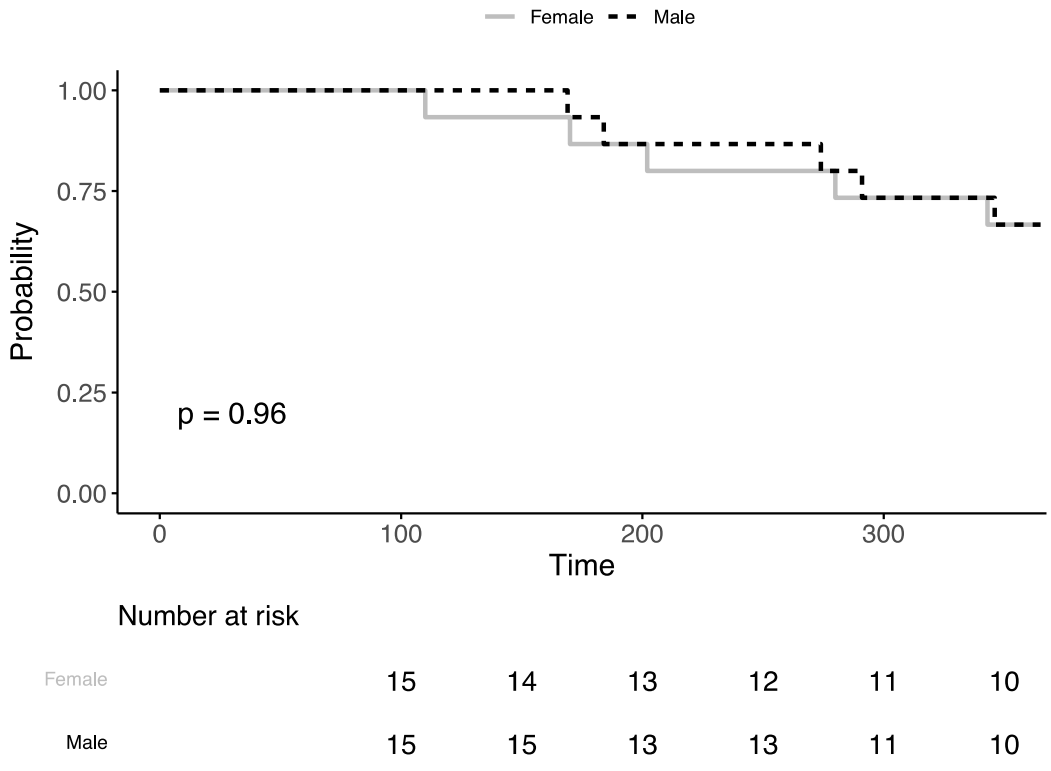


Figure 7-6 | SARS-CoV-2 positivity in first year by sex
Test = univariate Cox proportional hazards model

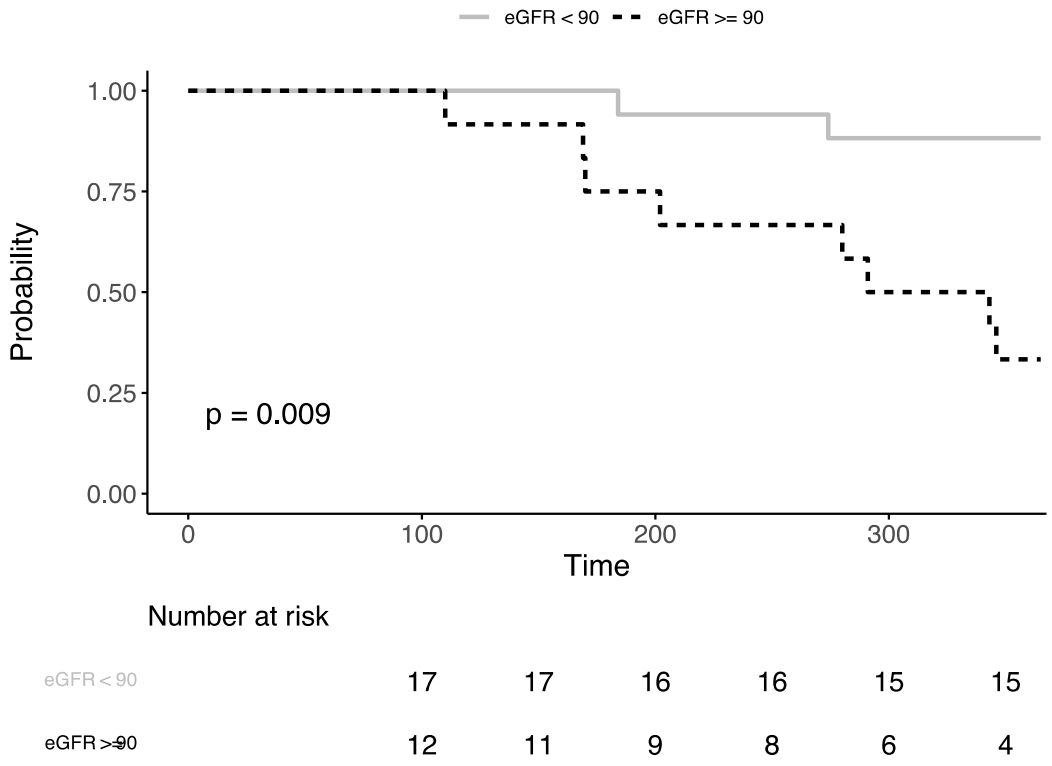


Figure 7-7 | SARS-CoV-2 positivity in first year by eGFR
Test = univariate Cox proportional hazards model

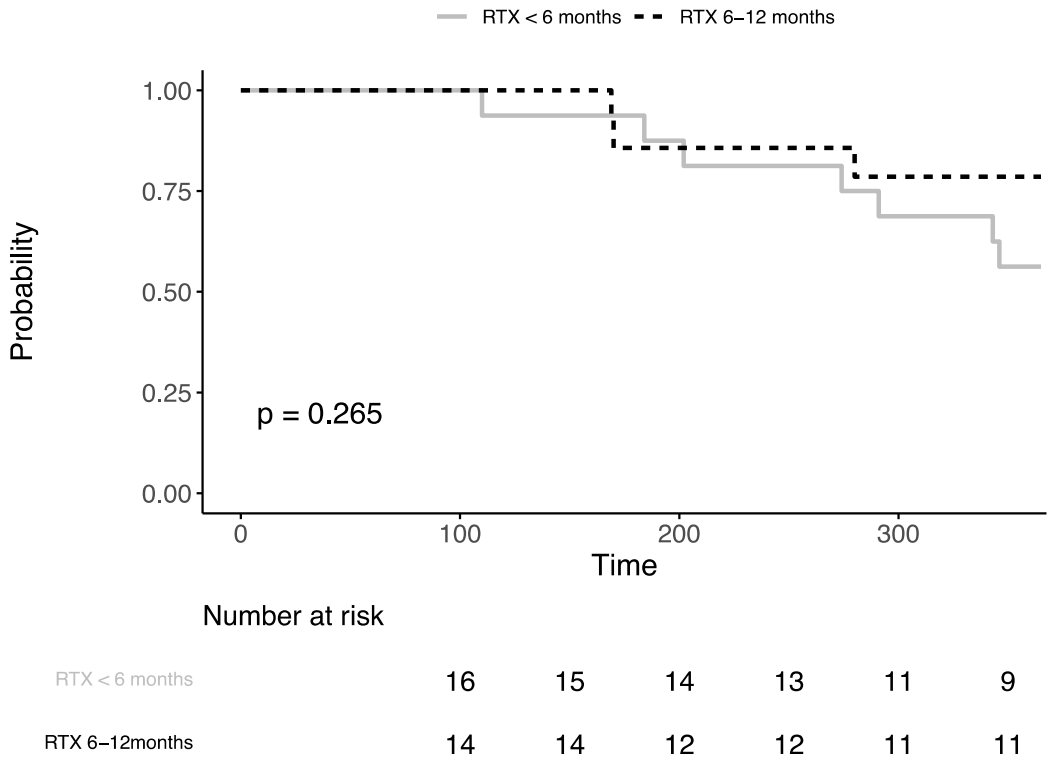


Figure 7-8 | SARS-CoV-2 positivity in first year by time since rituximab
Test = univariate Cox proportional hazards model

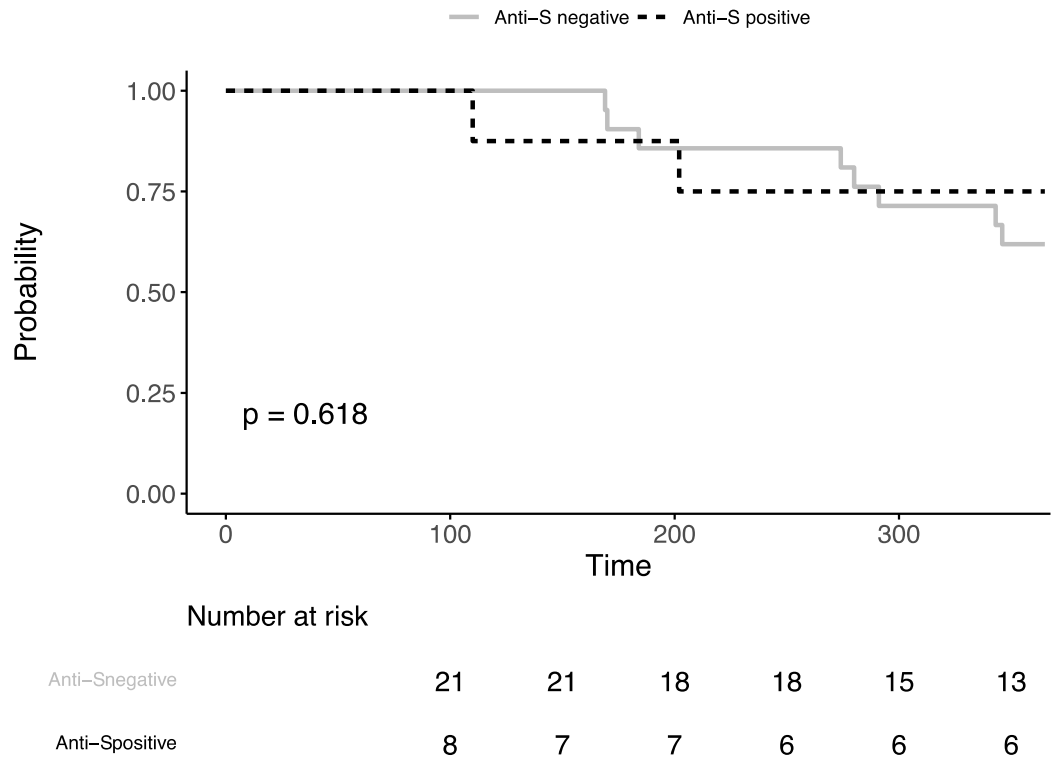


Figure 7-9 | SARS-CoV-2 positivity in first year by antibody response to vaccination
Test = univariate Cox proportional hazards model

Table 7-9 | Association between patient characteristics and SARS-CoV-2 positivity

Variable		n (%)	HR (univariable)
Age (continuous)	Mean (SD)	54.4 (16.4)	0.93 (0.89-0.97, p=0.001)
Age (categorical)	< 50 years	10 (33.3)	-
	>= 50 years	20 (66.7)	0.16 (0.04-0.61, p=0.008)
Sex	Female	15 (50.0)	-
	Male	15 (50.0)	0.96 (0.28-3.32, p=0.949)
eGFR (CKD-EPI, ml/ml/1.73m^2)	< 90	17 (58.6)	-
	≥90	12 (41.4)	7.86 (1.66-37.24, p=0.009)
Time since rituximab	< 6 months	16 (53.3)	-
	6-12 months	14 (46.7)	0.46 (0.12-1.79, p=0.265)
Anti-S (binary)	negative	21 (72.4)	-
	positive	8 (27.6)	0.67 (0.14-3.18, p=0.618)
Anti-S (value)	Mean (SD)	172.1 (622.3)	0.99 (0.97-1.01, p=0.480)
T-cell response	Mean (SD)	183.3 (241.5)	1.00 (1.00-1.00, p=0.311)

Anti-S = anti-S antibody level measured 28 days after the second vaccine dose, eGFR = estimated glomerular filtration rate, IQR = interquartile range, SD = standard deviation

7.6 Discussion

7.6.1 Key results

The primary aim of this sub-study of OCTAVE was to describe the immune response to SARS-CoV-2 vaccination in 30 AAV patients who had received rituximab within the previous 12 months. Baseline characteristics, incidence of virologically confirmed SARS-CoV-2 infection, mortality rates, antibody response and T cell mediated immune response before and after the second dose of vaccination are reported. Results are compared to healthy control responses from the main OCTAVE study report, though individual healthy control data was not available to allow detailed statistical comparison.

Analysis of baseline characteristics in the current study shows a younger population compared to typical AAV populations reported in epidemiological studies. Typical age ranges at disease onset are reported as 45-65 years for GPA, 55-75 years for MPA and 38-54 years for EGPA (Kitching et al., 2020). The population in the current study are likely to have been on average within a few years of diagnosis as they had all had recent treatment with rituximab. The median age at recruitment in the current study was 55.3 (IQR 39.2 – 68.2) years. This can be explained by a predominance of individuals with GPA, which has a lower age of onset compared to MPA. Fifty percent were female. Most of the participants in the study had GPA (25, 83.3%) and the same number had anti-PR3 positivity (25, 83.3%) – there was almost complete overlap between these groups. 13 (43.3%) received rituximab within 6 months prior to inclusion in the study (RTX 0-6 months), 17 (56.7%) received rituximab between 6 to 12 months prior to inclusion (RTX 6-12 months). Those in the RTX 0-6 months group were typically younger and had better renal function than the 6 to 12 months group. Comparison to available data for healthy controls reveals that although AAV is a disease typically associated with older people, the median age of healthy controls was higher and a higher proportion had evidence of previous Covid-19.

A substantially blunted humoral immune response was identified in AAV patients recently treated with rituximab. Although there was an increase in the mean anti-S antibody titre before and after the second SARS-CoV-2 vaccine (5.15 to 172.15 IU/mL, p=0.042, Wilcoxon test), the median did not alter from an

undetectable level (<0.4 IU/mL). This is reflected by the proportion of AAV patients who achieved seropositivity after the second vaccine dose, at 8 of 29 (28%) patients, compared to 222 of 225 (98.7%) of healthy controls ($p < 0.0001$, Fisher's exact test). After the second vaccine dose, only three individuals (10.3%) of 29 had a titre greater than the healthy control lower bound (380 IU/mL = 2.58 on log scale in figures) after the second vaccine dose. Anti-N was negative in all but one AAV patient, indicating that natural infection with Covid-19 was unlikely to have occurred in these individuals prior to or during the study, although it may have been possible for a participant to have been infected with SARS-CoV-2 during the study but not yet have seroconverted for anti-N to become positive at the time of testing.

The T-cell response showed a numerical, but not statistically significant, increase after the second dose of vaccine. The T-cell response was robust and comparable to that of healthy control data described in the main OCTAVE report (Barnes et al., 2023). There was no significant T-cell response to nucleocapsid antigen before or after the second vaccine doses.

After the second vaccine dose, individuals in the RTX 6-12 months groups had a numerically higher anti-S response, though this was not statistically significant. Similarly, a moderate strength, but non-statistically significant positive correlation was identified between anti-S response and time since rituximab. The converse was identified for T-cell response in relation to time since rituximab: the RTX 6-12 months groups had a lower T-cell response after the second dose and there was a moderate strength, non-significant, negative correlation between T-cell response and time since rituximab. As highlighted above, there were potentially important differences in baseline characteristics between the rituximab groups; RTX 6-12 months group participants were typically older and had worse renal function. This may have confounded the lack of statistically significant association between time since rituximab and immune response. These potential confounders may have diminished the apparent relationship between anti-S response and time since rituximab. No relationship was found on an individual level between anti-S response and T-cell response, including when stratified according to RTX 0-6 months and RTX 6-12 months groups.

One third (10 of 30, 33.3%) of individuals in the AAV group tested positive for SARS-CoV-2 when tested as part of routine clinical care over the subsequent 12 months following enrolment. Two factors were identified on univariate analysis as being associated with subsequent SARS-CoV-2 infection: younger age and better renal function. That such characteristics would be associated with increase susceptibility seems counterintuitive given that these are factors closely associated with improved immunity, as discussed above. It seems likely that these relationships are substantially confounded by self-protective measures. Older and frailer individuals would be far more likely to adopt self-protective measures. In this setting worse renal function can be considered a marker of frailty (Chowdhury et al., 2017). Adopting strict self-protective measures would make an individual potentially far less likely to contract SARS-CoV-2 (Talic et al., 2021). Similarly, younger and non-frail individuals may have been more likely to have occupational responsibilities that may have required them to have increased SARS-CoV-2 exposure.

7.6.2 Strengths

This study is one of the largest reported describing the immune response to SARS-CoV-2 vaccination in AAV patients receiving rituximab. The number of individuals in the AAV group was sufficiently large such that a clear difference was identified between the humoral response of that group and healthy controls, with a high degree of statistical confidence. It was prospectively designed and, while the sub-study was conducted in a single centre, had several advantages of a multi-centre study with study team expertise being drawn across multiple centres. The prospective design guards against potential conscious or unconscious selection bias on the part on study investigators whereby participants with a particular outcome are more or less likely to be included in the study due to that outcome. Prospective design also is more likely to achieve complete and accurate baseline data, which is crucial for an accurate description of the cohort and to allow statistical adjustment for confounders where appropriate. Patient participation was maximised through the availability of travel reimbursement and arranged transport for study subjects, thus minimising selection bias and enhancing generalisability.

Many of the initial studies of vaccine immune response in immune-suppressed individuals focused on the humoral immune response only (Carr, Wu, et al., 2021; Alexander et al., 2022; Redjoul et al., 2021). The current study examined both the antibody and T-cell mediated immune responses to vaccination, across a broad range of SARS-CoV-2 antigens. It also reports key clinical outcomes, namely SARS-CoV-2 infection and death, which were often not reported in earlier studies. Many studies also focused on diverse cohorts such as haemodialysis patients or a broad range of rheumatic diseases (Carr, Wu, et al., 2021; Boyarsky et al., 2021). The current study focuses on a clearly defined cohort within a disease area. Inclusion and exclusion criteria were explicit, thus enhancing generalisability to patient populations. Availability of a healthy control population is an additional key strength of the current study, an attribute not present in other studies. The studies from which the control population was derived, PITCH and CONSENSUS, followed methodology which was closely aligned to that of the immune-suppressed OCTAVE cohort. This enabled a clinically informative threshold to be established - the anti-S suboptimal response threshold. This was calculated as the upper bound of the lowest response decile in the healthy control group at 380 U/mL. This facilitated reporting in the current study that only three AAV patients (10.3%) of 29 achieved an antibody titre after the second dose of SARS-CoV-2 vaccine that was comparable to healthy controls.

Rare disease research is frequently hampered by small available sample size for clinical studies. A high participation rate was achieved in this study, as described above. Statistical analysis was carefully considered to avoid overfitting relating to the relatively small sample size in the AAV group: analyses were univariate and at most were only stratified by one additional variable.

7.6.3 Limitations

Though efforts were made to minimise its effects, selection bias is an issue for any prospective clinical study where recruitment occurs. High levels of participation amongst potential participants to the study were achieved, though some individuals did choose not to participate. Only a small number of individuals (fewer than five) declined, citing reasons including frailty and poor mobility. The results of this study are highly likely to be generalisable to most

individuals with AAV treated with rituximab in the prior 12 months, though a degree of caution should be used if generalising this data to significantly frail individuals. As noted above, the population in the current study had a moderately younger median age than what is considered typical for AAV onset. This may be representative of frail individuals declining participation, though this being a major effect was not apparent during recruitment. More likely is the predominance of individuals with GPA, which has a younger typical age of onset, as described above. The study focused on a clearly defined subset of individuals with AAV, those recently treated with rituximab. While this is advantageous for achieving clear results and helped identify a vulnerable group of individuals who had the poorest response to vaccination of all disease groups within OCTAVE, whether the results are generalisable to individuals with AAV who have not been recently treated with rituximab is less clear.

Small sample size is a recurring challenge for rare disease research. Recruitment of AAV participants exceeded expectations despite the challenging environment of the Covid-19 pandemic, but the relative numbers of AAV patients in OCTAVE was small despite this. The risk of spurious results increases in the setting of small samples, but this is unlikely to be the case for the main finding of the current study of the substantially attenuated antibody response in AAV patients compared to healthy control data reported in OCTAVE, where 27.6% of AAV patients on rituximab had a serological response compared to 98.7% of controls ($p < 0.0001$). Other analyses were mostly negative, it is possible that this represents type II error (i.e. false negatives) for some of these analyses. Therefore, results that are numerically of potential interest, but fall short of the traditional threshold for statistical significance, could be viewed as questions that may be worth exploring in future studies where greater power may be available.

Information bias, where measurement of outcomes varies systematically across different groups in a study, is an important consideration, though is unlikely to have significantly impacted on the current study. The pattern of blood sampling did vary across disease and control groups in OCTAVE. The main time point of interest for immunological outcomes in this study was following the second vaccination dose. In OCTAVE, this sampling was done 28 (plus or minus 3) days following the second vaccine. The healthy controls for OCTAVE were selected

from the UK PITCH and CONSENSUS studies. In PITCH sampling was done 28 (plus or minus 7) days after the second vaccine, while in CONSENSUS there was a fixed schedule with 3-4 week intervals, detailed in methods section 7.4.8.

Confounding likely impacted the magnitude of response relative to healthy controls. Older age is associated with lower antibody titres following vaccination (Ward et al., 2022). Although AAV is typically associated with older age, the AAV population in the current study was younger on average than the control population. The median age of the AAV group was 55.3 years, while the median age for the healthy control group was within the 66-74 years bracket (the available data for the OCTAVE control group was reported in four age brackets as per Table 7-1). Therefore, if it were possible for age to be controlled for, for example via a regression analysis or for an age-matched comparator to be available, an even greater difference for anti-S outcomes between AAV patients and controls may have been expected. This is because a younger control group could be expected to have a higher proportion of seroconversion and a higher average antibody titre. However, the reported results would have been unlikely to have differed as the percentage of those who seroconverted in the healthy control group was extremely high at 222 of 225 (98.7%) individuals and the median response was not specified in the OCTAVE main study report. The main OCTAVE report did describe a substantial difference in the median anti-RBD titre of the AAV group compared to the healthy control group, reported as a z-score of 8.42 ($p<0.001$). This z-score may have had a greater magnitude if controls in OCTAVE were directly matched to AAV participants on characteristics such as age on an individual basis (Barnes et al., 2023).

The comparison between rituximab timing groups similarly may have been affected by confounding. This compared individuals in the AAV groups: those who received rituximab within 6 months (RTX 6 months) of enrolment into the study versus those who received rituximab within 6-12 months of enrolment (RTX 6-12 months). The groups differed moderately on three potential confounders which are recognised as impacting the immune response to SARS-CoV-2 vaccination: age, kidney function and concomitant immunosuppressive treatment. The RTX 6-12 months group was older (median age 58.5 years compared to 47.4 years) and had lower average kidney function (eGFR 77.2 mL/min/1.73 m^2 compared to 91.2 mL/min/1.73 m^2). These are both factors

which have been demonstrated to be associated with limited seroconversion in response to SARS-CoV-2 (Chvatal-Medina et al., 2021; Barda et al., 2021). Conversely concomitant immunosuppressive treatments were present at a lower proportion in the RTX 6-12 months group, though the difference was moderate (3 individuals [23.1%] on concomitant immunosuppression in RTX 6 months group compared to 1 individual [5.9%] in the RTX 6-12 months group) and there was not a statistically significant difference (p = 0.138, Wilcoxon test). Given that the relative impact of these potential confounders was difficult to quantify, accurately determining the effect was difficult. A multivariable model would not have been appropriate to use due to the small sample size of this subset of the AAV data. Given that age and renal function are recognised as having a substantial impact on humoral response to SARS-CoV-2 vaccination, it may be that the magnitude of the difference between this group was diminished and the true effect of time since rituximab is more clinically and statistically significant than the current data suggests.

A strength of the main OCTAVE study was that it applied consistent methodology across multiple immunosuppressed disease groups, with heterogeneous pathophysiology, clinical phenotypes and immunosuppressive treatments. However this meant that some important clinical characteristics for some of these individual conditions were not likely to incorporated. One example with respect to AAV was that CD19-positive cell counts were not measured. CD19-positive cell count depletion is well recognised as a biological marker of effective rituximab treatment. CD19-positive cells reconstitute at different rates across individuals following rituximab treatment and their suppression may be both a marker of decreased likelihood of AAV relapse and also of diminished immune response to natural infections and vaccination (Charles et al., 2018). It would have been of potential biological and clinical interest to determine if CD19-positive cell reconstitution was associated with improved immune response to SARS-CoV-2 vaccination. This may have provided clinicians with additional means of determining whether patients were likely to respond to vaccination.

The approach used to determine occurrence of Covid-19 had limitations. For this sub-study, the presence in the clinical record of a positive laboratory or near-patient test for SARS-CoV-2 was used as a proxy for Covid-19. For the purposes of the current study, it was not possible to determine whether such a test was

associated with significant Covid-19 and related symptoms, or whether the positive test was an incidental finding on asymptomatic screening. This approach may not have been optimal at identifying symptomatic Covid-19, which is ultimately a more clinically relevant outcome. If a stricter definition of Covid-19 or severe Covid-19 were to be applied, different results may have been obtained.

7.6.4 Interpretation and generalisability

This study identified a considerably worse immune response to SARS-CoV-2 vaccination in AAV patients recently treated with rituximab compared to a group of healthy controls. There was low risk that this difference was related to confounding due to age, as the comparator population was older than the AAV cohort, which, if anything, would have diminished the apparent difference between the groups. A small proportion of 8 of 29 (28%) AAV participants achieved seroconversion after the second dose of vaccine. This finding was of immediate relevance to AAV patients, clinicians and researchers. It highlighted that individuals with AAV on rituximab may have to consider different self-protective measures to non-immunosuppressed individuals. Clinicians have had to be conscious of this when counselling AAV patients regarding vaccination and self-protective measures following vaccination. The findings of the current study have implications for subsequent studies. Basic scientists will wish to understand the underlying biology of the reduced vaccine response of individuals with AAV treated with rituximab. Clinical researchers will seek to discover strategies and other therapeutic options that may help reduce the risk of the severe sequelae of Covid-19 in immunosuppressed individuals, such as the group described here. This may involve enhanced vaccination schedules, evaluation of self-protective measures and novel SARS-CoV-2 treatments. The findings of the current study are likely to be highly generalisable to individuals with AAV recently treated with rituximab. Whether the findings are as relevant for individuals with AAV not treated with rituximab or for individuals who were treated with rituximab for different clinical conditions is less clear.

Comparable studies with a significant representation of AAV patents are few in number. A retrospective analysis of 11 consecutive patients with immune-mediated kidney disease was mostly comprised of individuals with AAV. All

participants in this study had recent treatment with rituximab. Of the nine patients who did not have detectable baseline anti-RBD antibodies, three (33%) achieved a humoral response to SARS-CoV-2 vaccination with two doses. Notably, of the three responders, two had detectable CD19-positive cells, whereas among the six non-responders only one had evidence of reconstituted CD19-positive cells (Demoulin et al., 2021). A prospective study of AAV patients receiving B-cell depleting treatment examined both the humoral and T-cell responses to SARS-CoV-2 vaccination. The study included 19 AAV patients previously treated with rituximab and seven healthy controls. It found that, among CD19-positive cell deplete individuals, none mounted a detectable antibody response. All CD19-positive cell replete and all healthy controls mounted an antibody response. Somewhat reassuringly, 91% of CD19-positive cell depleted individuals mounted a detectable SARS-CoV-2 specific T-cell response (Marty et al., 2022).

A study of 140 immune-suppressed patients with autoimmune rheumatic and glomerular disease included 45 patients with vasculitis (either AAV or anti-glomerular basement membrane disease). Participants received either BNT162b2 (Pfizer/BioNTech) or ChAdOx1 nCoV-19 (Oxford–AstraZeneca) vaccines. A high proportion of the cohort, 114 (81.4%) had previously been exposed to rituximab, most had received treatment within the last six months and most were CD19-positive cell deplete. Of the 34 individuals with vasculitis who had sampling performed following the second vaccine dose, 17 (50%) did not seroconvert. All healthy controls in this study seroconverted after the second vaccine dose. The authors found a significant difference among those exposed to rituximab in terms of time since exposure: 71% seroconverted if they had received rituximab over six months prior, while only 41% seroconverted if they had received rituximab within the prior six months. Rituximab did not appear to have a major impact on T-cell response. In the CD19-positive cell deplete group, 15 of 18 (83.3%) had a detectable T-cell response (Prendecki et al., 2021).

Considering a broader immunosuppressed population, a prospective cohort study of individuals with immune-suppressive conditions examined 632 patients. Only 11 individuals had systemic vasculitis. A large majority of patients in the study only had baseline serology performed. Among those who had follow-up serology and who had not experienced prior Covid-19, the authors found that 92% of immune-suppressed participants seroconverted after two vaccine doses,

compared to 95% of healthy controls. It was not clear how many individuals with vasculitis were included in this part of the analysis (Boekel et al., 2021).

A systematic review of SARS-CoV-2 mRNA vaccine response in immune-mediated inflammatory diseases examined 25 reports. Three studies described the response after two doses of mRNA vaccine in vasculitis patients. The meta-analysis showed a substantially attenuated response in individuals with vasculitis, with 70% seroconverting. Eight studies evaluated the impact of anti-CD20 therapies, of which rituximab is the most commonly used. This found a response rate of 39% after two doses of SARS-CoV-2 mRNA vaccine. These data, and those described above, are consistent with the findings of the current study that AAV patients recently exposed to rituximab have a severely attenuated antibody response to SARS-CoV-2 vaccination.

Individuals with AAV on rituximab clearly have a blunted immune response to SARS-CoV-2 vaccination. There is also substantial variation within this population with respect to immune response. It may be that CD19-positive cell levels have a strong relationship with the antibody response. Having characterised the immune response to SARS-CoV-2 vaccination in AAV patients, it is clear that there is a high risk of some AAV patients remaining vulnerable to Covid-19, which may be severe. As well as highlighting this potentially important risk, it may be that the immune response to vaccination in this population can act as a predictor of further Covid-19 risk.

The findings of the current study are generalisable to real-world populations with AAV, specifically those recently treated with rituximab. Findings may be relevant for other individuals with AAV. Notably substantial differences between rituximab and cyclophosphamide were not identified in clinical trials in terms of their impact on likelihood of severe infections, thus it seems that, in broad terms, both therapeutic strategies equally blunt the general immune response to infection (Stone et al., 2010). It must be borne in mind however, that rituximab specifically targets the humoral immune response, so there may be important differences in the underlying immune response to both infection and vaccination for individuals who receive rituximab compared to those who receive cyclophosphamide. Therefore, caution should be used in generalising these results beyond those exposed to rituximab. As described above, a small number

of potential subjects declined participation citing reasons such as frailty and poor mobility. The AAV group was also moderately younger than a typical AAV population. Therefore, the results of this study may be less easily generalised to individuals with AAV who are particularly frail or elderly. However it seems likely, based on knowledge of the relationship between vaccination response and age, that such individuals may be even more vulnerable to a limited vaccine response than younger, less frail AAV patients (Ward et al., 2022).

7.7 Conclusion

Taken in the context of existing literature, this study provides evidence that AAV patients treated with rituximab had a substantially blunted humoral immune response, but preserved T-cell response, to SARS-CoV-2 vaccination with ChAdOx1 nCoV-19. The antibody response for the RTX 6 months group was numerically lower than the RTX 6-12 months group, though this difference was not statistically different – perhaps relating to a lack of statistical power for this question. Other literature indicates that increased time since rituximab is closely linked to an increasingly normal humoral immune response. We did not detect an association between antibody titre and T-cell response, therefore based on the current data, there is no clear evidence supporting an enhanced T-cell response compensating for a poor humoral response. This study and others described above, highlight a clear need for additional focus on individuals vulnerable to the impacts of a severely attenuated SARS-CoV-2 vaccine response. Such individuals will have likely benefited from enhanced self-protective measures such as physical distancing, mask wearing and handwashing (Talic et al., 2021). Research efforts should be directed to understand this phenomenon in greater depth and to identify potential strategies which may improve outcomes for this vulnerable group. This study described variability in terms of the immune response to SARS-CoV-2 in this AAV population. The immune response to vaccination may be a prognostic marker for future Covid-19 risk and poor outcomes and should be considered for inclusion in studies examining prognostic markers and developing prognostic scores in vulnerable populations such as those with AAV treated with potent immunosuppressive agents such as rituximab.

7.8 Summary

In earlier chapters of this thesis the incidence of severe infection in AAV, glucocorticoids as a prognostic factor for severe infection and prognostic models relating to severe infection in AAV were explored. Covid-19 in systemic vasculitis patients was then examined, by identifying prognostic factors for severe disease. The current chapter explored the immune response to SARS-CoV-2 vaccination in a vulnerable group – AAV patients recently treated with rituximab. That the antibody response to vaccination was substantially attenuated compared to healthy controls has implications for future biological, clinical and predictive research in immune-suppressive populations. The next chapter will comprise an overall discussion of the findings presented in this thesis, themes present across chapters and implications for future work.

8 Overall thesis discussion

8.1 Overview

This chapter presents a discussion of the overall thesis. Research findings from Chapters 2 to 7 will be summarised. Themes relating to strengths and limitations across the included studies will be discussed. Research findings will be placed in the context of existing related literature. The implications of these findings will be considered across research, clinical and health policy domains. Finally, future directions relating to prognosis research in AAV will be presented.

8.2 Summary of findings

8.2.1 Data quality and incidence of severe infection in AAV registries (Chapter 2)

This study implemented novel semantic web technology to harmonise and integrate AAV registry data as part of the EU FAIRVASC project. A data quality (DQ) analysis was undertaken at all seven participating pilot registries. Severe infection incidence was derived though registries which contain this data using the prototype data retrieval interface.

FAIRVASC registry data was deemed to be high quality. Uniqueness assessed the degree of duplication of patient data. Uniqueness was 100% across registries when assessed using patient registry ID. Duplication of patients under different registry IDs was 0% across all registries, with the exception of the POLVAS registry at 3.6%. Consistency according to data format was 100% across all registries and variables. Consistency when evaluated by logic tests was typically 100% and was 98% at lowest in one registry. Consistency when evaluated by plausibility tests was effectively 100%. Completeness was high for variables such as age, date of birth and date of death, but was low for some variables such as serum ANCA autoantibody status, which varied from 52% in one registry to 100% in several others. Correctness was high for most variables at 90 to 100%.

Severe infection incidence was reported for the RKD and Skane registries, both separately and combined. The combined severe infection incidence over the first

year, the second year, years 3 – 5 and beyond 5 years was 179.2, 64.2, 41.5 and 35.1 events per thousand person years, respectively.

8.2.2 Glucocorticoids as a prognostic factor for severe infection in AAV (Chapter 3)

Analysing individual prognostic factors with a targeted approach is important to gain understanding of pathophysiology and to determine variables that are important to consider for prognostic modelling studies. Glucocorticoid exposure, in daily prednisolone equivalent, was evaluated in multivariable survival models in a large population-based dataset from the VOICES study. Glucocorticoid exposure ≥10 mg was strongly predictive of severe infection, showing a 93% increased risk compared to zero mg (hazard ratio 1.93 [1.19-3.13, p=0.008]). Multiple dose thresholds were then examined. Thresholds of ≥5 mg to 10mg, ≥10 to 20 mg, >20 mg all demonstrated similar associations with increased severe infection risk of 73 – 122% increased risk (see for hazard ratios). The >0 mg to 5mg exposure group point estimate suggested decreased risk for severe infection, though confidence intervals were wide and therefore it remains unclear whether this dose range is predictive of increased or decreased risk (hazard ratio 0.72 [0.17-3.09, p=0.659]). It may be that some AAV patients have glucocorticoids prescribed in primary care for them to have available in case of relapse, as opposed to actually taking the drug. This may be due to patient or clinician concern. An additional exposure window of 5 mg to 7.5 mg was explored. Although not significant, the point estimate suggested a possible 155% increased risk for severe infection (hazard ratio 2.55 [0.72-8.94, p=0.145]). Lastly, glucocorticoid exposure was examined as a continuous variable. On average, every 1 mg increment predicts a 1% increase in severe infection risk over the subsequent year infection (hazard ratio 1.01 [1.00-1.02, p=0.047]).

8.2.3 Prediction of first severe infection in AAV (Chapter 4)

This study developed and internally validated a prediction model for first severe infection in individuals with AAV. There was a high incidence of severe infection, with 20.6% of the cohort experiencing this event within one year. A four-component prognosis model was developed. Variables included in the final model were a non-linear transformation of age, socio-economic deprivation,

presence of prior renal disease and prior diabetes. Optimism-adjusted performance measures showed fair discrimination (C-index 0.60, C/D AUC_t 0.62), very good calibration (O/E ratio 1.01, calibration intercept -0.01, calibration slope 0.94) and very low prediction error (scaled Brier Score 2.6%). A calibration plot showed that estimated risks derived from the model closely matched observed risks.

8.2.4 Prediction of early mortality after severe infection in AAV (Chapter 5)

This study developed and internally validated a prediction model for mortality prior to hospital discharge or within 30 days of discharge. The size of the study cohort was 1,015 individuals of whom 157 (15.5%) did not survive. The final model included age, time since AAV diagnosis, presence of specific comorbidities (liver disease, metastatic cancer, renal disease and diabetes) and recent glucocorticoid exposure. Optimism-adjusted performance measures included a calibration slope of 0.967, calibration-in-the-large of 0.004 and, representing discriminative ability, a c-statistic of 0.713. A calibration plot demonstrated good calibration.

8.2.5 Prognostic factors for severe Covid-19 in systemic vasculitis (Chapter 6)

This was a retrospective study of Covid-19 outcomes in systemic vasculitis patients. At the outset of the Covid-19 pandemic, cases with a history of systemic vasculitis and a diagnosis of Covid-19 were submitted to the UK and Ireland vasculitis registries using a prospectively designed case report form (CRF) or a registry web-app hosted version of the CRF. The total number of submitted cases was 105, 84 (80%) of whom had AAV. The frequency of Covid-19 symptoms and complications were reported. Dyspnoea was the most common symptom in 61% and respiratory failure was the most common complication in 53%. The composite severe outcome consisted of hospitalisation requiring advanced oxygen therapy or invasive ventilation, or death. A severe outcome was experienced in 36% of the cohort. Glucocorticoid exposure greater than 5 mg daily prednisolone dose equivalents was predictive of the severe outcome with adjusted odds ratio 4.7 (95% confidence interval 1.5 – 17.3). Glucocorticoid exposure at any dose also was associated with adjusted odds ratio 2.9 (95% CI

1.1 – 8.4). Any immunosuppressive therapy, as opposed to none, had an adjusted odds ratio 5.9 (95% CI 1.8 – 27.7). Cyclophosphamide exposure was associated with severe outcome, adjusted odds ratio 3.6 (95% CI 1.1 – 12.0). Immune outcomes to vaccination were not associated with subsequent SARS-CoV-2 positivity.

8.2.6 SARS-CoV-2 vaccine immune response in AAV (Chapter 7)

This sub-study of the UK multicentre, prospective OCTAVE study described the immune response to SARS-CoV-2 vaccination in 30 AAV patients who had received rituximab in the prior 12 months. The immune outcomes aspect of this study is a form of overall prognosis research, in that immune outcomes in response to vaccination are quantified in this group. Considering the association between baseline factors and immune response or SARS-CoV-2 infection represents prognostic factor research. A severely blunted humoral immune response was evident with only 8 of 29 (28%) of the AAV group achieving seropositivity for anti-spike antibody, while 99% of health controls were seropositive ($p < 0.0001$). This was after two vaccine doses. Only 3 of 29 individuals (10%) had a titre above the lower bound of the healthy controls. The SARS-CoV-2 specific T-cell response was robust and comparable to healthy controls. There was a numerical, but not statistically significant, positive association between time since rituximab and antibody response on univariable analysis. There was no significant correlation between antibody and T-cell response. Positivity for SARS-CoV-2 was 33.3% over one year. Younger age was associated with infection with SARS-CoV-2. Age over 50 years had hazard ratio 0.16 (0.04-0.61, p=0.008) compared to those below 50 years. Better renal function was predictive of SARS-CoV-2 infection, eGFR over 90 ml/min/1.73m^2 had hazard ratio 7.86 (1.66-37.24, p=0.009), compared to those below that threshold. Importantly these were unadjusted analyses.

8.3 Strengths and limitations

Strengths

This thesis utilised real-world data to address several important epidemiological questions relating to AAV and severe infection. FARIVASC data in Chapter 2

comes from European vasculitis registries. By their nature, registries are typically more inclusive than RCTs or clinical studies involving multiple visits. Registries represented in FAIRVASC recruit patients from wide geographies, multiple centres and across specialities resulting in a highly representative sample of AAV patients. In Chapters 3, 4 and 5 the principal source of such data was the VOICES quantitative study. This dataset was derived through advanced data linkage capabilities available in Scotland, which are not present in many other countries. Part of Public Health Scotland, the Electronic Data Research and Innovation Service (eDRIS) has access to multiple health-related datasets with national coverage. This includes demographic, socioeconomic and hospital admissions data. Mortality data is obtained from the National Records of Scotland. A nationwide patient identifier, the Community Health Index (CHI) number, enables highly accurate data linkage to be performed. Such routinely collected data has the advantage of low levels of selection bias: individuals who may be poorly represented in clinical studies due to socioeconomic status, frailty or cultural reasons will typically be included in such data. The datasets are recognised as having high levels of completeness and data quality (Public Health Scotland: Data and Intelligence, 2014). Although these are technically retrospective studies, population-based research has the advantage of data being recorded prospectively, thus ensuring that the outcome could not influence the measurement of predictors and eliminating survival bias. Employing data linkage and real-world data allowed coverage of, theoretically, the whole Scottish population, with the resultant findings being highly generalisable to other similar populations. Chapter 6 included data submitted by centres affiliated with the UK and Ireland vasculitis registries. Similar to the other FAIRVASC registries, these centres are highly geographically distributed with representation from secondary and tertiary care. As a result, such data is likely to be generalisable to other AAV populations.

This thesis aimed to address gaps in the AAV literature related to severe infection. There is a substantial need for clearly defined prognostic factor studies. Such studies should have an *a priori* design such that they aim to evaluate a specific individual prognostic factor for its ability to predict a specific outcome. Many studies in the AAV literature highlight specific prognostic factors, but often do so retrospectively, after the analysis has been done. Often the

factor has not been clearly defined in advance. This reduces confidence that such reports have not succumbed to publication bias. Chapter 3 examined the prognostic value of glucocorticoids in relation to severe infection. It had clear prespecified objectives including to evaluate novel thresholds of glucocorticoid exposure, not yet considered in the AAV literature. Chapter 5 leveraged the mature networks associated with the UK and Ireland national vasculitis registries to examine prognostic factors in a novel disease, Covid-19. Evaluating the prognostic effects of immunosuppressive therapy was a predetermined objective. The relative lack of prognosis factor research in AAV presents a challenge in the development of prognostic models. Candidate predictors for a prognostic modelling study are best identified from previous literature that has confirmed a specific prognostic factor is predictive of the outcome in question and from the knowledge and experience of expert clinicians. This thesis utilised the best available studies of predictive factors for severe infection and infection-related mortality, as well as expert clinician knowledge.

AAV is a rare disease where adequate samples for epidemiological research are challenging to construct. An important strength of this thesis was that the studies included were typically larger than all comparable research. Sufficient sample size is crucial to enable the detection of important effects, minimise spurious findings and to produce accurate estimations of effect sizes. Study size is of substantial importance in predictive epidemiology, with established tools to determine the necessary sample size in prognosis modelling studies (Riley et al., 2020). While sample size was effectively fixed in this thesis for the described studies, such sample size estimation tools were utilised to ensure that an appropriate number of candidate predictors were included in the development of prognostic models. A lack of comparable studies made estimation of parameters to enter into sample size calculations somewhat challenging, thus studies with similar objectives in other populations were used. Multiple sample size calculations were also performed under a range of different assumptions to provide added confidence that study power was being appropriately considered. While larger than many comparable studies, study power was limiting to aspects of the prognostic modelling studies, this is discussed further below. In Chapters 6 and 7, which focused on severe Covid-19 and SARS-CoV-2 vaccination

respectively, the data reported has among the highest described sample sizes for studies on these areas in AAV.

Prediction models in this thesis used modern techniques, reported results according to accepted frameworks and considered bias using a validated tool. Modern prediction modelling approaches adopted in this thesis included utilising a large data set and carefully considering study size with recommended sample size calculations (Riley et al., 2019). This resulted in the number of candidate parameters that were entered into the modelling process being appropriately limited and carefully selected. Continuous variables were appropriately handled to maximise power and minimise overfitting by avoiding dichotomisation and using fractional polynomials for non-linear modelling. The recommended automatic variable selection procedure of backward elimination was used. Internal validation was done by bootstrapping, which is recognised as resulting in stable estimates with minimal bias (Steyerberg et al., 2001). It could be considered a weakness of the prognostic models that external validation was not carried out. As Steyerberg and Harrell state, “independent validation of previous research findings is a general scientific principle”. However, they go on to clarify that if an external validation dataset is available at the time of model development, then it is more appropriate to incorporate this data into development of the initial model using an “internal-external” cross-validation procedure by which one centre is excluded, a model is developed in the remaining centres, validated in the excluded centre and then the process is repeated for all centres. The final model is derived from the full dataset. Later, when new data is available, the same or different authors can perform external validation. Arguably, internal-external validation could have been performed in the current study using different health boards to represent different centres, but given that data for the VOICES study was obtained by the same methodology through data linkage at a national level, it seems reasonable that internal validation using bootstrapping was done without attempts at internal-external validation or a somewhat forced notion of external validation (Steyerberg and Harrell, 2016). To assess clinical utility, an analysis of net benefit can also be carried out using decision curve analysis, however this is appropriate to do at the stage of formal external validation (Van Calster et al., 2018).

Well-conducted epidemiological studies involve careful consideration of potential sources of bias and this thesis has sought to minimise bias wherever possible. Confounding is a major source of bias in studies of disease aetiology. Through a focus on prognosis research, where confounders do not impact predictions, confounding was not a concerning source of bias in this thesis. Selection bias, where study associations are biased relating to procedures for selecting patients or factors affecting participation, was minimised through a focus on population-level data or maximising participation. Chapter 5 was likely affected by a form of selection bias, but this was addressed in the study question, discussed further below. A focus on novel or statistically significant results can lead to bias and subsequent publication bias. A particular form of this is "p-hacking". This is where multiple or repeated statistical analyses are conducted and "significant" results are selectively retained for reporting (Head et al., 2015). For all analyses in this thesis, the approach was prespecified and all results were treated in the same manner, whether positive or negative, statistically significant or otherwise. In the prognosis factors and prognostic modelling studies, consideration was given to potential sources of bias specific to such studies, as described in QUIPS, a tool for assessing bias in prognostic factor studies, and in PROBAST, a tool for assessing bias and applicability in prediction model studies (Hayden et al., 2013; Moons et al., 2019). Importantly, all predictors and outcomes were defined and ascertained in a highly similar way for all participants. Assessment of the presence of predictors and outcomes was made independently of each other, ensuring no bias was introduced by the researchers in question. This ensures that a prognostic association between predictors and outcomes is more reliable if detected. Advice related to statistical methodology was applied to maximise internal and external validity. Bias relating to subject participation, study attrition, predictor measurement and outcome measurement were all minimised via the population-based study design. In the Covid-19 study in Chapter 5, ascertainment bias, a form of selection bias, was a potential risk were it not appropriately addressed. Ascertainment bias occurs when some members of the target population are systematically less likely to be included in the final study sample (Freedman and Pfeiffer, 2017). This potentially leads to the study population having important differences to the target population, reducing generalisability. In Chapter 5, it was considered highly likely that more severe cases of Covid-19 would come to

the attention of submitting clinicians and therefore be more likely to be included in the study sample. For this reason, it was decided prior to data collection that outcome frequencies would not be reported as generalisable to the wider population with systemic vasculitis. A beneficial effect of this bias was a form of enrichment – as more severely affected individuals were more likely to be included in the study, the number of events will have been increased, thus increasing the power to accurately detect prognostic effects.

Limitations

A wide range of data quality dimensions were assessed in the FAIRVASC study in Chapter 2. One area where only limited checks were possible was correctness. Correctness was assessed across six registries for eight variables, if the variable was present, with at least 10 records sampled for each registry. Registry data was directly compared to a local "gold standard" source of information, such as the clinical record. In total, at least 370 data points were sampled. Given limited resources available for this task, this was a satisfactory initial check of correctness, but a more comprehensive assessment would be desirable, particularly when a research question relies upon correctness of specific variables. For example, the DQ assessment process was not designed to check data related to infection and its associated severity. For the subsequent study of severe infection incidence, it would have been useful to have had a specific assessment of correctness of severe infection data. In Chapter 2, this limitation was addressed by focusing on two registries where it was known that recent extensive data collection relating to clinical outcomes had been performed.

Chapters 3, 4 and 5 utilised the VOICES study linkage data derived from Scottish national routinely collected administrative data. While the data linkage studies in these chapters represent a novel and efficient approach to prognosis research studies, a preferred source of data for such studies may be a prospective cohort specifically designed to address the study question. Such a study would have a rigorously defined inclusion and exclusion criteria to ensure the sample represents the population of interest. Similarly, predictors and outcomes would be carefully defined. Residual confounding is a challenge in epidemiology, most prominently in aetiological research. Retrospective studies are unlikely to be able to collect all important known confounders. For prognostic studies, where

confounding is not a specific issue, the analogous concern would be missing variables that allow a specific prognostic factor to appear to have more predictive qualities, but only in the absence of other important factors. Potentially important variables of interest that were not available in this thesis include vasculitis disease activity, immunosuppressive medication data and biological characteristics such as immune system parameters. Unknown confounders or important predictors by definition will not be accounted for, though this is the case regardless of whether a study is prospective or retrospective. Prospective studies may also have weaknesses. Limited human resource, lack of access to important clinical information systems and reliance on participant self-reporting outcomes may all significantly diminish data completeness and correctness. In some circumstances, routinely collected administrative data may have more detailed and extensive data than a prospective study would be able to compile. It leads to better representation of real-world populations as discussed above. Using routinely collected data is also substantially more efficient and cost-effective. While having broad representation of important variables is important in all modelling, this thesis limited the impact of residual confounding through a clear and predefined focus on predictive research, which does not require all confounders to be included in models. Nevertheless, predictive research can still be improved upon by inclusion of a wide range of potentially important predictors.

Correctness, or accuracy, of data is crucial in epidemiological research and the retrospective design of studies in this thesis may have impacted upon this. Case ascertainment was a potential limitation in this thesis. AAV cases were identified using ICD-10 codes assigned in a national administrative data by trained clinical coders. Hospital admission data that listed an AAV code as a relevant condition was used to identify AAV cases. The first such admission was used as an index date, deemed likely to represent the time of diagnosis of a substantial proportion of study subjects. While this may suitably identify all true AAV cases, this is difficult to quantify, as it was not possible to perform a nested data quality check. Comorbid conditions were identified by similar means, which again relies on a hospital admission to pick up the condition. Data quality checks of such clinical coding is performed at a national level with such data deemed to be of high quality by national DQ processes. Glucocorticoid exposure was a key

predictor explored in the data linkage studies in this thesis. There was availability of a rich source of community prescribed medication from the PIS data set. Notably this records medications that are dispensed from community pharmacies, not the exact intended prescription from primary or secondary care. Differences between what is intended and what is dispensed may arise. AAV patients may keep a 'backup' supply of glucocorticoids in case they suffer a relapse and need immediate access to drug. Patients also may have very high glucocorticoid exposure that is not documented on community data, as the drug is supplied by secondary care. As some patients may have been exposed to glucocorticoid, even if this was not evident based on their community medication data, it raises the possibility of systemic bias in the correctness of glucocorticoid exposure data utilised in Chapters 3, 4 and 5.

Epidemiology studies often utilise factors that are more easily ascertained or measured as markers to represent exposures or outcomes of interest. This occurred in the VOICES study. Due to the nature of routinely-collected data, accuracy of some data derived for the VOICES dataset may be diminished. National administrative datasets currently utilise dedicated personnel to code data surrounding hospital admissions. Currently for hospital admissions data, clinical information relating to patients' background comorbid conditions is not optimally collected for public health or research purposes. When an individual's hospital admission is coded, one main condition and up to five additional conditions are documented. The main condition is the medical or social condition primarily responsible for the patient's need for investigation or treatment. Other conditions are those which coexist or develop during the health care episode. Therefore, it may be difficult for investigators to distinguish between new or pre-existing conditions. Many patients will have multiple issues during the health care episode. Being only able to document a maximum of five coexisting conditions means that relevant comorbid conditions may be missed. Furthermore, certain disease groups are prioritised above others for inclusion. For example, cancer and cardiovascular disease are higher priority than renal disease or hypertension (Public Health Scotland: Data and Intelligence, 2014). Where a condition is included in the data, using this data to define study populations, exposures or outcomes may still be problematic. For example, an important condition in this thesis was severe infection. A hospital

admission associated with infection was to define severe infection. This varies from the commonly used definition of “infection requiring hospital admission or requiring intravenous therapy” that is frequently used in epidemiological studies. If coded as an “other condition”, it is not possible to determine if the infection event in question was the principal reason for hospital admission or if it simply occurred around the time of the care episode and was not actually a severe event. An approach to address this could be a sensitivity analysis where only infections coded as “main condition” are used to define the outcome. A further instance of using an available measure to represent an outcome of interest was in Chapter 8. SARS-CoV-2 positivity in the clinical record was used as a marker of possible Covid-19 occurring during follow-up. While informative, it remains unclear whether a positive test for SARS-CoV-2 represented an important event for a given individual or whether the test result was an incidental finding on “routine screening”.

Adequate statistical power is crucial in clinical research to detect important signals, to limit the occurrence of spurious findings and to provide accurate estimate of effects. Although the prediction modelling studies in this thesis are substantially larger than any comparators in the AAV literature, they are still substantially smaller than high quality prediction models published in other settings with access to more data. For example, the PREDICT breast cancer survival model was developed on a dataset of 5,694 individuals and the QRISK3 model for cardiovascular event prediction was derived from a data set of 7.9 million individuals (Wishart et al., 2010; Hippisley-Cox et al., 2017). As a result, the thesis models had to limit the included number of predictors to minimise vulnerability to overfitting. The Covid-19 study in Chapter 6 was limited by sample size, despite substantial efforts to maximise case submission. As a result, it was only possible to perform multivariable analysis with a small number of covariates. Similarly, the immunological study in Chapter 7 was limited by a small sample, resulting in only basic univariable analyses being performed. A principal objective of the study, to quantify the humoral immune response in AAV patients treated with rituximab, was none the less achieved. Despite challenges around study power, biologically plausible and informative prognostic factors were still identified in these studies.

In the immunological data reported in Chapter 7, older age and better renal function were both associated with increased infection with SARS-CoV-2 over 12 months following vaccination on a univariable analysis. Crucially, this should not be interpreted as either a causative or prognostic factor, as the analysis was unadjusted. Important confounders were not possible to include, both due to the sample size being inadequate for multivariable analysis and due to lack of data on certain confounders. Older age and worse renal function are both markers of frailty. An important confounder, that would be particularly challenging to measure, is propensity towards self-protective measures against viral infection. Older, more comorbid and frail individuals would have been far more likely to adopt strict self-protective measures, which are known to reduce incidence of Covid-19 (Talic et al., 2021).

8.4 Place in literature

This thesis makes a significant contribution to the literature surrounding AAV and severe infection. The findings discussed above will be considered in the context of important related work.

Chapter 2 included an analysis of DQ in European AAV registries. High levels of DQ were found across most registries, domains and variables evaluated. DQ in FAIRVASC registries was comparable to other published reports, such as an analysis of heart failure data which found DQ scores above 95% across a range of DQ domains similar to those evaluated in Chapter 2 (Aerts et al., 2021). An analysis of the Italian National Rare Disease registry showed 100% completeness for most variables, but substantial missing data for some such as orphan drug use at 85% and date of disease onset at 53% (Taruscio et al., 2014). Methods used in the thesis study were similar to these studies. DQ in FAIRVASC registries is likely at least of as high quality as other similar registries.

Chapter 2 reported the incidence of severe infection across two major European AAV registries, across different time periods. The incidence was comparable to other reports in the published literature and reflected the predominance of severe infections in the early period after AAV diagnosis. The institute which hosts the Swedish data recently published data relating to the incidence of severe infection. Reassuringly, the Swedish incidence rates derived from the

FAIRVASC infrastructure, and reported in Chapter 2, yield highly similar results to that published on the same question from the group that hosts the southern Swedish data as part of FAIRVASC. The study in question utilised the same data as FAIRVASC and their analysis had comparable, but not identical, methodology (Rathmann et al., 2021).

Chapter 3 investigated glucocorticoid exposure across a range of dose thresholds, many of which have not been explored in the AAV literature. As detailed in Chapter 1 (section 1.13.2), dose thresholds have been examined in adjacent IMIDs such as RA. George and colleagues found that low-dose prednisone exposure up to 5 mg conferred a clinically important increased risk for severe infection (George et al., 2022). While this may be assumed to apply to AAV also, AAV has substantially different underlying disease processes, therapeutic strategies and propensity to severe infection. The results presented in Chapter 3 confirm that low-dose glucocorticoid exposure between 5 mg to 7.5 mg may confer substantially increased severe infection risk. It also adds to the body of literature that supports glucocorticoid exposure at higher doses as a major contributor to severe infection risk (Lai et al., 2014; Walsh et al., 2022).

Chapter 4 described the first clinical prediction model developed in AAV using a large routinely collected data set. It reported that 20.6% of AAV patients experienced a severe infection event, consistent with reports detailed in the introductory chapter that 15 to 22% will have this outcome. Only one other study, by McClure and colleagues, reported a comparable prediction model for severe infection. That study examined a different question, aiming to predict severe infections or three non-serious infections over five years following maintenance treatment with rituximab. The study sample size was 147 patients, whereas the study in Chapter 4 included 2,078 patients. The McClure study yielded an optimism-adjusted C-index of 0.64, comparable to the study in this thesis. An advantage of the McClure study was access to useful biological parameters such as serums IgG levels. A limitation was consideration of sample size. Their modelling included 13 candidate parameters, while only having 88 clinical events (Events per candidate predictor (EPP) = 6.7). This leaves the study vulnerable to overfitting. The study in this thesis had 11 candidate predictors and 438 events (EPP = 39.8), however non-linear modelling was used for continuous variables which will substantially decrease the effective EPP. Age

was also unnecessarily dichotomised in the McClure study, thus losing information and potentially degrading model performance, as opposed to the non-linear approach used in this thesis.

Chapter 5 derived the first known prognosis model that predicts mortality in individuals with AAV and severe infection. The optimism adjusted c-statistic for this model was 0.71. Similar models exist for the general population such as the Mortality in Emergency Department Sepsis (MEDS) score and the Pneumonia Severity Index and CURB-65 scores for community acquired pneumonia. The MEDS score was derived in 2,070 patients and had a c-statistic of 0.78 in a validation data set (Shapiro et al., 2003). The Pneumonia Severity Index had a c-statistic of 0.81 in a validation cohort, while CURB-65 had a c-statistic of 0.76 in the same cohort. The model in this thesis has not yet been validated in an external data set, however internal validation was performed using bootstrapping. Our model is the only known to be targeted at a highly immunosuppressed population, such as AAV.

Chapter 6 examined the immune response to SARS-CoV-2 vaccination and occurrence of subsequent SARS-CoV-2, in an AAV population treated with rituximab. Only other small studies have been reported. A study of 11 AAV patients treated with rituximab found that only three seroconverted (Demoulin et al., 2021). A similar study of 19 AAV patients found that among CD19-positive cell deplete individuals, none mounted a detectable antibody response. All CD19-positive cell replete individuals did however mount an antibody response (Marty et al., 2022).

8.5 Implications

8.5.1 Research implications

Ensuring high quality data for clinical research is clearly important, but there are challenges in applying data quality concepts and methodology. Interpretation of DQ results is also challenging, as often the relative importance of DQ domains such as completeness and correctness is unknown. Different epidemiological studies and different areas of clinical research likely require different thresholds of DQ and the importance of DQ domains may vary across

studies. The field would benefit from work that quantifies the effect of differing data quality in studies with different objectives and across clinical domains. Researchers should be aware of DQ and apply principles and methodology from the field in their work. Starting with high quality data is crucial and Chapter 2, section 2.6.3, contains suggestions for optimising this.

One of the main objectives of FAIRVASC was to develop semantic web-based technology to make rare disease registry data "FAIR" – findable, accessible, interoperable and reusable. AAV was selected as the exemplar condition. The EU-funded consortium has demonstrated that the FAIRVASC infrastructure gives "proof of concept" that semantic web technology can be used effectively to federate registry data and achieve FAIR objectives. This role of this type of infrastructure can now be expanded to other rare disease settings and beyond, to make registry, biobank and other biomedical data increasingly FAIR. This is compatible with data protection legislation and would increasingly allow researchers to harness research data to improve patient outcomes. The study in Chapter 2 described incidence rates of severe infection. By combining two registries with high quality severe infection data, these results are based on the largest data set yet published on the topic. This provides highly accurate estimates of incidence, which will facilitate future prognosis research.

There is a greater need for clearly defined prognostic factor research in the medical literature in general and AAV is no exception. Many studies are performed without clearly stating that the objectives are aetiological research or predictive research. The PROGRESS framework, whose themes are incorporated into this thesis, serves as an excellent structure on which researchers can clearly define and report high-quality prognosis research (Hemingway et al., 2013).

Glucocorticoid exposure has been established as strongly predictive of severe infection both in this thesis and in other studies (McGregor et al., 2012). It also has an aetiological role, most clearly evidenced in the PEXIVAS therapeutic trial and other randomised data (Walsh et al., 2022). There is a need to study glucocorticoid exposure as a prognostic factor with greater statistical power and an aim to determine dose thresholds that impact risk. Risk may not be linear, therefore modern approaches to non-linear continuous variable modelling should

be utilised. Understanding the underlying mechanisms by which glucocorticoids confer increased risk could pave the way for novel therapeutic strategies to decrease severe infections and their associated mortality. A more comprehensive understanding of the impact of glucocorticoid dose will provide a strong foundation for subsequent prognosis modelling studies and stratified medicine research.

The prognostic modelling studies in this thesis apply modern statistical methodology to highly generalisable real-world data sets with careful consideration of sample size. These studies have not undergone external validation or assessment of clinical impact. Future prognostic modelling studies in these areas should seek to add additional prognostic factors which were not available in this thesis. In their current form, the models are not appropriate to deploy in clinical practice. This work does, however, represent important first steps in developing the prognostic modelling field in the setting of AAV and severe infection. Such models could be used in therapeutic trials to identify patients at risk of serious infection. Novel therapies or approaches to clinical care could be evaluated in RCTs examining such high-risk populations.

8.5.2 Clinical implications

Many AAV patients are maintained on long-term low-dose glucocorticoid, particularly in EGPA (Hellmich et al., 2023; Kitching et al., 2020). While this may be an appropriate strategy for some patients to control disease activity, it should not be routinely employed for all patients. Chapter 2 raised concern that low-dose glucocorticoid between 5 mg to 7.5 mg may be associated with a clinically important risk of severe infection. When making decisions with patients about ongoing glucocorticoid therapy, the risk of severe infections should be considered.

As described above, the models reported in this thesis are not appropriate for deployment in routine clinical care. In future, models informed by this thesis will ideally be used in the routine care of AAV patients which could inform patients about what to expect in the future and could assist with the application of stratified medicine: the selection of individualised treatments to maximise effectiveness and minimise adverse events.

Chapter 7 demonstrated the substantially diminished humoral immune response to SARS-CoV-2 vaccination in rituximab treated AAV patients. This had direct clinical implications for AAV patients. Based on this data, it was not possible to reassure such patients that their immune response to vaccination was normal. It was important for many patients to know that protection afforded by vaccination may be limited. As a result, they were able to consider adopting stricter self-protection measures. There was, however, a partially reassuring message from the data, that the T-cell response appeared to be functioning similarly to healthy controls.

An important message from this thesis is that AAV, and importantly its therapy, confer a high risk of severe infection. While adverse outcomes in this setting are many, infection does not have a prominent place in communication with patients, whether in the clinical setting or in patient information literature. Communication around what patients can expect following a diagnosis should be patient centred and based on the best available data. Severe infection may need more prominence in discussions with patients and in information given to them.

8.5.3 Health policy implications

Prognosis research tools have a wide range of potential applications such as helping clinicians and patients make treatment decisions based on individualised risk, communicating prognosis to patients and families and for selecting patients at high risk of a given outcome for therapeutic trials aiming to ameliorate that risk. Applications in health policy can also be envisioned. Prognostic tools could be applied to specific groups to forecast the likelihood of clinical events at a population level. This could enable optimisation of the structure and funding of services by taking future events into account. In general, as the quality and predictive ability of prognostic tools improve, their integration into routine clinical practice should be considered and facilitated. Incorporating useful clinical prognosis tools into electronic health care applications in a convenient and intuitive manner would be highly valuable for clinicians and patients. In order to facilitate future delivery of such tools, consideration should be given to optimising routinely collected data for population-based research, including prognosis research. Administrative data sets could be altered, even in subtle ways, to enable better population research, which in turn could provide direct

clinical benefit in a relatively short time frame. An example of such a potential change is the number of comorbid conditions that is possible to derive from a care episode in the current Scottish administrative data. As detailed above in section 8.3, there are limitations to the number of comorbid conditions that existing clinical coders are able to document for a given care episode. Artificial intelligence (AI) tools are under development aiming to summarise medical records (Zhang et al., 2023). Utilising such tools in future could enable more comprehensive documentation of patient's health conditions, while existing human resource could be deployed to adjudicate cases where an AI tool's results have low certainty and for auditing of clinical coding. Ultimately, clinical coding and data linkage should be adequately resourced to maximise the utility of data for patient benefit, clinical research and policy making.

8.6 Future directions

An objective of the FAIRVASC consortium is to expand use cases for federated research data such that advance statistical techniques can be used to analyses the data in greater depth. At present semantic web technology typically only allows basic arithmetic operations to be performed. The FAIRVASC consortium is investigating the use of novel encrypted federated learning techniques that will allow such statistical techniques, including machine learning, to be performed on distributed data, meaning that the underlying data will not be transferred out of the host institution for the analysis. Crucially this allows any concerns around data protection to be comprehensively addressed. More complex predictive models will be able to be developed on suitably large datasets to better predict outcomes for patients. The FAIRVASC infrastructure is highly scalable, therefore can be extended to other AAV data sets, other rare disease settings and beyond.

Chapter 2 highlighted glucocorticoids as a prognostic factor for severe infection in AAV. Multiple observational studies and randomised data support an aetiological role for glucocorticoids in this regard. Research into the underlying biological mechanisms underlying development of severe infection in AAV, including the biology of the glucocorticoid effect, is warranted. This could reveal biomarkers for severe infection risk that could be incorporated into increasingly powerful prognostic tools. Severe infections represent a huge global

burden in terms of morbidity and mortality. Understanding the biology could have beneficial effects far beyond AAV.

It is clear that severe infections are common in AAV and can lead to a substantial impact on morbidity and mortality. However, quantification of the morbidity impact is lacking and the mechanisms by which it leads to or predicts mortality in AAV are not understood. It is not clear whether a non-fatal severe infection simply predicts other severe infections, which could be fatal, or impacts aspects of physiology, vasculitis treatment or other disease processes that leads to early mortality in a causative fashion. Quality of life (QOL) is highly likely to be impacted be severe infection, in the very least due to excess time spent in hospital and patient anxiety. However, it is unclear if other aspects of QOL are impacted. Prospective studies of patients with AAV who have suffered infections could address these questions by assessing aspects of biology, clinical care and outcomes. Comparison to controls, including AAV patients who have not suffered infections would be important.

Prognosis research will be of increasing importance in all disease settings, including AAV. Adverse clinical events in AAV include cardiovascular disease, malignancy, venous thromboembolism and severe infection, which this thesis has presented as an important, unmet clinical need. There is a clear requirement for adequately powered, prospective prognosis research in these areas. This would include thorough assessment of known, and novel biomarkers; prognostic factors; prognostic modelling studies executed using modern methology described in this thesis; external validation and clinical impact studies of these models, which is often missing from the wider medical literature; and stratified medicine research to help patients use prognostic tools to choose treatment strategies that maximise clinical benefit while minimising adverse effects.

Many studies have demonstrated that the most common cause of early mortality in AAV is severe infection. Other work has highlighted the severe increase in excess mortality in AAV due to infection. More patients die of severe infection than active vasculitis. It is clear that our treatment strategies are not adequately tailored to minimise this risk. Individuals with AAV have benefitted considerably from the international vasculitis research community's ability to execute therapeutic trials despite the rarity of the disease. Future

interventional trials should be focused therapies and strategies to reduce adverse outcomes, such as severe infection and infection-related mortality. This may include tailoring therapy according to individualised data from prognostic tools or enhanced antimicrobial prophylaxis in those at the highest risk.

8.7 Conclusion

It is well recognised that individuals with AAV are vulnerable to the potentially devastating impacts of severe infection. Based on a structure provided by the prognosis research literature and the PROGRESS framework, this thesis has explored the high incidence of severe infection in AAV, the potential for glucocorticoid exposure as a prognostic marker for severe infection and prognostic modelling of both the occurrence of severe infection and early death after the event. A novel infectious disease, Covid-19, was the subject of chapters which investigated prognostic factors for severe disease and the immunological impact of SARS-CoV-2 vaccination in this high-risk population. Prognosis research has the potential to deliver immense health benefits to individuals with rare diseases. Its increased application, in aiming to predict severe infection in AAV, is no exception.

9 Appendices

9.1 Literature search

Embase 1947-Present, updated daily

1 exp ANCA associated vasculitis/ 7788
2 (Anti-Neutrophil Cytoplasmic Antibody-Associated Vasculitis or Anti Neutrophil Cytoplasmic Antibody Associated Vasculitis or ANCA-Associated Vasculitis or ANCA Associated Vasculitis or Pauci-Immune Vasculitis or Pauci Immune Vasculitis or Pauci-Immune Vasculitides or ANCA-Associated Vasculitides or ANCA Associated Vasculitides or ANCA-Associated Vasculitide or Churg-Strauss Syndrome or Churg Strauss Syndrome or Churg-Strauss Vasculitis or Eosinophilic Granulomatous Vasculitis or Eosinophilic Granulomatous Vasculitides or Granulomatosis with Polyangiitis or Granulomatosis with Polyangiitides or Wegener Granulomatosis or Wegener's Granulomatosis or Wegeners Granulomatosis or Microscopic Polyangiitis or Microscopic Polyangiitides or Necrotizing vasculi$ or antineutrophil cytoplasmic antibody-associated vasculitis or Anti-neutrophil cytoplasmic antibody ANCA associated vasculitis or Antineutrophil cytoplasmic autoantibody-associated vasculitis).mp. [mp=title, abstract, heading word, drug trade name, original title, device manufacturer, drug manufacturer, device trade name, keyword heading word, floating subheading word, candidate term word] 29491
3 1 or 2 29491
4 exp infection/ 4159535
5 (Infectio$ or Antibiotic$ or Antimicrobial$ or antivi$ or Pathogen$ or Bacter$ or Sepsis or Septic$ or Microbe$ or Fungus or Fungal or Fungemia or Fungaemia or Parasite or Parasitic or Virus$ or Viral).mp. [mp=title, abstract, heading word, drug trade name, original title, device manufacturer, drug manufacturer, device trade name, keyword heading word, floating subheading word, candidate term word] 6751809
6 4 or 5 7737736
7 3 and 6 12125
8 (Epidemiol$ or retrospective or Case-Control or Matched$ or case control or case-control or (Cohort and study) or Matched-cohort$ or Observ$ or Longitudinal or cross sectional or cross-sectional or predict$ or scor$ or Data Linkage or Risk$ or Incidence or Prevalence or Multivaria$ or Regression or Population$).mp. [mp=title, abstract, heading word, drug trade name, original title, device manufacturer, drug manufacturer, device trade name, keyword heading word, floating subheading word, candidate term word] 15737462
9 7 and 8 5529
10 (animals/ not humans/) or comment/ or editorial/ or exp review/ or meta analysis/ or consensus/ or exp guideline/ 5151934
11 hi.fs. or case report.mp. 2860812
12 10 or 11 7902305
13 9 not 12 3350

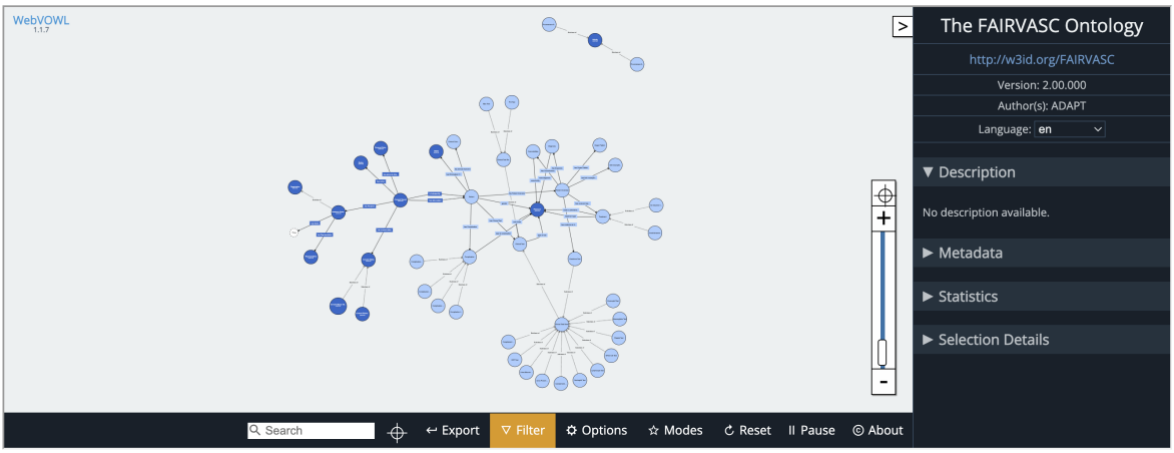
Ovid MEDLINE(R) and In-Process, In-Data-Review & Other Non-Indexed Citations <1946 to February 25, 2022>

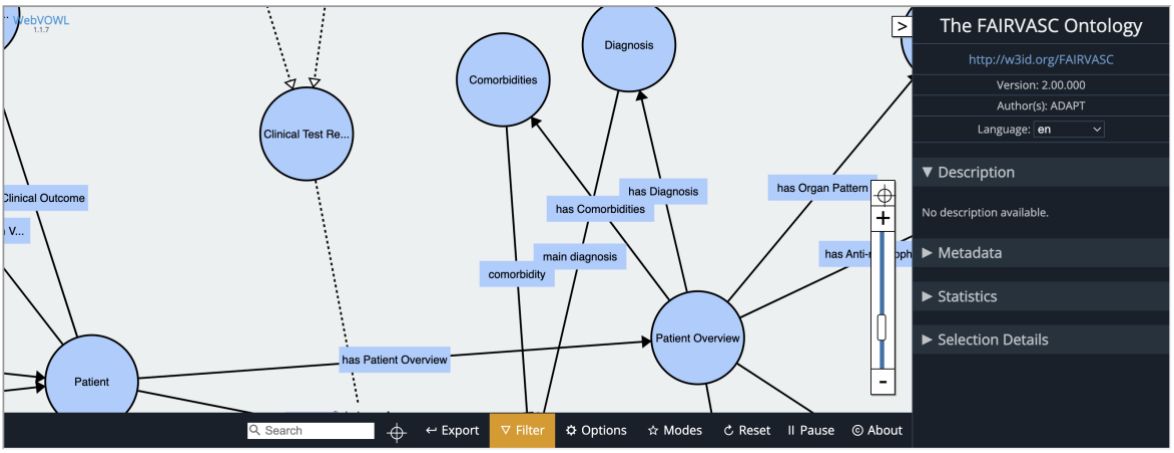
1 exp Anti-Neutrophil Cytoplasmic Antibody-Associated Vasculitis/ 11518

2 (Anti-Neutrophil Cytoplasmic Antibody-Associated Vasculitis or Anti Neutrophil Cytoplasmic Antibody Associated Vasculitis or ANCA-Associated Vasculitis or ANCA Associated Vasculitis or Pauci-Immune Vasculitis or Pauci Immune Vasculitis or Pauci-Immune Vasculitides or ANCA-Associated Vasculitides or ANCA Associated Vasculitides or ANCA-Associated Vasculitide or Churg-Strauss Syndrome or Churg Strauss Syndrome or Churg-Strauss Vasculitis or Eosinophilic Granulomatous Vasculitis or Eosinophilic Granulomatous Vasculitides or Granulomatosis with Polyangiitis or Granulomatosis with Polyangiitides or Wegener Granulomatosis or Wegener's Granulomatosis or Wegeners Granulomatosis or Microscopic Polyangiitis or Microscopic Polyangiitides or Necrotizing vasculi$ or antineutrophil cytoplasmic antibody-associated vasculitis or Anti-neutrophil cytoplasmic antibody ANCA associated vasculitis or Antineutrophil cytoplasmic autoantibody-associated vasculitis).mp. [mp=title, abstract, original title, name of substance word, subject heading word, floating sub-heading word, keyword heading word, organism supplementary concept word, protocol supplementary concept word, rare disease supplementary concept word, unique identifier, synonyms] 16490
3 1 or 2 16490
4 exp Infections/ 2883889
5 (Infectio$ or Antibiotic$ or Antimicrobial$ or antivi$ or Pathogen$ or Bacter$ or Sepsis or Septic$ or Microbe$ or Fungus or Fungal or Fungemia or Fungaemia or Parasite or Parasitic or Virus$ or Viral).mp. [mp=title, abstract, original title, name of substance word, subject heading word, floating sub-heading word, keyword heading word, organism supplementary concept word, protocol supplementary concept word, rare disease supplementary concept word, unique identifier, synonyms] 4927623
6 4 or 5 5756114
7 3 and 6 4748
8 (Epidemiol$ or retrospective or Case-Control or Matched$ or case control or case-control or (Cohort and study) or Matched-cohort$ or Observ$ or Longitudinal or cross sectional or cross-sectional or predict$ or scor$ or Data Linkage or Risk$ or Incidence or Prevalence or Multivaria$ or Regression or Population$).mp. [mp=title, abstract, original title, name of substance word, subject heading word, floating sub-heading word, keyword heading word, organism supplementary concept word, protocol supplementary concept word, rare disease supplementary concept word, unique identifier, synonyms] 11372668
9 7 and 8 1887
10 (animals/ not humans/) or comment/ or editorial/ or exp review/ or meta analysis/ or consensus/ or exp guideline/ 9076672
11 hi.fs. or case report.mp. 660103
12 10 or 11 9648633
13 9 not 12 1301

9.2 FAIRVASC ontology

Screenshots of interactive ontology visualisation from http://ontologies.adaptcentre.ie/fairvasc/index-en.html:





Example of FAIRVASC ontology coded in JSON format: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  "@id" : "_:genid1",
  "@type" : [ "http://www.w3.org/2002/07/owl#Restriction" ],
  "http://www.w3.org/2002/07/owl#hasValue" : [ {
    "@id" : "http://identifiers.org/ncit:C64548"
  } ],
  "http://www.w3.org/2002/07/owl#onProperty" : [ {
    "@id" : "http://w3id.org/FAIRVASC#testType"
  } ]
}, {
  "@id" : "_:genid10",
  "@type" : [ "http://www.w3.org/2002/07/owl#Restriction" ],
  "http://www.w3.org/2002/07/owl#hasValue" : [ {
    "@id" : "http://identifiers.org/ncit:C67255"
  } ],
  "http://www.w3.org/2002/07/owl#onProperty" : [ {
    "@id" : "http://w3id.org/FAIRVASC#testUnit"
  } ]
}, {
```

```
  "@id" : "_:genid11",
  "@type" : [ "http://www.w3.org/2002/07/owl#Restriction" ],
  "http://www.w3.org/2002/07/owl#hasValue" : [ {
    "@id" : "http://identifiers.org/ncit:C64848"
  } ],
  "http://www.w3.org/2002/07/owl#onProperty" : [ {
    "@id" : "http://w3id.org/FAIRVASC#testType"
  } ]
}, {
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  "@type" : [ "http://www.w3.org/2002/07/owl#Restriction" ],
  "http://www.w3.org/2002/07/owl#hasValue" : [ {
    "@id" : "http://identifiers.org/ncit:C64783"
  } ],
  "http://www.w3.org/2002/07/owl#onProperty" : [ {
    "@id" : "http://w3id.org/FAIRVASC#testUnit"
  } ]
```

9.3 Glucocorticoid dose exposure: full models

Table 9-1 | Glucocorticoid 0 mg vs >0-10 mg vs >10 mg: full model

		n (%)	HR (95% CI): univ
Glucocorticoid: 0mg vs >0 to 10mg vs >10mg	Zero	290 (29.7)	-
	>0 to 10 mg	120 (12.3)	1.61 (0.84-3.08, p
	>= 10 mg	568 (58.1)	1.67 (1.04-2.67, p
Age	Mean (SD)	59.7 (15.3)	1.03 (1.01-1.04, p
Sex	Female	513 (52.5)	1.51 (1.03-2.22, p
Diabetes		65 (6.6)	1.68 (0.90-3.14, p
Cancer (all)		54 (5.5)	1.35 (0.66-2.77, p
Chronic respiratory disease		180 (18.4)	2.17 (1.46-3.24, p
Chronic heart failure		31 (3.2)	2.28 (1.06-4.90, p
Liver disease		15 (1.5)	1.93 (0.61-6.08, p
Renal disease		92 (9.4)	3.50 (2.27-5.41, p
CVD		47 (4.8)	2.42 (1.30-4.51, p
SIMD (deciles)	Mean (SD)	5.6 (2.8)	0.98 (0.92-1.05, p

95% CI = 95 percent confidence interval, CVD = cerebrovascular disease, mg = milligrams, SD = standard deviation, SIMD = Scottish Index of Multiple Deprivation.

Table 9-2 | Glucocorticoid multiple dose thresholds: full model

		n (%)	HR (95% CI):
Glucocorticoid: 0mg vs >0 to 10mg vs >10mg	Zero	290 (29.7)	-
	>0 to 5 mg	38 (3.9)	0.64 (0.15-2.
	>5 to 10 mg	82 (8.4)	2.10 (1.06-4.
	>10 to 20 mg	198 (20.2)	1.64 (0.93-2.
	>20 mg	370 (37.8)	1.68 (1.02-2.
Age	Mean (SD)	59.7 (15.3)	1.03 (1.01-1.
Sex	Female	513 (52.5)	1.51 (1.03-2.
Diabetes		65 (6.6)	1.68 (0.90-3.
Cancer (all)		54 (5.5)	1.35 (0.66-2.
Chronic respiratory disease		180 (18.4)	2.17 (1.46-3.
Chronic heart failure		31 (3.2)	2.28 (1.06-4.
Liver disease		15 (1.5)	1.93 (0.61-6.
Renal disease		92 (9.4)	3.50 (2.27-5.
CVD		47 (4.8)	2.42 (1.30-4.
SIMD (deciles)	Mean (SD)	5.6 (2.8)	0.98 (0.92-1.

95% CI = 95 percent confidence interval, CVD = cerebrovascular disease, mg = milligrams, SD = standard deviation, SIMD = Scottish Index of Multiple Deprivation.

Table 9-3 | Glucocorticoid 5-7.5 mg dose: full model

		n (%)	HR (95% CI): univariable	H
Glucocorticoid: 0mg vs 5-7.5 mg	Zero	290 (93.5)	-	-
	5 to 7.5 mg	20 (6.5)	1.98 (0.59-6.59, p=0.267)	2
Age	Mean (SD)	58.4 (17.1)	1.03 (1.00-1.05, p=0.053)	1
Sex	Female	180 (58.1)	1.17 (0.53-2.57, p=0.704)	0
Diabetes		20 (6.5)	1.31 (0.31-5.53, p=0.716)	0
Cancer (all)		21 (6.8)	0.54 (0.07-3.97, p=0.544)	0
Chronic respiratory disease		55 (17.7)	2.18 (0.95-5.02, p=0.067)	2
Chronic heart failure		7 (2.3)	4.53 (1.07-19.20, p=0.040)	2
Liver disease		7 (2.3)	0.00 (0.00-Inf, p=0.997)	0
Renal disease		31 (10.0)	4.48 (1.95-10.31, p<0.001)	4
CVD		20 (6.5)	3.80 (1.43-10.08, p=0.007)	3
SIMD (deciles)	Mean (SD)	5.4 (2.9)	1.02 (0.89-1.16, p=0.823)	0

95% CI = 95 percent confidence interval, CVD = cerebrovascular disease, mg = milligrams, SD = standard deviation, SIMD = Scottish Index of Multiple Deprivation.

Table 9-4 | Glucocorticoid dose - continuous variable: full model

		n (%)	HR (95% CI): univariable	HR (95%
Glucocorticoid dose (mg)	Mean (SD)	20.3 (23.1)	1.00 (1.00-1.01, p=0.314)	1.01 (1.0
Age	Mean (SD)	59.7 (15.3)	1.03 (1.01-1.04, p<0.001)	1.02 (1.0
Sex	Female	513 (52.5)	1.51 (1.03-2.22, p=0.034)	1.28 (0.8
Diabetes		65 (6.6)	1.68 (0.90-3.14, p=0.101)	1.10 (0.6
Cancer (all)		54 (5.5)	1.35 (0.66-2.77, p=0.412)	1.09 (0.5
Chronic respiratory disease		180 (18.4)	2.17 (1.46-3.24, p<0.001)	1.85 (1.2
Chronic heart failure		31 (3.2)	2.28 (1.06-4.90, p=0.035)	1.25 (0.5
Liver disease		15 (1.5)	1.93 (0.61-6.08, p=0.262)	1.77 (0.5
Renal disease		92 (9.4)	3.50 (2.27-5.41, p<0.001)	2.87 (1.8
CVD		47 (4.8)	2.42 (1.30-4.51, p=0.005)	2.21 (1.2
SIMD (deciles)	Mean (SD)	5.6 (2.8)	0.98 (0.92-1.05, p=0.611)	0.99 (0.9

95% CI = 95 percent confidence interval, CVD = cerebrovascular disease, mg = milligrams, SD = standard deviation, SIMD = Scottish Index of Multiple Deprivatio

9.4 Example UKIVAS Covid-19 case report form

Notes

- This form is designed to be used either electronically **OR** by printing and scanning
- For UKIVAS sites patient information can also be submitted via ukivas.org – please refer to the accompanying flow chart or contact ggc.vasculitis-covid@nhs.scot for advice
- Please email completed forms to **ggc.vasculitis-covid@nhs.scot**
- We understand you will likely be short of time at present, in which case we would be grateful if you could prioritise the sections highlighted in light blue. **We understand you may not be able to complete the blue section** in which case please keep note of the patient for retrospective data collection and, if possible, email us minimal information.

Baseline information (highlighted in light blue for prioritisation)

Patient information	
Age at C-19 diagnosis (years)	
Gender (please select one)	☐ Male ☐ Female ☐ Other
Country of birth	
Vasculitis diagnosis	
Date of formal vasculitis diagnosis	
Vasculitis on biopsy	☐ not done ☐ positive ☐ negative ☐ non-diagnostic
Vasculitis diagnosis confidence	☐ Possible ☐ Probable ☐ Definite

Past vasculitis organ system involvement	☐ cutaneous ☐ mucous membranes ☐ eyes ☐ ENT ☐ chest ☐ cardiovascular ☐ abdominal ☐ renal ☐ nervous system ☐ other Detail for 'other' / any comments:

Desirable information (highlighted in light blue for prioritisation)

Comorbidities		
Comorbidities (check all that apply)	☐ Asthma ☐ Atrial fibrillation ☐ Cerebrovascular accident (stroke or TIA) ☐ COPD (not including late onset asthma) ☐ Coronary heart disease (Ischaemic heart disease) ☐ Diabetes mellitus ☐ Hyperlipidaemia ☐ Hypertension requiring treatment ☐ Hypothyroidism ☐ Renal disease (kidney disease) ☐ Thromboembolic disease ☐ Obesity (BMI >= 40) ☐ Pulmonary HTN ☐ Other lung disease (non-vasculitis e.g. ILD) ☐ H/O organ transplant ☐ Myocardial infarction	☐ Congestive cardiac failure ☐ Peripheral vascular disease ☐ Dementia ☐ Connective tissue disorder ☐ Peptic ulcer disease ☐ Liver disease ☐ Hemiplegia ☐ Solid tumour – if present: ☐ localised OR ☐ metastatic ☐ Leukemia ☐ Lymphoma ☐ AIDS ☐ None ☐ Other (free text):

Vasculitis / Disease status at time of C-19 diagnosis	
Disease activity at this encounter (physician global, please check one)	☐ 1 Remission ☐ 2 Minimal or Low disease activity ☐ 3 Moderate disease activity ☐ 4 Severe or High disease activity ☐ 5 Unknown

Immunosuppressive Medication *** Medication questions relate to those taken at time of COVID-19 diagnosis (or up to 2 weeks prior)** *** For medications with prolonged duration of action (e.g. rituximab), the patient should be considered 'on' this medication if this medicine is thought to be still having its clinical effect**		
Immunosuppressive status (please check one)	☐ Currently on immunosuppression ☐ Discontinuation of immunosuppression within 6 months prior to this encounter ☐ Discontinuation of immunosuppression > 6 months prior to this encounter ☐ Treatment Naïve	
If above answer 'Treatment Naïve' please move to next section 'Other medication'		
On corticosteroids?	☐ Yes ☐ No	
If on corticosteroids, current corticosteroid dose:	mg (in oral prednisolone equivalents)	
On other immunosuppressive medication? (please check all that apply)	☐ Abatacept ☐ Alfa1 antitrypsin ☐ Anakinra ☐ Apremilast ☐ Azathioprine ☐ Belimumab ☐ Bevacizumab ☐ Ciclesonide ☐ Ciclosporin ☐ Chloroquine ☐ Cyclophosphamide - Oral ☐ Cyclophosphamide - IV ☐ Hydroxychloroquine ☐ IVIG: Immunoglobulins ☐ Leflunomide ☐ Mepolizumab ☐ Methotrexate ☐ Mycophenolate mofetil	☐ Rituximab ☐ Secukinumab ☐ Sulfasalazine ☐ Tacrolimus (including Advagraf, Prograf, etc.) ☐ Thalidomide ☐ Tocilizumab ☐ Tofacitinib ☐ Tumor necrosis factor alpha (TNF-) inhibitors ☐ Ustekinumab ☐ None ☐ Other (please specify):
If on corticosteroids, dose change in response to this clinical encounter/episode	☐ Increased ☐ No change ☐ Reduced	☐ Stopped ☐ Unknown
If on other Immunosuppressive medication, dose change in response to this clinical encounter/episode	Immunosuppressive 1 name:	☐ Immunosuppressive increased ☐ No change in Immunosuppressive ☐ Immunosuppressive reduced ☐ Immunosuppressive stopped ☐ Unknown
If on 2nd other Immunosuppressive medication, dose change in response to this clinical encounter/episode	Immunosuppressive 2 name:	☐ Immunosuppressive increased ☐ No change in Immunosuppressive ☐ Immunosuppressive reduced ☐ Immunosuppressive stopped ☐ Unknown
If on > 2 immunosuppressive please provide details regarding dose change in response to this encounter / episode here:		

Other medication (please check one box for each drug class)			
Angiotensin-converting-enzyme inhibitor (ACE-i) at C19 diagnosis	☐ Yes	☐ No	☐ Unknown
Angiotensin II receptor blocker (ARB) at C19 diagnosis	☐ Yes	☐ No	☐ Unknown
Non-steroidal anti-inflammatory drug (NSAID) at C19 diagnosis	☐ Yes	☐ No	☐ Unknown

COVID-19 – initial assessment		
Date of C-19 symptom onset (if known)		
Date of C-19 diagnosis		
Admission to hospital required (check one box) **If** Yes: Date of admission	☐ Yes ☐ No ☐ Unknown	
Admission to ICU during admission (check one box) **If** Yes: Date of admission	☐ Yes ☐ No ☐ Unknown	
Clinical features at outset (check all that apply)	☐ Fever ☐ Malaise ☐ Headache ☐ Irritability or confusion ☐ Arthralgia ☐ Myalgia ☐ Conjunctivitis ☐ Rhinorrhea ☐ Sore Throat ☐ Cough ☐ Sputum production ☐ Shortness of Breath ☐ Chest pain	☐ RR >24 breaths/min ☐ Abdominal pain ☐ Nausea ☐ Vomiting ☐ Diarrhoea ☐ Altered taste ☐ Altered smell ☐ Haemoptysis ☐ Epistaxis ☐ **None** (asymptomatic) ☐ Unknown ☐ **Other:** (please specify)

COVID-19 – treatment and outcomes	
Were antibiotics administered?	☐ Yes ☐ No ☐ Unknown
Was treatment administered for C-19 infection (other than best supportive care)? (check all that apply)	☐ No treatment except supportive care ☐ Kaletra (Lopinavir/ritonavir) ☐ Remdesivir ☐ Chloroquine ☐ Hydroxychloroquine ☐ Neuraminidase inhibitors, direct acting antivirals (e.g. Oseltamivir) ☐ Azithromycin ☐ Tocilizumab ☐ Bevacizumab ☐ Tofacitinib ☐ Ciclesonide ☐ Plasma (from recovered patients) ☐ Other (please specify):
Complications / Disease Course (check all that apply)	☐ Acute Respiratory Distress Syndrome (ARDS) ☐ Acute respiratory failure ☐ Pneumothorax ☐ Acute liver injury ☐ Acute heart failure ☐ Myocarditis ☐ Cardiac arrhythmia ☐ Cardiac ischaemia ☐ Acute Kidney Injury (AKI) ☐ Sepsis ☐ Vasopressor dependence at any time ☐ Disseminated Intravascular Coagulation ☐ Severe anaemia (below 80 g/L) ☐ Gastrointestinal haemorrhage ☐ Encephalitis ☐ Pregnancy-related complications ☐ Hyperglycaemia ☐ Hypoglycaemia ☐ Rhabdomyolysis ☐ Metabolic acidosis ☐ Secondary infection ☐ Macrophage activation syndrome ☐ **None** ☐ Unknown ☐ **Other (please specify):**
C-19 Outcome	☐ 1. Not hospitalized, no limitations on activities ☐ 2. Not hospitalized, limitation on activities ☐ 3. Hospitalized, not requiring supplemental oxygen ☐ 4. Hospitalized, requiring supplemental oxygen ☐ 5. Hospitalized, on non-invasive ventilation or high flow oxygen devices ☐ 6. Hospitalized, on invasive mechanical ventilation or ECMO ☐ 7. Death ☐ 8. Unknown
If C-19 outcome = '7. Death' Date of death	
If C-19 outcome = '7. Death' Cause of death	☐ COVID-19 / presumed COVID-19 / complication of COVID-19 ☐ other (please specify):

Learning	
Would you like to share any lessons or other aspects from this case? Please include as much information as desired, this will greatly help patients and colleagues.	

UKIVas COVID-19 Case Report Form

Additional information – complete if possible, but please note can be completed at later date

Patient information (part 2)	
Please select any additional databases this patient's details have been shared with	☐ Irish RKD ☐ UKIVAS ☐ EULAR Global COVID-RHEUM ☐ SCAR19 (Scottish COVID19 Autoimmune Registry) ☐ Other (please specify):
Ethnicity (please select one)	☐ White British W1 ☐ White Irish W2 ☐ Any Other White Background W9 ☐ Indian A1 ☐ Pakistani A2 ☐ Bangladeshi A3 ☐ Any Other Asian Background A9 ☐ Caribbean B1 ☐ African B2 ☐ Any Other Black Background B9 ☐ White and Black Caribbean M1 ☐ White and Black African M2 ☐ White and Asian M3 ☐ Any Other Mixed Background M9 ☐ Chinese O1 ☐ Any Other Ethnic Group O9 ☐ Not Stated NS
Country of residence	
Employment status (at C-19 diagnosis, please select one)	☐ unknown ☐ not working ☐ disability benefit ☐ employed full-time ☐ employed part-time ☐ retired ☐ student ☐ home-maker ☐ n/a – pre-school age
Education (please select one)	☐ Primary School ☐ High school ☐ University ☐ Unknown or Prefer not to answer
Smoking history (please select one)	☐ Current Smoker ☐ Former Smoker ☐ Never Smoked ☐ Unknown smoking status
E-cigarette or Vape use (please select one)	☐ Yes ☐ No ☐ Unknown

Patient information (part 2) - Vasculitis diagnosis	
ANCA serology (at time of vasculitis diagnosis)	☐ PR3 ☐ MPO ☐ PR3 and MPO ☐ ELISA negative ☐ No ELISA performed ☐ Other (free text):
Biopsy performed	☐ Yes ☐ No ☐ Unknown
Histologically confirmed diagnosis	☐ Yes ☐ No ☐ Unknown

Vasculitis / Disease status at time of C-19 diagnosis	
Urinalysis Done	☐ Yes ☐ No ☐ Unknown
If Urinalysis = Yes: Urinalysis Protein	☐ Negative ☐ +1 ☐ +2 ☐ > = +3
If Urinalysis = Yes: Urinalysis Blood	☐ Negative ☐ +1 ☐ +2 ☐ > = +3
End-stage Kidney Disease (ESKD) prior to C-19 diagnosis?	☐ Yes ☐ No ☐ Unknown
If ESKD = Yes Type of Renal Replacement Therapy (RRT)	☐ Functioning renal transplant ☐ Haemodialysis ☐ Peritoneal Dialysis ☐ Sustained CKD V
If ESKD = Yes Date of onset of ESKD	
Last eGFR prior to C-19 diagnosis	Please enter NA if the patient was dialysis dependent at that time
Weight (kg)	
Height (m)	
Do you think vasculitis is relapsing in this encounter?	☐ High probability ☐ Possibly ☐ No ☐ Unknown
Adjudicated probability of relapse? (completed retrospectively by senior clinician)	☐ Definite ☐ High Probability ☐ Possibly ☐ No

UKIVas COVID-19 Case Report Form

Birmingham Vasculitis Activity Score (at time of C-19 diagnosis) only complete if training undertaken

Tick an item **only** if attributable to active vasculitis. If there are no -system.

	None	Active disease
1. General	☐	
Myalgia		☐
Arthralgia / arthritis		☐
		☐
		☐
2. Cutaneous	☐	
Infarct		☐
Purpura		☐
Ulcer		☐
♦**Gangrene**		☐
Other skin vasculitis		☐
3. Mucous membranes / eyes	☐	
Mouth ulcers		☐
Genital ulcers		☐
Adnexal inflammation		☐
Significant proptosis		☐
Scleritis / Episcleritis		☐
Conjunctivitis / Blepharitis / Keratitis		☐
Blurred vision		☐
Sudden visual loss		☐
Uveitis		☐
♦**Retinal changes (vasculitis / thrombosis / exudate / haemorrhage)**		☐
4. ENT	☐	
Bloody nasal discharge / crusts / ulcers / granulomata		☐
Paranasal sinus involvement		☐
Subglottic stenosis		☐
Conductive hearing loss		☐
♦**Sensorineural hearing loss**		☐
5. Chest	☐	
Wheeze		☐
Nodules or cavities		☐
Pleural effusion / pleurisy		☐
Infiltrate		☐
Endobronchial involvement		☐
♦**Massive haemoptysis / alveolar haemorrhage**		☐
♦**Respiratory failure**		☐

	None	Active disease
6. Cardiovascular	☐	
Loss of pulses		☐
Valvular heart disease		☐
Pericarditis		☐
♦**Ischaemic cardiac pain**		☐
♦**Cardiomyopathy**		☐
♦**Congestive cardiac failure**		☐
7. Abdominal	☐	
Peritonitis		☐
Bloody diarrhoea		☐
♦**Ischaemic abdominal pain**		☐
8. Renal	☐	
Hypertension		☐
Proteinuria >1+		☐
		☐
Creatinine 125-249µ/L(1.41-2.82mg/dl)*		☐
Creatinine 250-499 µ/L(2.83-5.64mg/dl)*		☐
		☐
♦**Rise in serum creatinine >30% or fall in creatinine clearance >25%**		☐
***Can only be scored on the first assessment**		
9. Nervous system	☐	
Headache		☐
Meningitis		☐
Organic confusion		☐
Seizures (not hypertensive)		☐
♦**Cerebrovascular accident**		☐
♦**Spinal cord lesion**		☐
♦**Cranial nerve palsy**		☐
Sensory peripheral neuropathy		☐
♦**Mononeuritis multiplex**		☐
10. Other	☐	
a.		☐
b.		☐
c.		☐
d.		☐
BVAS Score not necessary to add up for COVID-19 data collection, will be done centrally		

♦ **Major items highlighted**

UKIVas COVID-19 Case Report Form

Vasculitis Damage Index (at time of C-19 diagnosis) only complete if training undertaken

This is for recording organ damage that has occurred in patients *since the onset of vasculitis*. Patients often have co-morbidity before they develop vasculitis, **which must not be scored.** Record features of active disease using the Birmingham Vasculitis Activity Score (BVAS). A new patient should ***usually have a VDI score of zero***, unless: (a) they have had vasculitis for more than three months of onset of disease **and** (b) the damage has developed or become worse since the onset of vasculitis

1. Musculoskeletal	**No**	**Yes**
None	☐	
Significant muscle atrophy or weakness		☐
Deforming/erosive arthritis		☐
Osteoporosis/vertebral collapse		☐
Avascular necrosis		☐
Osteomyelitis		☐
2. Skin/Mucous membranes		
None	☐	
Alopecia		☐
Cutaneous ulcers		☐
Mouth ulcers		☐
3. Ocular		
None	☐	
Cataract		☐
Retinal change		☐
Optic atrophy		☐
Visual impairment/diplopia		☐
Blindness in one eye		☐
Blindness in second eye		☐
Orbital wall destruction		☐
4. ENT		
None	☐	
Hearing loss		☐
Nasal blockage/chronic discharge/crusting		☐
Nasal bridge collapse/septal perforation		☐
Chronic sinusitis/radiological damage		☐
Subglottic stenosis (no surgery)		☐
Subglottic stenosis (with surgery)		☐
5. Pulmonary		
None	☐	
Pulmonary hypertension		☐
Pulmonary fibrosis		☐
Pulmonary infarction		☐
Pleural fibrosis		☐
Chronic asthma		☐
Chronic breathlessness		☐
Impaired lung function		☐
6. Cardiovascular		
None	☐	
Angina/angioplasty		☐
Myocardial infarction		☐
Subsequent myocardial infarction		☐
Cardiomyopathy		☐
Valvular disease		☐
[illegible]		☐
[illegible]		☐

7. Peripheral vascular disease	**No**	**Yes**
None	☐	
Absent pulses in one limb		☐
2nd episode of absent pulses in one limb		☐
Major vessel stenosis		☐
Claudication >3 mths		☐
Minor tissue loss		☐
Major tissue loss		☐
Subsequent major tissue loss		☐
Complicated venous thrombosis		☐
8. Gastrointestinal		
None	☐	
Gut infarction/resection		☐
Mesenteric insufficiency/pancreatitis		☐
Chronic peritonitis		☐
Oesophageal stricture/surgery		☐
9. Renal		
None	☐	
[illegible]		☐
[illegible]		☐
End stage renal disease		☐
10. Neuropsychiatric		
None	☐	
Cognitive impairment		☐
Major psychosis		☐
Seizures		☐
Cerebrovascular accident		☐
2nd cerebrovascular accident		☐
Cranial nerve lesion		☐
Peripheral neuropathy		☐
Transverse myelitis		☐
11. Other		
None	☐	
Gonadal failure		☐
Marrow failure		☐
Diabetes		☐
Chemical cystitis		☐
Malignancy		☐
Other		☐

Total VDI Score not necessary to add up for COVID-19 data collection, will be done centrally

COVID-19 – diagnosis	
Location at which C-19 diagnosis was made (please select one)	☐ 1, Home or standalone testing ☐ 2, Nursing home or assisted living facility ☐ 3, Outpatient facility ☐ 4, Emergency Department ☐ 5, Inpatient/Hospital ☐ 6, Other (free text): ☐ 7, Unknown
Method of C-19 testing (select the most objective option)	☐ 1, symptoms (presumptive) ☐ 2, PCR ☐ 3, antibody ☐ 4, metagenomic testing ☐ 5, CT scan ☐ 6, other (free text): ☐ 7, Laboratory assay, type unknown
If Method = PCR: Level of Sars-CoV-2 (COVID-19) PCR	
If antibody testing done: Sars-CoV-2 (COVID-19) IgM level Sars-CoV-2 (COVID-19) IgG level	
Infection Acquisition	☐ High-risk travel to endemic area ☐ Contact of known or suspected person ☐ Attendance to a healthcare facility/ward where C-19 infections are managed ☐ None of the above (community acquired) ☐ Unknown
Presumed infection acquisition date (if known)	

COVID-19 – additional clinical features / investigations			
Body temperature (highest recorded, °C)		Splenomegaly or hepatomegaly	☐ Yes ☐ No ☐ Unknown
Note: All laboratory tests at first assessment / admission			
CRP (mg/L)		D-dimer (mg/L)	
Creatinine (μmol/L)		Troponin	Check units: ☐ ng/L ☐ ng/mL ☐ ng/dL
AST (U/L)		Creatinine kinase (U/L)	
ALT (U/L)		Ferritin (ug/L)	
Haemoglobin (g/dL)		Lactate dehydrogenase (U/L)	
Total White Cell Count x 10^9/L		Lactate (mg/dL)	
Neutrophil count x 10^9/L		Triglycerides (mmol/L)	
Lymphocyte count x 10^9/L		Fibrinogen (g/L)	
Platelet count x 10^9/L		Urine Protein Creatinine ratio (uPCR, mg/mmol)	
Prothrombin time (s)			
Anti-PR3 level (IU/ml)		Anti-MPO level (IU/mL)	
Radiological evidence of chest disease? (plain film or CT)	☐ Inflammation ☐ Ischaemia ☐ Infarction ☐ Haemorrhage ☐ Bony destruction ☐ Metastasis ☐ Mass / Tumour ☐ Abscess ☐ Polyp ☐ Ulceration ☐ Fibrosis	☐ Stenosis ☐ Calcification ☐ Effusion ☐ Embolism ☐ Nodules ☐ Consolidation ☐ Organomegaly ☐ Single granuloma ☐ Multiple granulomas ☐ None	
If consolidation present:	☐ unilateral ☐ bilateral		

COVID-19 – additional infection / outcome data	
Concomitant respiratory pathogens detected (select all that apply):	☐ Influenza A ☐ Influenza B ☐ NON-COVID-19 Coronavirus ☐ Respiratory syncytial virus (RSV) ☐ Adenovirus ☐ None ☐ Other (free text):
If Secondary Infection present: Type of Infection (in addition to those selected above)	☐ Bacteraemia ☐ Pneumonia ☐ Pyelonephritis ☐ Gastroenteritis ☐ Encephalitis ☐ Cellulitis ☐ Osteomyelitis ☐ Other (free text):
If Secondary Infection present: Organism(s) (if known)	
If AKI present: Dialysis required	☐ Yes ☐ No ☐ Unknown
If dialysis required: Date of dialysis start	
If dialysis required: Date of dialysis stop	or ☐ remains dialysis dependent
Date of symptom resolution (if known)	*Note: first date patient is asymptomatic, signifying recovery*
Have patient's symptoms resolved at time of initial report?	☐ 1 yes ☐ 2 no ☐ 3 unknown ☐ 4 Asymptomatic patient (just tested positive)
Date of hospital discharge (if known)	

Bibliography

Abani, O. et al. (2021) Tocilizumab in patients admitted to hospital with COVID-19 (RECOVERY): a randomised, controlled, open-label, platform trial. *The Lancet*. 397 (10285), 1637–1645.

Aerts, H. et al. (2021) Quality of Hospital Electronic Health Record (EHR) Data Based on the International Consortium for Health Outcomes Measurement (ICHOM) in Heart Failure: Pilot Data Quality Assessment Study. *JMIR medical informatics*. 9 (8), e27842.

Agarwal, N. et al. (2012) Are immunosuppressive medications associated with decreased responses to routine immunizations? A systematic review. *Vaccine*. 30 (8), 1413–1424.

Aiyegbusi, O. et al. (2021) ANCA-associated renal vasculitis is associated with rurality but not seasonality or deprivation in a complete national cohort study. *RMD open*. [Online] 7 (2), . [online]. Available from: http://dx.doi.org/10.1136/rmdopen-2020-001555.

Alexander, J. L. et al. (2022) COVID-19 vaccine-induced antibody responses in immunosuppressed patients with inflammatory bowel disease (VIP): a multicentre, prospective, case-control study. *The lancet. Gastroenterology & hepatology*. 7 (4), 342–352.

Almaani, S. et al. (2021) ANCA-Associated Vasculitis: An Update. *Journal of clinical medicine research*. [Online] 10 (7), . [online]. Available from: http://dx.doi.org/10.3390/jcm10071446.

Al-Shayeb, B. et al. (2020) Clades of huge phages from across Earth's ecosystems. *Nature*. 578 (7795), 425–431.

Amirthalingam, G. et al. (2021) Serological responses and vaccine effectiveness for extended COVID-19 vaccine schedules in England. *Nature communications*. 12 (1), 7217.

Andersen, P. K. & Perme, M. P. (2010) Pseudo-observations in survival analysis. *Statistical methods in medical research*. 19 (1), 71–99.

Anderson, G. et al. (1992) Wegener's granuloma. A series of 265 British cases seen between 1975 and 1985. A report by a sub-committee of the British Thoracic Society Research Committee. *The Quarterly journal of medicine*. 83 (302), 427–438.

Andrews, M. et al. (1990) Systemic vasculitis in the 1980s--is there an increasing incidence of Wegener's granulomatosis and microscopic polyarteritis? *Journal of the Royal College of Physicians of London*. 24 (4), 284–288.

Angyal, A. et al. (2022) T-cell and antibody responses to first BNT162b2 vaccine dose in previously infected and SARS-CoV-2-naive UK health-care workers: a multicentre prospective cohort study. *The Lancet. Microbe*. 3 (1), e21–e31.

Anon (2016) *REGULATION (EU) 2016/679 OF THE EUROPEAN PARLIAMENT AND OF THE COUNCIL of 27 April 2016 on the protection of natural persons with regard to the processing of personal data and on the free movement of such data, and repealing Directive 95/46/EC (General Data Protection Regulation)*. [online]. Available from: https://eur-lex.europa.eu/eli/reg/2016/679/oj (Accessed 22 September 2022).

Arts, D. G. T. et al. (2002) Defining and improving data quality in medical registries: a literature review, case study, and generic framework. *Journal of the American Medical Informatics Association: JAMIA*. 9 (6), 600–611.

Austin, P. C. & Steyerberg, E. W. (2017) Events per variable (EPV) and the relative performance of different strategies for estimating the out-of-sample validity of logistic regression models. *Statistical methods in medical research*. 26 (2), 796–808.

Ayorinde, A. et al. (2023) Health inequalities in infectious diseases: a systematic overview of reviews. *BMJ open*. 13 (4), e067429.

Bahlis, L. F. et al. (2021) Charlson Comorbidity Index and other predictors of in-hospital mortality among adults with community-acquired pneumonia. *Jornal brasileiro de pneumologia: publicacao oficial da Sociedade Brasileira de Pneumologia e Tisilogia*. 47 (1), e20200257.

Baneshi, M. R. et al. (2013) On the Use of Fractional Polynomial Models to Assess Preventive Aspect of Variables: An Example in Prevention of Mortality Following HIV Infection. *International journal of preventive medicine*. 4 (4), 414–419.

Barber, C. et al. (2013) Systematic review of validation studies of the use of administrative data to identify serious infections. *Arthritis care & research*. 65 (8), 1343–1357.

Barda, N. et al. (2021) BNT162b2 mRNA Covid-19 Vaccine in a Nationwide Mass Vaccination Setting. Reply. The New England journal of medicine 384 (20) p.1970.

Barnes, E. et al. (2023) SARS-CoV-2-specific immune responses and clinical outcomes after COVID-19 vaccination in patients with immune-suppressive disease. *Nature medicine*. 29 (7), 1760–1774.

Basu, N. et al. (2014) The characterisation and determinants of quality of life in ANCA associated vasculitis. *Annals of the rheumatic diseases*. 73 (1), 207–211.

Batini, C. et al. (2009) Methodologies for data quality assessment and improvement. *ACM Comput. Surv*. 41 (3), 1–52.

Berden, A. E. et al. (2010) Histopathologic classification of ANCA-associated glomerulonephritis. *Journal of the American Society of Nephrology: JASN*. 21 (10), 1628–1636.

Berti, A. et al. (2017) The Epidemiology of Antineutrophil Cytoplasmic Autoantibody-Associated Vasculitis in Olmsted County, Minnesota: A Twenty-Year US Population-Based Study. *Arthritis & rheumatology (Hoboken, N.J.)*. 69 (12), 2338–2350.

Blazek, K. et al. (2020) A practical guide to multiple imputation of missing data in nephrology. *Kidney international*. [Online] [online]. Available from: https://doi.org/10.1016/j.kint.2020.07.035.

Boekel, L. et al. (2021) Antibody development after COVID-19 vaccination in patients with autoimmune diseases in the Netherlands: a substudy of data from two prospective cohort studies. *The Lancet. Rheumatology*. 3 (11), e778–e788.

Boomsma, M. M. et al. (2000) Prediction of relapses in Wegener's granulomatosis by measurement of antineutrophil cytoplasmic antibody levels: a prospective study. *Arthritis and rheumatism*. 43 (9), 2025–2033.

Booth, A. et al. (2022) *Systematic Approaches to a Successful Literature Review*. SAGE Publications Ltd.

Boyarsky, B. J. et al. (2021) Antibody response to a single dose of SARS-CoV-2 mRNA vaccine in patients with rheumatic and musculoskeletal diseases. *Annals of the rheumatic diseases*. 80 (8), 1098–1099.

Cain, D. W. & Cidlowski, J. A. (2017) Immune regulation by glucocorticoids. *Nature reviews. Immunology*. 17 (4), 233–247.

Cannon, L. et al. (2023) Secondary immunodeficiencies and infectious considerations of biologic immunomodulatory therapies. *Annals of allergy, asthma & immunology: official publication of the American College of Allergy, Asthma, & Immunology*. 130 (6), 718–726.

Cao, Y. et al. (2011) DRB1*15 allele is a risk factor for PR3-ANCA disease in African Americans. *Journal of the American Society of Nephrology: JASN*. 22 (6), 1161–1167.

Carabelli, A. M. et al. (2023) SARS-CoV-2 variant biology: immune escape, transmission and fitness. *Nature reviews. Microbiology*. 21 (3), 162–177.

Carr, E. J., Wu, M., et al. (2021) Neutralising antibodies after COVID-19 vaccination in UK haemodialysis patients. The Lancet 398 (10305) p.1038–1041.

Carr, E. J., Kronbichler, A., et al. (2021) Review of Early Immune Response to SARS-CoV-2 Vaccination Among Patients With CKD. *Kidney international reports*. 6 (9), 2292–2304.

Cereda, D. et al. (2021) The early phase of the COVID-19 epidemic in Lombardy, Italy. *Epidemics*. 37100528.

Cervera, R. et al. (2003) Morbidity and mortality in systemic lupus erythematosus during a 10-year period: a comparison of early and late manifestations in a cohort of 1,000 patients. *Medicine*. 82 (5), 299–308.

Cevik, M. et al. (2021) SARS-CoV-2, SARS-CoV, and MERS-CoV viral load dynamics, duration of viral shedding, and infectiousness: a systematic review and meta-analysis. *The Lancet. Microbe*. 2 (1), e13–e22.

Charles, P. et al. (2018) Comparison of individually tailored versus fixed-schedule rituximab regimen to maintain ANCA-associated vasculitis remission: results of a multicentre, randomised controlled, phase III trial (MAINRITSAN2). *Annals of the rheumatic diseases*. 77 (8), 1143–1149.

Charles, P. et al. (2020) Long-Term Rituximab Use to Maintain Remission of Antineutrophil Cytoplasmic Antibody-Associated Vasculitis: A Randomized Trial. *Annals of internal medicine*. 173 (3), 179–187.

Charlson, M. E. et al. (1987) A new method of classifying prognostic comorbidity in longitudinal studies: development and validation. *Journal of chronic diseases*. 40 (5), 373–383.

Chiang, C.-J. et al. (2015) Quality assessment and improvement of nationwide cancer registration system in Taiwan: a review. *Japanese journal of clinical oncology*. 45 (3), 291–296.

Chowdhury, M. Z. I. & Turin, T. C. (2020) Variable selection strategies and its importance in clinical prediction modelling. *Family medicine and community health*. 8 (1), e000262.

Chowdhury, R. et al. (2017) Frailty and chronic kidney disease: A systematic review. *Archives of gerontology and geriatrics*. 68135–142.

Churg, J. & Strauss, L. (1951) Allergic granulomatosis, allergic angiitis, and periarteritis nodosa. *The American journal of pathology*. 27 (2), 277–301.

Chvatal-Medina, M. et al. (2021) Antibody Responses in COVID-19: A Review. *Frontiers in immunology*. 12633184.

Cigni, A. et al. (2008) Hormonal strategies for fertility preservation in patients receiving cyclophosphamide to treat glomerulonephritis: a nonrandomized trial and review of the literature. *American journal of kidney diseases: the official journal of the National Kidney Foundation*. 52 (5), 887–896.

Coggon, D. et al. (2003) *Epidemiology for the Uninitiated*. BMJ Books.

Cohen, P. et al. (2007) Churg-Strauss syndrome with poor-prognosis factors: A prospective multicenter trial comparing glucocorticoids and six or twelve cyclophosphamide pulses in forty-eight patients. *Arthritis and rheumatism*. 57 (4), 686–693.

Colevas, A. D. & Setser, A. (2004) The NCI Common Terminology Criteria for Adverse Events (CTCAE) v 3.0 is the new standard for oncology clinical trials. *Journal of clinical orthodontics: JCO*. 22 (14_suppl), 6098–6098.

Collins, G. S. et al. (2015) Transparent Reporting of a multivariable prediction model for Individual Prognosis or Diagnosis (TRIPOD): the TRIPOD statement. *Annals of internal medicine*. 162 (1), 55–63.

Comber, L. et al. (2021) Airborne transmission of SARS-CoV-2 via aerosols. *Reviews in medical virology*. 31 (3), e2184.

COVID-19 Forecasting Team (2022) Variation in the COVID-19 infection-fatality ratio by age, time, and geography during the pre-vaccine era: a systematic analysis. *The Lancet*. 399 (10334), 1469–1488.

Crowson, C. S. et al. (2011) The lifetime risk of adult-onset rheumatoid arthritis and other inflammatory autoimmune rheumatic diseases. *Arthritis and rheumatism*. 63 (3), 633–639.

Curtis, J. R. et al. (2006) Population-based assessment of adverse events associated with long-term glucocorticoid use. *Arthritis and rheumatism*. 55 (3), 420–426.

Damen, J. A. A. G. et al. (2016) Prediction models for cardiovascular disease risk in the general population: systematic review. *BMJ* . 353i2416.

DBpedia collaboration (2022) *DBpedia Ontology* [online]. Available from: https://dbpedia.org/ontology/ (Accessed 22 September 2022).

Demoulin, N. et al. (2021) Low rates of humoral response to BNT162b2 SARS-CoV-2 vaccination in patients with immune-mediated kidney diseases treated with rituximab. *Clinical kidney journal*. 14 (9), 2132–2133.

Dick, J. et al. (2018) C5a receptor 1 promotes autoimmunity, neutrophil dysfunction and injury in experimental anti-myeloperoxidase glomerulonephritis. *Kidney international*. 93 (3), 615–625.

Dickson, W. E. C. (1908) Polyarteritis acuta nodosa and periarteritis nodosa. *The Journal of pathology and bacteriology*. 12 (1), 31–57.

Didar, T. F. et al. (2015) Improved treatment of systemic blood infections using antibiotics with extracorporeal opsonin hemoadsorption. *Biomaterials*. 67382–392.

Dixon, W. G. et al. (2012) Immediate and delayed impact of oral glucocorticoid therapy on risk of serious infection in older patients with rheumatoid arthritis: a nested case-control analysis. *Annals of the rheumatic diseases*. 71 (7), 1128–1133.

Dixon, W. G. et al. (2011) The association between systemic glucocorticoid therapy and the risk of infection in patients with rheumatoid arthritis: systematic review and meta-analyses. *Arthritis research & therapy*. 13 (4), R139.

Docherty, A. B. et al. (2020) Features of 20 133 UK patients in hospital with covid-19 using the ISARIC WHO Clinical Characterisation Protocol: prospective observational cohort study. *BMJ* . 369m1985.

Donnelly, M. & McDonagh, M. (2019) Health Research, Consent and the GDPR Exemption. *European journal of health law*. 26 (2), 97–119.

Doran, M. F. et al. (2002) Frequency of infection in patients with rheumatoid arthritis compared with controls: a population-based study. *Arthritis and rheumatism*. 46 (9), 2287–2293.

Dumanski, S. M. & Ahmed, S. B. (2019) Fertility and reproductive care in chronic kidney disease. *Journal of nephrology*. 32 (1), 39–50.

Dusetzina, S. B. et al. (2014) *An Overview of Record Linkage Methods*. Agency for Healthcare Research and Quality (US).

Eder, L. et al. (2022) COVID-19 Hospitalizations, Intensive Care Unit Stays, Ventilation, and Death Among Patients With Immune-mediated Inflammatory Diseases Compared to Controls. *The Journal of rheumatology*. 49 (5), 523–530.

Eder, L. et al. (2023) Understanding COVID-19 Risk in Patients With Immune-Mediated Inflammatory Diseases: A Population-Based Analysis of SARS-CoV-2 Testing. *Arthritis care & research*. 75 (2), 317–325.

Eisenberg, R. A. et al. (2013) Rituximab-treated patients have a poor response to influenza vaccination. *Journal of clinical immunology*. 33 (2), 388–396.

von Elm, E. et al. (2007) The Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) statement: guidelines for reporting observational studies. *Annals of internal medicine*. 147 (8), 573–577.

Emmi, G. et al. (2023) Evidence-Based Guideline for the diagnosis and management of eosinophilic granulomatosis with polyangiitis. *Nature reviews. Rheumatology*. 19 (6), 378–393.

England, B. R. et al. (2021) Risk of COVID-19 in Rheumatoid Arthritis: A National Veterans Affairs Matched Cohort Study in At-Risk Individuals. *Arthritis & rheumatology (Hoboken, N.J.)*. 73 (12), 2179–2188.

Evans, J. M. & MacDonald, T. M. (1999) Record-linkage for pharmacovigilance in Scotland. *British journal of clinical pharmacology*. 47 (1), 105–110.

Exley, A. R. et al. (1997) Development and initial validation of the Vasculitis Damage Index for the standardized clinical assessment of damage in the systemic vasculitides. *Arthritis and rheumatism*. 40 (2), 371–380.

Fadahunsi, K. P. et al. (2019) Protocol for a systematic review and qualitative synthesis of information quality frameworks in eHealth. *BMJ open*. 9 (3), e024722.

Fahey, J. L. et al. (1954) Wegener's granulomatosis. *The American journal of medicine*. 17 (2), 168–179.

Falagas, M. E. et al. (2007) Infection-related morbidity and mortality in patients with connective tissue diseases: a systematic review. *Clinical rheumatology*. 26 (5), 663–670.

Falk, R. J. et al. (1990) Anti-neutrophil cytoplasmic autoantibodies induce neutrophils to degranulate and produce oxygen radicals in vitro. *Proceedings of the National Academy of Sciences of the United States of America*. 87 (11), 4115–4119.

Falk, R. J. et al. (2011) Granulomatosis with polyangiitis (Wegener's): an alternative name for Wegener's granulomatosis. *Arthritis and rheumatism*. 63 (4), 863–864.

Fauci, A. S. et al. (1971) Effect of cyclophosphamide upon the immune response in Wegener's granulomatosis. *The New England journal of medicine*. 285 (27), 1493–1496.

Fauci, A. S. et al. (1983) Wegener's granulomatosis: prospective clinical and therapeutic experience with 85 patients for 21 years. *Annals of internal medicine*. 98 (1), 76–85.

Faurschou, M. et al. (2012) Brief Report: long-term outcome of a randomized clinical trial comparing methotrexate to cyclophosphamide for remission induction in early systemic antineutrophil cytoplasmic antibody-associated vasculitis. *Arthritis and rheumatism*. 64 (10), 3472–3477.

Ferraro, A. J. et al. (2008) Levels of autoantibodies, unlike antibodies to all extrinsic antigen groups, fall following B cell depletion with Rituximab. *European journal of immunology*. 38 (1), 292–298.

Fine, M. J. et al. (1997) A prediction rule to identify low-risk patients with community-acquired pneumonia. *The New England journal of medicine*. 336 (4), 243–250.

Flossmann, O. et al. (2011) Long-term patient survival in ANCA-associated vasculitis. *Annals of the rheumatic diseases*. 70 (3), 488–494.

FOAF collaboration (2022) *FOAF Vocabulary Specification* [online]. Available from: http://xmlns.com/foaf/0.1/ (Accessed 22 September 2022).

Freedman, D. M. & Pfeiffer, R. M. (2017) Ascertainment Bias in Statin Use and Alzheimer Disease Incidence. JAMA neurology 74 (7) p.868.

Furer, V. et al. (2019) Incidence and prevalence of vaccine preventable infections in adult patients with autoimmune inflammatory rheumatic diseases (AIIRD): a systemic literature review informing the 2019 update of the EULAR recommendations for vaccination in adult patients with AIIRD. *RMD open*. 5 (2), e001041.

Furuta, S. et al. (2021) Effect of Reduced-Dose vs High-Dose Glucocorticoids Added to Rituximab on Remission Induction in ANCA-Associated Vasculitis: A Randomized Clinical Trial. *JAMA: the journal of the American Medical Association*. 325 (21), 2178–2187.

Gasparini, A. (2018) comorbidity: An R package for computing comorbidity scores. *Journal of open source software*. 3 (23), 648.

Gatenby, P. A. et al. (2009) Antineutrophil cytoplasmic antibody-associated vasculitides: could geographic patterns be explained by ambient ultraviolet radiation? *Arthritis and rheumatism*. 61 (10), 1417–1424.

Geetha, D. et al. (2018) Comparisons of Guidelines and Recommendations on Managing Antineutrophil Cytoplasmic Antibody-Associated Vasculitis. *Kidney international reports*. 3 (5), 1039–1049.

van Geloven, N. et al. (2022) Validation of prediction models in the presence of competing risks: a guide through modern methods. *BMJ* . 377e069249.

Gene Ontology Consortium (2022) *Gene Ontology Resource* [online]. Available from: http://geneontology.org/ (Accessed 22 September 2022).

Geneletti, S. et al. (2009) Adjusting for selection bias in retrospective, case-control studies. *Biostatistics* . 10 (1), 17–31.

George, M. D. et al. (2020) Risk for Serious Infection With Low-Dose Glucocorticoids in Patients With Rheumatoid Arthritis : A Cohort Study. *Annals of internal medicine*. 173 (11), 870–878.

George, M. D. et al. (2022) Risk of Serious Infection With Low-dose Glucocorticoids in Patients With Rheumatoid Arthritis: An Instrumental Variable Analysis. *Epidemiology* . 33 (1), 65–74.

Gianfrancesco, M. et al. (2020) Characteristics associated with hospitalisation for COVID-19 in people with rheumatic disease: data from the COVID-19 Global Rheumatology Alliance physician-reported registry. *Annals of the rheumatic diseases*. [Online] [online]. Available from: http://dx.doi.org/10.1136/annrheumdis-2020-217871.

Godeau, B. et al. (1995) Factors associated with Pneumocystis carinii pneumonia in Wegener's granulomatosis. *Annals of the rheumatic diseases*. 54 (12), 991–994.

Godman, G. C. & Churg, J. (1954) Wegener's granulomatosis: pathology and review of the literature. *A.M.A. archives of pathology*. 58 (6), 533–553.

Gómez-Puerta, J. A. et al. (2013) The association between silica exposure and development of ANCA-associated vasculitis: systematic review and meta-analysis. *Autoimmunity reviews*. 12 (12), 1129–1135.

Goulabchand, R. et al. (2022) Hospitalizations for infections in primary Sjögren's syndrome patients: a nationwide incidence study. *Annals of medicine*. 54 (1), 2672–2680.

Grau, R. G. (2015) Drug-Induced Vasculitis: New Insights and a Changing Lineup of Suspects. *Current rheumatology reports*. 17 (12), 71.

Grayson, P. C. et al. (2022) 2022 American College of Rheumatology/European Alliance of Associations for Rheumatology Classification Criteria for Eosinophilic Granulomatosis with Polyangiitis. *Annals of the rheumatic diseases*. 81 (3), 309–314.

Grijalva, C. G. et al. (2008) Changing patterns of medication use in patients with rheumatoid arthritis in a Medicaid population. *Rheumatology* . 47 (7), 1061–1064.

Grijalva, C. G. et al. (2011) Initiation of tumor necrosis factor-α antagonists and the risk of hospitalization for infection in patients with autoimmune diseases. *JAMA: the journal of the American Medical Association*. 306 (21), 2331–2339.

Guillevin, L. et al. (1997) A prospective, multicenter, randomized trial comparing steroids and pulse cyclophosphamide versus steroids and oral cyclophosphamide in the treatment of generalized Wegener's granulomatosis. *Arthritis and rheumatism*. 40 (12), 2187–2198.

Haas, E. J. et al. (2021) Impact and effectiveness of mRNA BNT162b2 vaccine against SARS-CoV-2 infections and COVID-19 cases, hospitalisations, and deaths following a nationwide vaccination campaign in Israel: an observational study using national surveillance data. *The Lancet*. 397 (10287), 1819–1829.

Harper, L. et al. (2012) Pulse versus daily oral cyclophosphamide for induction of remission in ANCA-associated vasculitis: long-term follow-up. *Annals of the rheumatic diseases*. 71 (6), 955–960.

Harrell, F. E. (2015) *Regression Modeling Strategies*. Springer International Publishing.

Harrell, F. E., Jr et al. (1996) Multivariable prognostic models: issues in developing models, evaluating assumptions and adequacy, and measuring and reducing errors. *Statistics in medicine*. 15 (4), 361–387.

Hayden, J. A. et al. (2013) Assessing bias in studies of prognostic factors. *Annals of internal medicine*. 158 (4), 280–286.

Head, M. L. et al. (2015) The extent and consequences of p-hacking in science. *PLoS biology*. 13 (3), e1002106.

Heijl, C. et al. (2017) Long-term patient survival in a Swedish population-based cohort of patients with ANCA-associated vasculitis. *RMD open*. 3 (1), e000435.

Heinze, G. et al. (2018) Variable selection - A review and recommendations for the practicing statistician. *Biometrical journal. Biometrische Zeitschrift*. 60 (3), 431–449.

Hellmich, B. et al. (2023) EULAR recommendations for the management of ANCA-associated vasculitis: 2022 update. *Annals of the rheumatic diseases*. [Online] [online]. Available from: http://dx.doi.org/10.1136/ard-2022-223764.

Hemingway, H. et al. (2013) Prognosis research strategy (PROGRESS) 1: a framework for researching clinical outcomes. *BMJ* . 346e5595.

Hemingway, H. et al. (2009) Ten steps towards improving prognosis research. *BMJ* . 339b4184.

Herlyn, K. et al. (2010) Patient-reported outcome assessment in vasculitis may provide important data and a unique perspective. *Arthritis care & research*. 62 (11), 1639–1645.

Hernán, M. A. & Robins, J. M. (2016) Using Big Data to Emulate a Target Trial When a Randomized Trial Is Not Available. *American journal of epidemiology*. 183 (8), 758–764.

Hespanhol, V. & Bárbara, C. (2020) Pneumonia mortality, comorbidities matter? *Pulmonology*. 26 (3), 123–129.

Hingorani, A. D. et al. (2013) Prognosis research strategy (PROGRESS) 4: stratified medicine research. *BMJ* . 346e5793.

Hippisley-Cox, J. et al. (2017) Development and validation of QRISK3 risk prediction algorithms to estimate future risk of cardiovascular disease: prospective cohort study. *BMJ* . 357j2099.

Hoffman, G. S. et al. (1992) Wegener granulomatosis: an analysis of 158 patients. *Annals of internal medicine*. 116 (6), 488–498.

Hollander, D. & Manning, R. T. (1967) The use of alkylating agents in the treatment of Wegener's granulomatosis. *Annals of internal medicine*. 67 (2), 393–398.

Hunder, G. G. et al. (1990) The American College of Rheumatology 1990 criteria for the classification of vasculitis. Introduction. *Arthritis and rheumatism*. 33 (8), 1065–1067.

Hwang, J. et al. (2016) Administrative data is as good as medical chart review for comorbidity ascertainment in patients with infections in Singapore. *Epidemiology and infection*. 144 (9), 1999–2005.

Iba, K. et al. (2021) Re-evaluation of the comparative effectiveness of bootstrap-based optimism correction methods in the development of multivariable clinical prediction models. *BMC medical research methodology*. 21 (1), 9.

Inada-Kim, M. et al. (2017) Defining and measuring suspicion of sepsis: an analysis of routine data. *BMJ open*. 7 (6), e014885.

Ishizu, A. et al. (2023) Expert perspectives on pathological findings in vasculitis. *Modern rheumatology / the Japan Rheumatism Association*. 33 (1), 1–11.

Jackson, C. B. et al. (2022) Mechanisms of SARS-CoV-2 entry into cells. *Nature reviews. Molecular cell biology*. 23 (1), 3–20.

Jayne, D. et al. (2003) A randomized trial of maintenance therapy for vasculitis associated with antineutrophil cytoplasmic autoantibodies. *The New England journal of medicine*. 349 (1), 36–44.

Jayne, D. (2009) The diagnosis of vasculitis. *Best practice & research. Clinical rheumatology*. 23 (3), 445–453.

Jayne, D. R. et al. (2000) Intravenous immunoglobulin for ANCA-associated systemic vasculitis with persistent disease activity. *QJM: monthly journal of the Association of Physicians*. 93 (7), 433–439.

Jayne, D. R. W. et al. (2021) Avacopan for the Treatment of ANCA-Associated Vasculitis. *The New England journal of medicine*. 384 (7), 599–609.

Jayne, D. R. W. et al. (2017) Randomized Trial of C5a Receptor Inhibitor Avacopan in ANCA-Associated Vasculitis. *Journal of the American Society of Nephrology: JASN*. 28 (9), 2756–2767.

Jayne, D. R. W. et al. (2007) Randomized trial of plasma exchange or high-dosage methylprednisolone as adjunctive therapy for severe renal vasculitis. *Journal of the American Society of Nephrology: JASN*. 18 (7), 2180–2188.

Jennette, J. C. et al. (2013) 2012 revised International Chapel Hill Consensus Conference Nomenclature of Vasculitides. *Arthritis and rheumatism*. 65 (1), 1–11.

Jennette, J. C. et al. (1994) Nomenclature of systemic vasculitides. Proposal of an international consensus conference. *Arthritis and rheumatism*. 37 (2), 187–192.

Johansson, M. W. et al. (1997) Myeloperoxidase mediates cell adhesion via the alpha M beta 2 integrin (Mac-1, CD11b/CD18). *Journal of cell science*. 110 (Pt 9)1133–1139.

Johnson, R. M. & Vinetz, J. M. (2020) Dexamethasone in the management of covid -19. *BMJ* . 370m2648.

Joint Formulary Committee (2022) *British National Formulary. [BNF online]. London: BMJ Group and Pharmaceutical Press;* [online]. Available from: https://bnf.nice.org.uk/treatment-summaries/glucocorticoid-therapy/ (Accessed 22 August 2022).

Jones, B. E. et al. (2017) Gene-Specific DNA Methylation Changes Predict Remission in Patients with ANCA-Associated Vasculitis. *Journal of the American Society of Nephrology: JASN*. 28 (4), 1175–1187.

Jones, R. B. et al. (2010) Rituximab versus cyclophosphamide in ANCA-associated renal vasculitis. *The New England journal of medicine*. 363 (3), 211–220.

Kain, R. et al. (2012) High prevalence of autoantibodies to hLAMP-2 in anti-neutrophil cytoplasmic antibody-associated vasculitis. *Journal of the American Society of Nephrology: JASN*. 23 (3), 556–566.

Kain, R. et al. (2008) Molecular mimicry in pauci-immune focal necrotizing glomerulonephritis. *Nature medicine*. 14 (10), 1088–1096.

Kant, S. et al. (2022) Antibody response to COVID-19 booster vaccine in rituximab-treated patients with anti-neutrophil cytoplasmic antibody-associated vasculitis. *Kidney international*. 101 (2), 414–415.

Kantidakis, G. et al. (2023) Statistical models versus machine learning for competing risks: development and validation of prognostic models. *BMC medical research methodology*. 23 (1), 51.

Karras, A. et al. (2017) Randomised controlled trial of prolonged treatment in the remission phase of ANCA-associated vasculitis. *Annals of the rheumatic diseases*. 76 (10), 1662–1668.

Kawasaki T. (1967) [Acute febrile mucocutaneous syndrome with lymphoid involvement with specific desquamation of the fingers and toes in children]. *Arerugi = [Allergy]*. 16 (3), 178–222.

Kessenbrock, K. et al. (2009) Netting neutrophils in autoimmune small-vessel vasculitis. *Nature medicine*. 15 (6), 623–625.

Keuning, B. E. et al. (2020) Mortality prediction models in the adult critically ill: A scoping review. *Acta anaesthesiologica Scandinavica*. 64 (4), 424–442.

Khan, A. et al. (2012) Risk factors for fractures and avascular osteonecrosis in type 1 Gaucher disease: a study from the International Collaborative Gaucher Group (ICGG) Gaucher Registry. *Journal of bone and mineral research: the official journal of the American Society for Bone and Mineral Research*. 27 (8), 1839–1848.

Khoury, D. S. et al. (2021) Neutralizing antibody levels are highly predictive of immune protection from symptomatic SARS-CoV-2 infection. *Nature medicine*. 27 (7), 1205–1211.

Kitching, A. R. et al. (2020) ANCA-associated vasculitis. *Nature reviews. Disease primers*. 6 (1), 71.

Klebanoff, S. J. (2005) Myeloperoxidase: friend and foe. *Journal of leukocyte biology*. 77 (5), 598–625.

Kronbichler, A. et al. (2023) Classification criteria for ANCA-associated vasculitis: one size does not fit all! Rheumatology 62 (3) p.993–995.

Kronbichler, A. et al. (2024) Diagnosis and management of ANCA-associated vasculitis. *The Lancet*. 403 (10427), 683–698.

Kronbichler, A. et al. (2015) Frequency, risk factors and prophylaxis of infection in ANCA-associated vasculitis. *European journal of clinical investigation*. 45 (3), 346–368.

Kronbichler, A. et al. (2018) Trimethoprim-sulfamethoxazole prophylaxis prevents severe/life-threatening infections following rituximab in antineutrophil cytoplasm antibody-associated vasculitis. *Annals of the rheumatic diseases*. 77 (10), 1440–1447.

Kroon, F. P. B. et al. (2022) Risk and prognosis of SARS-CoV-2 infection and vaccination against SARS-CoV-2 in rheumatic and musculoskeletal diseases: a systematic literature review to inform EULAR recommendations. *Annals of the rheumatic diseases*. 81 (3), 422–432.

Lafarge, A. et al. (2020) Predictive factors of severe infections in patients with systemic necrotizing vasculitides: data from 733 patients enrolled in five randomized controlled trials of the French Vasculitis Study Group. *Rheumatology* . 59 (9), 2250–2257.

Lai, Q. Y. et al. (2014) Predictors for mortality in patients with antineutrophil cytoplasmic autoantibody-associated vasculitis: A study of 398 Chinese patients. *The Journal of rheumatology*. 41 (9), 1849–1855.

Lamers, M. M. & Haagmans, B. L. (2022) SARS-CoV-2 pathogenesis. *Nature reviews. Microbiology*. 20 (5), 270–284.

Lau, D. et al. (2005) Myeloperoxidase mediates neutrophil activation by association with CD11b/CD18 integrins. *Proceedings of the National Academy of Sciences of the United States of America*. 102 (2), 431–436.

Leavitt, R. Y. et al. (1990) The American College of Rheumatology 1990 criteria for the classification of Wegener's granulomatosis. *Arthritis and rheumatism*. 33 (8), 1101–1107.

Lecluse, L. L. A. et al. (2009) National registries of systemic treatment for psoriasis and the European "Psonet" initiative. *Dermatology* . 218 (4), 347–356.

Li, J. et al. (2018) The frequency of ANCA-associated vasculitis in a national database of hospitalized patients in China. *Arthritis research & therapy*. 20 (1), 226.

Li, L. et al. (2019) Search strategies to identify observational studies in MEDLINE and Embase. *Cochrane database of systematic reviews* . 3 (3), MR000041.

Li, S. et al. (2020) High-dose but not low-dose corticosteroids potentially delay viral shedding of patients with COVID-19. *Clinical infectious diseases: an official publication of the Infectious Diseases Society of America*. [Online] [online]. Available from: http://dx.doi.org/10.1093/cid/ciaa829.

Lim, W. S. et al. (2003) Defining community acquired pneumonia severity on presentation to hospital: an international derivation and validation study. *Thorax*. 58 (5), 377–382.

Little, M. A. et al. (2012) Anti-proteinase 3 anti-neutrophil cytoplasm autoantibodies recapitulate systemic vasculitis in mice with a humanized immune system. *PloS one*. 7 (1), e28626.

Little, M. A. et al. (2010) Early mortality in systemic vasculitis: relative contribution of adverse events and active vasculitis. *Annals of the rheumatic diseases*. 69 (6), 1036–1043.

Lood, C. & Hughes, G. C. (2017) Neutrophil extracellular traps as a potential source of autoantigen in cocaine-associated autoimmunity. *Rheumatology* . 56 (4), 638–643.

Luna, C. M. et al. (2016) The Impact of Age and Comorbidities on the Mortality of Patients of Different Age Groups Admitted with Community-acquired Pneumonia. *Annals of the American Thoracic Society*. 13 (9), 1519–1526.

MacKenna, B. et al. (2022) Risk of severe COVID-19 outcomes associated with immune-mediated inflammatory diseases and immune-modifying therapies: a nationwide cohort study in the OpenSAFELY platform. *The Lancet. Rheumatology*. 4 (7), e490–e506.

Marty, P. K. et al. (2022) Antigen Specific Humoral and Cellular Immunity Following SARS-CoV-2 Vaccination in ANCA-Associated Vasculitis Patients Receiving B-Cell Depleting Therapy. *Frontiers in immunology*. 13834981.

Masi, A. T. et al. (1990) The American College of Rheumatology 1990 criteria for the classification of Churg-Strauss syndrome (allergic granulomatosis and angiitis). *Arthritis and rheumatism*. 33 (8), 1094–1100.

Maskell, D. J. & Wood, J. L. N. (2020) "*Biology of pathogenic microorganisms*," in Oxford University Press. p.

Matteson, E. L. et al. (1996) Long-term survival of patients with Wegener's granulomatosis from the American College of Rheumatology Wegener's Granulomatosis Classification Criteria Cohort. *The American journal of medicine*. 101 (2), 129–134.

McClure, M. E. et al. (2021) Long-term maintenance rituximab for ANCA-associated vasculitis: Relapse and infection prediction models. *Rheumatology*. 60 (3), 1491–1501.

McGlinn, K. et al. (2022) FAIRVASC: A semantic web approach to rare disease registry integration. *Computers in biology and medicine*. 105313.

McGregor, J. G. et al. (2015) Adverse events and infectious burden, microbes and temporal outline from immunosuppressive therapy in antineutrophil cytoplasmic antibody-associated vasculitis with native renal function. *Nephrology, dialysis, transplantation: official publication of the European Dialysis and Transplant Association - European Renal Association*. [Online] 30 (Supplement.1), . [online]. Available from: http://ndt.oxfordjournals.org/.

McGregor, J. G. et al. (2012) Glucocorticoids and relapse and infection rates in anti-neutrophil cytoplasmic antibody disease. *Clinical journal of the American Society of Nephrology: CJASN*. 7 (2), 240–247.

Merino-Vico, A. et al. (2021) B Lineage Cells in ANCA-Associated Vasculitis. *International journal of molecular sciences*. [Online] 23 (1), . [online]. Available from: http://dx.doi.org/10.3390/ijms23010387.

Merkel, P. A. et al. (2005) Brief communication: high incidence of venous thrombotic events among patients with Wegener granulomatosis: the Wegener's Clinical Occurrence of Thrombosis (WeCLOT) Study. *Annals of internal medicine*. 142 (8), 620–626.

Meyerholz, D. K. et al. (2016) Dipeptidyl Peptidase 4 Distribution in the Human Respiratory Tract: Implications for the Middle East Respiratory Syndrome. *The American journal of pathology*. 186 (1), 78–86.

Miloslavsky, E. M. et al. (2013) Clinical outcomes of remission induction therapy for severe antineutrophil cytoplasmic antibody-associated vasculitis. *Arthritis and rheumatism*. 65 (9), 2441–2449.

Mohammad, A. J. et al. (2017) Severe infection in antineutrophil cytoplasmic antibody-associated vasculitis. *The Journal of rheumatology*. 44 (10), 1468–1475.

Mok, C. C. et al. (1998) Risk factors for ovarian failure in patients with systemic lupus erythematosus receiving cyclophosphamide therapy. *Arthritis and rheumatism*. 41 (5), 831–837.

Moons, K. G. M. et al. (2019) PROBAST: A Tool to Assess Risk of Bias and Applicability of Prediction Model Studies: Explanation and Elaboration. *Annals of internal medicine*. 170 (1), W1–W33.

Morgan, M. D. et al. (2016) Association of Low B Cell Count and IgG Levels with Infection, and Poor Vaccine Response with All-Cause Mortality in an Immunosuppressed Vasculitis Population. *Arthritis care and research: the official journal of the Arthritis Health Professions Association*. 68 (6), 853–860.

Movsisyan, M. et al. (2022) Kinetics of anti-nucleocapsid IgG response in COVID-19 immunocompetent convalescent patients. *Scientific reports*. 12 (1), 12403.

MRC Regulatory Support Centre (2023) *MRC Regulatory Support Centre Courses* [online]. Available from: https://bygsystems.net/mrcrsc-lms/course/index.php?categoryid=14 (Accessed 26 June 2023).

Mukhtyar, C. et al. (2009) Modification and validation of the Birmingham Vasculitis Activity Score (version 3). *Annals of the rheumatic diseases*. 68 (12), 1827–1832.

Musen, M. A. et al. (2012) The National Center for Biomedical Ontology. *Journal of the American Medical Informatics Association: JAMIA*. 19 (2), 190–195.

Mv, P. et al. (2023) New-onset systemic vasculitis following SARS-CoV-2 infection and vaccination: the trigger, phenotype, and outcome. *Clinical rheumatology*. 42 (10), 2761–2775.

Naing, N. N. (2000) Easy way to learn standardization : direct and indirect methods. *The Malaysian journal of medical sciences: MJMS*. 7 (1), 10–15.

National Institutes of Health. National Cancer Institute. (2017) *Common terminology criteria for adverse events (CTCAE) v5.0*. [online]. Available from: https://ctep.cancer.gov/protocoldevelopment/electronic_applications/docs/CTCAE_v5_Quick_Reference_8.5x11.pdf.

NHS National Services Scotland (2017) *Scottish clinical coding standards ICD-10 Consolidation*.

Nilsen, A. T. et al. (2020) Increasing incidence and prevalence of ANCA-associated vasculitis in Northern Norway. *Rheumatology* . 59 (9), 2316–2324.

Noma, H. et al. (2021) Confidence intervals of prediction accuracy measures for multivariable prediction models based on the bootstrap-based optimism correction methods. *Statistics in medicine*. 40 (26), 5691–5701.

Noordzij, M. et al. (2013) When do we need competing risks methods for survival analysis in nephrology? *Nephrology, dialysis, transplantation: official publication of the European Dialysis and Transplant Association - European Renal Association*. 28 (11), 2670–2677.

Nyberg, T. et al. (2022) Comparative analysis of the risks of hospitalisation and death associated with SARS-CoV-2 omicron (B.1.1.529) and delta (B.1.617.2) variants in England: a cohort study. *The Lancet*. 399 (10332), 1303–1312.

O'Reilly, G. M. et al. (2016) Classifying, measuring and improving the quality of data in trauma registries: A review of the literature. *Injury*. 47 (3), 559–567.

Osler, W. (1914) THE VISCERAL LESIONS OF PURPURA AND ALLIED CONDITIONS. *British medical journal*. 1 (2775), 517–525.

Pearce, F. A., Hubbard, R. B., et al. (2018) Can granulomatosis with polyangiitis be diagnosed earlier in primary care? A case-control study. *QJM: monthly journal of the Association of Physicians*. 111 (1), 39–45.

Pearce, F. A., Craven, A., et al. (2017) Global ethnic and geographic differences in the clinical presentations of anti-neutrophil cytoplasm antibody-associated vasculitis. *Rheumatology* . 56 (11), 1962–1969.

Pearce, F. A. et al. (2016) Incidence of ANCA-associated vasculitis in a UK mixed ethnicity population. *Rheumatology* . 55 (9), 1656–1663.

Pearce, F. A., McGrath, C., et al. (2018) Outcomes and compliance with standards of care in anti-neutrophil cytoplasmic antibody-associated vasculitis-insights from a large multiregion audit. *Rheumatology Advances in Practice*. 2 (2), 1–7.

Pearce, F. A., Grainge, M. J., et al. (2017) The incidence, prevalence and mortality of granulomatosis with polyangiitis in the UK Clinical Practice Research Datalink. *Rheumatology* . 56 (4), 589–596.

Pendergraft, W. F., 3rd et al. (2004) Autoimmunity is triggered by cPR-3(105-201), a protein complementary to human autoantigen proteinase-3. *Nature medicine*. 10 (1), 72–79.

Persaud, C. et al. (2021) Highest Recorded Serum Creatinine. *Case reports in nephrology and urology*. [Online] 2021. [online]. Available from: https://www.hindawi.com/journals/crin/2021/6048919/ (Accessed 6 December 2023).

Phung, M. T. et al. (2019) Prognostic models for breast cancer: a systematic review. *BMC cancer*. 19 (1), 230.

Polack, F. P. et al. (2020) Safety and Efficacy of the BNT162b2 mRNA Covid-19 Vaccine. *The New England journal of medicine*. 383 (27), 2603–2615.

Premkumar, L. et al. (2020) The receptor binding domain of the viral spike protein is an immunodominant and highly specific target of antibodies in SARS-CoV-2 patients. *Science immunology*. [Online] 5 (48), . [online]. Available from: http://dx.doi.org/10.1126/sciimmunol.abc8413.

Prendecki, M. et al. (2021) Humoral and T-cell responses to SARS-CoV-2 vaccination in patients receiving immunosuppression. *Annals of the rheumatic diseases*. 80 (10), 1322–1329.

Pritchard, E. et al. (2021) Impact of vaccination on new SARS-CoV-2 infections in the United Kingdom. *Nature medicine*. 27 (8), 1370–1378.

Public Health Scotland (2024) *CHI Linkage and Indexing (CHILI)* [online]. Available from: https://publichealthscotland.scot/services/chi-linkage-and-indexing-chili/about-the-chi-linkage-and-indexing-team-chili/community-health-index-chi/ (Accessed 2024).

Public Health Scotland (2022) *National Data Catalogue: Prescribing Information System (PIS)* [online]. Available from: https://www.ndc.scot.nhs.uk/National-Datasets/data.asp?SubID=9 (Accessed 22 August 2022).

Public Health Scotland (2023) *What is clinical coding?* [online]. Available from: https://publichealthscotland.scot/our-areas-of-work/isd-services-development/terminology-services-and-clinical-coding/what-is-clinical-coding/clinical-classifications/assigning-codes/ (Accessed 26 June 2023).

Public Health Scotland: Data and Intelligence (2014) *Scottish Clinical Coding Standards* [online]. Available from: https://www.isdscotland.org/Products-and-Services/Terminology-Services/Clinical-Coding-Guidelines/ (Accessed 27 June 2023).

Puéchal, X. et al. (2017) Adding Azathioprine to Remission-Induction Glucocorticoids for Eosinophilic Granulomatosis With Polyangiitis (Churg-Strauss), Microscopic Polyangiitis, or Polyarteritis Nodosa Without Poor Prognosis Factors: A Randomized, Controlled Trial. *Arthritis & rheumatology (Hoboken, N.J.)*. 69 (11), 2175–2186.

Quan, H. et al. (2005) Coding algorithms for defining comorbidities in ICD-9-CM and ICD-10 administrative data. *Medical care*. 43 (11), 1130–1139.

R Core Team (2022) *R: A Language and Environment for Statistical Computing*. [online]. Available from: https://www.R-project.org/.

Ramspek, C. L. et al. (2021) Prediction or causality? A scoping review of their conflation within current observational research. *European journal of epidemiology*. 36 (9), 889–898.

Rathmann, J. et al. (2021) Incidence and predictors of severe infections in ANCA-associated vasculitis: A population-based cohort study. *Rheumatology*. 60 (6), 2745–2754.

RECOVERY Collaborative Group et al. (2020) Dexamethasone in Hospitalized Patients with Covid-19 — Preliminary Report. *The New England journal of medicine*. [Online] [online]. Available from: http://dx.doi.org/10.1056/NEJMoa2021436.

Redjoul, R. et al. (2021) Antibody response after second BNT162b2 dose in allogeneic HSCT recipients. *The Lancet*. 398 (10297), 298–299.

Reinhold-Keller, E. et al. (2000) An interdisciplinary approach to the care of patients with Wegener's granulomatosis: Long-term outcome in 155 patients. *Arthritis and rheumatism*. 43 (5), 1021–1032.

Rhee, C. et al. (2017) Incidence and Trends of Sepsis in US Hospitals Using Clinical vs Claims Data, 2009-2014. *JAMA: the journal of the American Medical Association*. 318 (13), 1241–1249.

Riley, R. D. et al. (2020) Calculating the sample size required for developing a clinical prediction model. *BMJ* . 368m441.

Riley, R. D. et al. (2019) Minimum sample size for developing a multivariable prediction model: PART II - binary and time-to-event outcomes. *Statistics in medicine*. 38 (7), 1276–1296.

Riley, R. D. et al. (2021) Penalization and shrinkage methods produced unreliable clinical prediction models especially when sample size was small. *Journal of clinical epidemiology*. 13288–96.

Riley, R. D. et al. (2013) Prognosis Research Strategy (PROGRESS) 2: prognostic factor research. *PLoS medicine*. 10 (2), e1001380.

Riley, R. D. et al. (2009) Prognostic markers in cancer: the evolution of evidence from single studies to meta-analysis, and beyond. *British journal of cancer*. 100 (8), 1219–1229.

Roberts, D. M. et al. (2015) Rituximab-associated hypogammaglobulinemia: incidence, predictors and outcomes in patients with multi-system autoimmune disease. *Journal of autoimmunity*. 5760–65.

Robertson, H. D. & Polk, H. C., Jr (1974) The mechanism of infection in patients with diabetes mellitus: a review of leukocyte malfunction. *Surgery*. 75 (1), 123–128.

Robson, J. et al. (2015) Damage in the anca-associated vasculitides: long-term data from the European vasculitis study group (EUVAS) therapeutic trials. *Annals of the rheumatic diseases*. 74 (1), 177–184.

Robson, J. C. et al. (2022) 2022 American College of Rheumatology/European Alliance of Associations for Rheumatology classification criteria for granulomatosis with polyangiitis. *Annals of the rheumatic diseases*. 81 (3), 315–320.

Roche (2023a) *Elecsys® Anti-SARS-CoV-2 N* [online]. Available from: https://diagnostics.roche.com/gb/en/products/params/elecsys-anti-sars-cov-2.html (Accessed 20 December 2023).

Roche (2023b) *Elecsys® Anti-SARS-CoV-2 S* [online]. Available from: https://diagnostics.roche.com/global/en/products/params/elecsys-anti-sars-cov-2-s.html (Accessed 20 December 2023).

Ross, R. K. et al. (2020) When Is a Complete-Case Approach to Missing Data Valid? The Importance of Effect-Measure Modification. *American journal of epidemiology*. 189 (12), 1583–1589.

Royston, P. & Altman, D. G. (2013) External validation of a Cox prognostic model: principles and methods. *BMC medical research methodology*. 1333.

Royston, P. & Altman, D. G. (1994) Regression Using Fractional Polynomials of Continuous Covariates: Parsimonious Parametric Modelling. *Journal of the Royal Statistical Society. Series C, Applied statistics*. 43 (3), 429–467.

Rudd, K. E. et al. (2020) Global, regional, and national sepsis incidence and mortality, 1990-2017: analysis for the Global Burden of Disease Study. *The Lancet*. 395 (10219), 200–211.

Rutherford, M. A. et al. (2021) Risk Factors for Severe Outcomes in Patients With Systemic Vasculitis and COVID-19: A Binational, Registry-Based Cohort Study. *Arthritis and Rheumatology*. 73 (9), 1713–1719.

Rutter, M. et al. (2022) COVID-19 infection, admission and death among people with rare autoimmune rheumatic disease in England: results from the RECORDER project. *Rheumatology* . 61 (8), 3161–3171.

Sakai, R. et al. (2019) Risk of opportunistic infections in patients with antineutrophil cytoplasmic antibody-associated vasculitis, using a Japanese health insurance database. *International journal of rheumatic diseases*. 22 (11), 1978–1984.

Sakellariou, G. et al. (2022) Influence of initial glucocorticoid co-medication on mortality and hospitalization in early inflammatory arthritis: an investigation by record linkage of clinical and administrative databases. *Arthritis research & therapy*. 24 (1), 144.

Sarica, S. (2018) *A study of comorbidities in ANCA-associated vasculitis*. PhD thesis. University of Aberdeen.

Sarica, S. et al. (2018) Tracking the risk of infections in ANCA-associated vasculitis: Results from a scottish matched-cohort study. *Arthritis and Rheumatology*. 70 (Supplement 9), 3055–3056.

Sarica, S. H. et al. (2020) Characterizing infection in anti-neutrophil cytoplasmic antibody-associated vasculitis: Results from a longitudinal, matched-cohort data linkage study. *Rheumatology*. 59 (10), 3014–3022.

Sarica, Shifa H. et al. (2020) Multimorbidity in Anti-Neutrophil Cytoplasmic Antibody-associated vasculitis: results from a longitudinal, multi-centre data-linkage study. *Arthritis & Rheumatology*. [Online] [online]. Available from: https://onlinelibrary.wiley.com/doi/10.1002/art.41557.

Sattui, S. E. et al. (2021) Outcomes of COVID-19 in patients with primary systemic vasculitis or polymyalgia rheumatica from the COVID-19 Global Rheumatology Alliance physician registry: a retrospective cohort study. *The Lancet Rheumatology*. 3 (12), e855–e864.

Sauerbrei, W. et al. (2020) State of the art in selection of variables and functional forms in multivariable analysis-outstanding issues. *Diagnostic and prognostic research*. 43.

Schenfeld, J. et al. (2017) Dose relationship between oral glucocorticoids and tumor necrosis factor inhibitors and the risk of hospitalized infectious events

among patients with rheumatoid arthritis. *Rheumatology international*. 37 (7), 1075–1082.

Schlieben, D. J. et al. (2005) Pulmonary-renal syndrome in a newborn with placental transmission of ANCAs. *American journal of kidney diseases: the official journal of the National Kidney Foundation*. 45 (4), 758–761.

Schreiber, A. et al. (2003) Membrane expression of proteinase 3 is genetically determined. *Journal of the American Society of Nephrology: JASN*. 14 (1), 68–75.

Scott, J. et al. (2020) Environmental risk factors associated with ANCA associated vasculitis: A systematic mapping review. *Autoimmunity reviews*. 102660.

Scottish Government (2020) *Introducing the Scottish Index of Multiple Deprivation 2020*. [online]. Available from: https://www.gov.scot/binaries/content/documents/govscot/publications/statistics/2020/01/scottish-index-multiple-deprivation-2020/documents/scottish-index-multiple-deprivation-2020/scottish-index-multiple-deprivation-2020/govscot%3Adocument/scottish-index-multiple-deprivation-2020.pdf.

Scottish Public Health Observatory (2022) *ISD Linked Database* [online]. Available from: https://www.scotpho.org.uk/publications/overview-of-key-data-sources/scottish-national-data-schemes/isd-linked-database (Accessed 17 August 2022).

Sebastiani, M. et al. (2020) Epidemiology and management of interstitial lung disease in ANCA-associated vasculitis. *Clinical and experimental rheumatology*. 38 Suppl 124 (2), 221–231.

Sedrakyan, A. et al. (2014) Comparative effectiveness of ceramic-on-ceramic implants in stemmed hip replacement: a multinational study of six national and regional registries. *The Journal of bone and joint surgery. American volume*. 96 Suppl 134–41.

Seeliger, B. et al. (2017) Are the 1990 American College of Rheumatology vasculitis classification criteria still valid? *Rheumatology* . 56 (7), 1154–1161.

Shapiro, N. I. et al. (2003) Mortality in Emergency Department Sepsis (MEDS) score: a prospectively derived and validated clinical prediction rule. *Critical care medicine*. 31 (3), 670–675.

Shaw, T. et al. (2003) B cell therapy for rheumatoid arthritis: the rituximab (anti-CD20) experience. *Annals of the rheumatic diseases*. 62 Suppl 2 (Suppl 2), ii55-9.

Shingu, M. et al. (1992) Activation of complement in normal serum by hydrogen peroxide and hydrogen peroxide-related oxygen radicals produced by activated neutrophils. *Clinical and experimental immunology*. 90 (1), 72–78.

Singh, J. A. et al. (2016) 2015 American College of Rheumatology Guideline for the Treatment of Rheumatoid Arthritis. *Arthritis & rheumatology (Hoboken, N.J.)*. 68 (1), 1–26.

Sinico, R. A. & Guillevin, L. (2019) *Anti-Neutrophil Cytoplasmic Antibody (ANCA) Associated Vasculitis*. Springer International Publishing.

Smith, R. M. et al. (2023) Rituximab versus azathioprine for maintenance of remission for patients with ANCA-associated vasculitis and relapsing disease: an international randomised controlled trial. *Annals of the rheumatic diseases*. 82 (7), 937–944.

Smolyansky, E. D. et al. (2021) Machine learning models for decision support in epilepsy management: A critical review. *Epilepsy & behavior: E&B*. 123108273.

SNOMED International (2022) *SNOMED Home page* [online]. Available from: https://www.snomed.org/ (Accessed 22 September 2022).

Solans-Laque, R. et al. (2017) Clinical characteristics and outcome of Spanish patients with ANCA-associated vasculitides: Impact of the vasculitis type, ANCA specificity, and treatment on mortality and morbidity. *Medicine*. 96 (8), e6083.

Solans-Laque, R. et al. (2013) Clinical features and mortality causes in a large cohort of spanish patients with anca associated vasculitides. *Annals of the rheumatic diseases*. [Online] 71 (SUPPL. 3), . [online]. Available from: http://ard.bmj.com/content/71/Suppl_3/231.3.abstract?sid=4214b495-9146-49ad-9eff-a445d07e20ce.

Stahn, C. & Buttgereit, F. (2008) Genomic and nongenomic effects of glucocorticoids. *Nature clinical practice. Rheumatology*. 4 (10), 525–533.

Stegeman, C. A. et al. (1994) Association of chronic nasal carriage of Staphylococcus aureus and higher relapse rates in Wegener granulomatosis. *Annals of internal medicine*. 120 (1), 12–17.

Steyerberg, E. W. (2019) *Clinical Prediction Models*. Springer International Publishing.

Steyerberg, E. W. et al. (2001) Internal validation of predictive models: efficiency of some procedures for logistic regression analysis. *Journal of clinical epidemiology*. 54 (8), 774–781.

Steyerberg, E. W. et al. (2013) Prognosis Research Strategy (PROGRESS) 3: prognostic model research. *PLoS medicine*. 10 (2), e1001381.

Steyerberg, E. W. & Harrell, F. E., Jr (2016) Prediction models need appropriate internal, internal-external, and external validation. *Journal of clinical epidemiology*. 69245–247.

Stone, J. H. et al. (2010) Rituximab versus cyclophosphamide for ANCA-associated vasculitis. *The New England journal of medicine*. 363 (3), 221–232.

Stone, J. H. & Wegener's Granulomatosis Etanercept Trial Research Group (2003) Limited versus severe Wegener's granulomatosis: baseline data on patients in the Wegener's granulomatosis etanercept trial. *Arthritis and rheumatism*. 48 (8), 2299–2309.

Strangfeld, A. et al. (2011) Treatment benefit or survival of the fittest: what drives the time-dependent decrease in serious infection rates under TNF inhibition and what does this imply for the individual patient? *Annals of the rheumatic diseases*. 70 (11), 1914–1920.

Strijker, M. et al. (2019) Systematic review of clinical prediction models for survival after surgery for resectable pancreatic cancer. *The British journal of surgery*. 106 (4), 342–354.

Sun, Y.-S. et al. (2024) Risk Factors and Incidence of Serious Infections in Patients With Systemic Lupus Erythematosus Undergoing Rituximab Therapy. *The Journal of rheumatology*. 51 (2), 160–167.

Suppiah, R. et al. (2022) 2022 American College of Rheumatology/European Alliance of Associations for Rheumatology Classification Criteria for Microscopic Polyangiitis. *Arthritis & rheumatology (Hoboken, N.J.)*. 74 (3), 400–406.

Takeuchi, Y. et al. (2017) The influence of the Great East Japan earthquake on microscopic polyangiitis: A retrospective observational study. *PloS one*. 12 (5), e0177482.

Talic, S. et al. (2021) Effectiveness of public health measures in reducing the incidence of covid-19, SARS-CoV-2 transmission, and covid-19 mortality: systematic review and meta-analysis. *BMJ* . 375e068302.

Taruscio, D. et al. (2014) The Italian National Rare Diseases Registry. *Blood transfusion = Trasfusione del sangue*. 12 Suppl 3s606-13.

Tejera Segura, B. et al. (2019) Can we validate a clinical score to predict the risk of severe infection in patients with systemic lupus erythematosus? A longitudinal retrospective study in a British Cohort. *BMJ open*. 9 (6), e028697.

Terrier, B. et al. (2018) Long-term efficacy of remission-maintenance regimens for ANCA-associated vasculitides. *Annals of the rheumatic diseases*. 77 (8), 1150–1156.

The COVID-19 Global Rheumatology Alliance (2020) *The COVID-19 Global Rheumatology Alliance* [online]. Available from: https://rheum-covid.org/ (Accessed 22 August 2020).

The OpenSAFELY Collaborative et al. (2020) OpenSAFELY: factors associated with COVID-19-related hospital death in the linked electronic health records of 17 million adult NHS patients. Epidemiology [online]. Available from: http://dx.doi.org/10.1101/2020.05.06.20092999.

Thery-Casari, C. et al. (2020) Severe infections in patients with anti-neutrophil cytoplasmic antibody-associated vasculitides receiving rituximab: A meta-analysis. *Autoimmunity reviews*. 19 (5), 102505.

Toniolo, A. et al. (2019) The diabetes pandemic and associated infections: suggestions for clinical microbiology. *Reviews in medical microbiology: a journal of the Pathological Society of Great Britain and Ireland*. 30 (1), 1–17.

Trama, A. et al. (2017) Data quality in rare cancers registration: the report of the RARECARE data quality study. *Tumori*. 103 (1), 22–32.

Trivioli, G. et al. (2022) Genetics of ANCA-associated vasculitis: role in pathogenesis, classification and management. *Nature reviews. Rheumatology*. 18 (10), 559–574.

Tuin, J. et al. (2019) Mycophenolate Mofetil Versus Cyclophosphamide for the Induction of Remission in Nonlife-Threatening Relapses of Antineutrophil Cytoplasmic Antibody-Associated Vasculitis: Randomized, Controlled Trial. *Clinical journal of the American Society of Nephrology: CJASN*. 14 (7), 1021–1028.

Van Calster, B. et al. (2019) Calibration: the Achilles heel of predictive analytics. *BMC medicine*. 17 (1), 230.

Van Calster, B. et al. (2018) Reporting and Interpreting Decision Curve Analysis: A Guide for Investigators. *European urology*. 74 (6), 796–804.

Vandenbroucke, J. P. et al. (2007) Strengthening the Reporting of Observational Studies in Epidemiology (STROBE): explanation and elaboration. *PLoS medicine*. 4 (10), e297.

Vanholder, R. & Ringoir, S. (1993) Infectious morbidity and defects of phagocytic function in end-stage renal disease: a review. *Journal of the American Society of Nephrology: JASN*. 3 (9), 1541–1554.

Vassilopoulos, A. et al. (2023) Incidence of serious infections in patients with ANCA-associated vasculitis receiving immunosuppressive therapy: A systematic review and meta-analysis. *Frontiers of medicine*. 101110548.

Verhulst, B. & Neale, M. C. (2021) Best Practices for Binary and Ordinal Data Analyses. *Behavior genetics*. 51 (3), 204–214.

Vickers, A. J. et al. (2016) Net benefit approaches to the evaluation of prediction models, molecular markers, and diagnostic tests. *BMJ* . 352i6.

Vincent, J. L. et al. (1996) The SOFA (Sepsis-related Organ Failure Assessment) score to describe organ dysfunction/failure. On behalf of the Working Group on Sepsis-Related Problems of the European Society of Intensive Care Medicine. *Intensive care medicine*. 22 (7), 707–710.

VOICES, University of Aberdeen (2022) *Vasculitis Outcomes In relation to Care ExperienceS (VOICES)* [online]. Available from: https://www.abdn.ac.uk/iahs/research/epidemiology/vasculitis-1616.php (Accessed 17 August 2022).

van Vollenhoven, R. F. et al. (2010) Longterm safety of patients receiving rituximab in rheumatoid arthritis clinical trials. *The Journal of rheumatology*. 37 (3), 558–567.

Volz, E. et al. (2021) Evaluating the Effects of SARS-CoV-2 Spike Mutation D614G on Transmissibility and Pathogenicity. *Cell*. 184 (1), 64-75.e11.

Voysey, M. et al. (2021) Safety and efficacy of the ChAdOx1 nCoV-19 vaccine (AZD1222) against SARS-CoV-2: an interim analysis of four randomised controlled trials in Brazil, South Africa, and the UK. *The Lancet*. 397 (10269), 99–111.

Wainwright, J. & Davson, J. (1950) The renal appearances in the microscopic form of periarteritis nodosa. *The Journal of pathology and bacteriology*. 62 (2), 189–196.

Wallace, Z. S. et al. (2020) All-cause and cause-specific mortality in ANCA-associated vasculitis: Overall and according to ANCA type. *Rheumatology*. 59 (9), 2308–2315.

Wallace, Z. S. & Miloslavsky, E. M. (2020) Management of ANCA associated vasculitis. *BMJ* . 368m421.

Walsh, M. et al. (2010) Effects of duration of glucocorticoid therapy on relapse rate in antineutrophil cytoplasmic antibody-associated vasculitis: A meta-analysis. *Arthritis care & research*. 62 (8), 1166–1173.

Walsh, M. et al. (2020) Plasma Exchange and Glucocorticoids in Severe ANCA-Associated Vasculitis. *The New England journal of medicine*. 382 (7), 622–631.

Walsh, M. et al. (2012) Risk factors for relapse of antineutrophil cytoplasmic antibody-associated vasculitis. *Arthritis and rheumatism*. 64 (2), 542–548.

Walsh, M. et al. (2022) The effects of plasma exchange in patients with ANCA-associated vasculitis: an updated systematic review and meta-analysis. *BMJ* . 376e064604.

Walton, E. W. (1958) Giant-cell granuloma of the respiratory tract (Wegener's granulomatosis). *British medical journal*. 2 (5091), 265–270.

Ward, H. et al. (2022) Population antibody responses following COVID-19 vaccination in 212,102 individuals. *Nature communications*. 13 (1), 907.

Watanabe-Imai, K. et al. (2017) Clinical characteristics of and risk factors for serious infection in Japanese patients within six months of remission induction therapy for antineutrophil cytoplasmic antibody-associated vasculitis registered in a nationwide, prospective, inception cohort study. *Modern rheumatology / the Japan Rheumatism Association*. 27 (4), 646–651.

Watts, R. et al. (2007) Development and validation of a consensus methodology for the classification of the ANCA-associated vasculitides and polyarteritis nodosa for epidemiological studies. *Annals of the rheumatic diseases*. 66 (2), 222–227.

Watts, R. A. et al. (2015) Classification, epidemiology and clinical subgrouping of antineutrophil cytoplasmic antibody (ANCA)-associated vasculitis. *Nephrology, dialysis, transplantation: official publication of the European Dialysis and Transplant Association - European Renal Association*. 30 Suppl 1i14-22.

Watts, R. A. et al. (2022) Global epidemiology of vasculitis. *Nature reviews. Rheumatology*. 18 (1), 22–34.

Wechsler, M. E. et al. (2017) Mepolizumab or Placebo for Eosinophilic Granulomatosis with Polyangiitis. *The New England journal of medicine*. 376 (20), 1921–1932.

Wegener, F. (1939) Über eine eigenartige rhinogene Granulomatose mit besonderer Beteiligung des Arterien systems und der Nieren. *Beiträge zur Pathologie*. 158 (1), 127–143.

Weiskopf, D. et al. (2009) The aging of the immune system. *Transplant international: official journal of the European Society for Organ Transplantation*. 22 (11), 1041–1050.

Weiskopf, N. G. & Weng, C. (2013) Methods and dimensions of electronic health record data quality assessment: enabling reuse for clinical research. *Journal of the American Medical Informatics Association: JAMIA*. 20 (1), 144–151.

Wiik, A. (2000) What you should know about PR3-ANCA. An introduction. *Arthritis research*. 2 (4), 252–254.

Wilkinson, M. D. et al. (2016) The FAIR Guiding Principles for scientific data management and stewardship. *Scientific data*. 3160018.

Wishart, G. C. et al. (2010) PREDICT: a new UK prognostic model that predicts survival following surgery for invasive breast cancer. *Breast cancer research: BCR*. 12 (1), R1.

Witko-Sarsat, V. et al. (1999) A large subset of neutrophils expressing membrane proteinase 3 is a risk factor for vasculitis and rheumatoid arthritis. *Journal of the American Society of Nephrology: JASN*. 10 (6), 1224–1233.

Wohlwill, F. (1923) Uber die mur mikroskopisch erkenbarre form der periarteritis nodosa. *Arch Pathol Anat*. 246377–411.

Wolff, R. F. et al. (2019) PROBAST: A Tool to Assess the Risk of Bias and Applicability of Prediction Model Studies. *Annals of internal medicine*. 170 (1), 51–58.

World Health Organization (2020) *WHO Director-General's opening remarks at the media briefing on COVID-19 - 11 March 2020* [online]. Available from: https://www.who.int/dg/speeches/detail/who-director-general-s-opening-remarks-at-the-media-briefing-on-covid-19---11-march-2020 (Accessed 14 September 2020).

van der Woude, F. J. et al. (1985) Autoantibodies against neutrophils and monocytes: tool for diagnosis and marker of disease activity in Wegener's granulomatosis. *The Lancet*. 1 (8426), 425–429.

Woywodt, A. et al. (2006) Wegener's granulomatosis. *The Lancet*. 367 (9519), 1362–1366.

Wrapp, D. et al. (2020) Cryo-EM structure of the 2019-nCoV spike in the prefusion conformation. *Science*. 367 (6483), 1260–1263.

Wu, Z. & McGoogan, J. M. (2020) Characteristics of and Important Lessons From the Coronavirus Disease 2019 (COVID-19) Outbreak in China: Summary of a Report of 72 314 Cases From the Chinese Center for Disease Control and Prevention. *JAMA: the journal of the American Medical Association*. 323 (13), 1239–1242.

Wyllie, D. et al. (2021) SARS-CoV-2 responsive T cell numbers and anti-Spike IgG levels are both associated with protection from COVID-19: A prospective cohort study in keyworkers. medRxiv p.2020.11.02.20222778. [online]. Available from: https://www.medrxiv.org/content/10.1101/2020.11.02.20222778v2.abstract (Accessed 20 December 2023).

Xiao, H. et al. (2007) Alternative complement pathway in the pathogenesis of disease mediated by anti-neutrophil cytoplasmic autoantibodies. *The American journal of pathology*. 170 (1), 52–64.

Xiao, H. et al. (2002) Antineutrophil cytoplasmic autoantibodies specific for myeloperoxidase cause glomerulonephritis and vasculitis in mice. *The Journal of clinical investigation*. 110 (7), 955–963.

Yang, J. et al. (2016) Histone modification signature at myeloperoxidase and proteinase 3 in patients with anti-neutrophil cytoplasmic autoantibody-associated vasculitis. *Clinical epigenetics*. 885.

Yang, L. et al. (2018) Risk factors for infectious complications of ANCA-associated vasculitis: A cohort study. *BMC nephrology*. 19 (1), 138.

Ye, Q. et al. (2020) The pathogenesis and treatment of the `Cytokine Storm' in COVID-19. *The Journal of infection*. 80 (6), 607–613.

Ye, X. et al. (2023) Associations of socioeconomic status with infectious diseases mediated by lifestyle, environmental pollution and chronic comorbidities: a comprehensive evaluation based on UK Biobank. *Infectious diseases of poverty*. 12 (1), 5.

Yoo, J. et al. (2018) Birmingham vasculitis activity and chest manifestation at diagnosis can predict hospitalised infection in ANCA-associated vasculitis. *Clinical rheumatology*. 37 (8), 2133–2141.

Zeng, L. et al. (2022) Plasma exchange and glucocorticoid dosing for patients with ANCA-associated vasculitis: a clinical practice guideline. *BMJ* . 376e064597.

Zhang, N. et al. (2023) *FaMeSumm: Investigating and Improving Faithfulness of Medical Summarization* Houda Bouamor et al. (eds.). 10915–10931.

Zhang, N. et al. (2022) Predicting bacterial infection risk in patients with ANCA-associated vasculitis in southwest China: development of a new nomogram. *Clinical rheumatology*. 41 (11), 3451–3460.

Zhao, K. et al. (2021) Increased risk of severe infections and mortality in patients with newly diagnosed systemic lupus erythematosus: a population-based study. *Rheumatology* . 60 (11), 5300–5309.

Zhou, F. et al. (2020) Clinical course and risk factors for mortality of adult inpatients with COVID-19 in Wuhan, China: a retrospective cohort study. *The Lancet*. 395 (10229), 1054–1062.

Zhu, N. et al. (2020) A Novel Coronavirus from Patients with Pneumonia in China, 2019. *The New England journal of medicine*. 382 (8), 727–733.

Zsichla, L. & Müller, V. (2023) Risk Factors of Severe COVID-19: A Review of Host, Viral and Environmental Factors. *Viruses*. [Online] 15 (1), . [online]. Available from: http://dx.doi.org/10.3390/v15010175.

Zubaid, M. et al. (2014) Design and Rationale of Gulf locals with Acute Coronary Syndrome Events (Gulf Coast) Registry. *The open cardiovascular medicine journal*. 888–93.