Biochemical and epidemiological investigations of non-steroidal anti-inflammatory drug usage and related side effects in equids

Marco Duz

MedVet MVM(Res) DipECEIM MRCVS

Submitted in fulfilment of the requirements for the
Degree of Doctor in Philosophy

School of Veterinary Medicine
College of Medical, Veterinary and Life Sciences
University of Glasgow

March 2016

Abstract

Non-steroidal anti-inflammatory drugs (NSAIDs) are widely used in equine veterinary practice. These drugs exert their effect by inhibiting cyclooxygenase (COX) enzymes, which control prostaglandin production, a major regulator of tissue perfusion. Two isoforms of COX enzymes exist: COX-1 is physiologically present in tissues, while COX-2 is up-regulated during inflammation and has been indicated as responsible for the negative effects of an inflammatory response. Evidence suggests that NSAIDs that inhibit only COX-2, preserving the physiological function of COX-1 might have a safer profile. Studies that evaluate the effect of NSAIDs on COX enzymes are all performed under experimental conditions and none uses actual clinical patients. The biochemical investigations in this work focus on describing the effect on COX enzymes activity of flunixin meglumine and phenylbutazone, two non-selective COX inhibitors and firocoxib, a COX-2 selective inhibitor, in clinical patients undergoing elective surgery. A separate epidemiological investigation was aimed at describing the impact that the findings of biochemical data have on a large population of equids. Electronic medical records (EMRs) from 454,153 equids were obtained from practices in the United Kingdom, United States of America and Canada. Information on prevalence and indications for NSAIDs use was extracted from the EMRs via a text mining technique, improved from the literature and described and validated within this Thesis. Further the prevalence of a clinical sign compatible with NSAID toxicity, such as diarrhoea, is reported along with analysis evaluating NSAID administration in light of concurrent administration of other drugs and comorbidities. This work confirms findings from experimental settings that NSAIDs firocoxib is COX-2 selective and that flunixin meglumine and phenylbutazone are non-selective COX inhibitors and therefore their administration carries a greater risk of toxicity. However the impact of this finding needs to be interpreted with caution as

epidemiological data suggest that the prevalence of toxicity is in fact small and the use of these drugs at the labelled dose is quite safe.

Table of Contents

List of Tables

List of Figures

List of Accompanying Material

Published papers:

1. Duz M, Parkin TDH, Cullander RM and Marshall JF, 2015. Effect of flunixin meglumine and firocoxib on ex- vivo cyclooxygenase activity in horses undergoing elective surgery. *American Journal of Veterinary Research*. 76(3), 208-215

Acknowledgments

I initially approached Dr Parkin with a preliminary idea of this work at a congress in Hannover in 2011. Ever since Dr Parkin has supported my intent to pursue the project of the epidemiologic investigations providing invaluable support with his world-class experience. I feel extremely lucky to have been able to work with Dr Parkin, a supportive, knowledgeable supervisor and friend.

I would like to equally thank Dr John Marshall for pushing me into completing successfully the biochemical investigations, presenting my findings at congresses and resulting in the accompanying publication. Dr Marshall is a very talented and driven supervisor, and I can only aspire to be such an inspiring supervisor for my future PhD students.

I also should thank all the veterinary surgeons and nurses at the Weipers Centre Equine Hospital for providing help in collecting the samples for the study in Chapter 2. In particular a thank you to Professor Sandy Love who has always been an invaluable support when taking decisions with my career, through my Master's, residency and PhD. Thank you also to all the PhD students with whom I shared the office over the years. In particular a thank you to Stamatis Georgopoulos, who has shown me the light at the end of the tunnel most certainly lost.

I would like to thank the University of Glasgow for funding these 3 years through the John Crawford Endowment fund.

Also I would like to really thank all the veterinary practices, which have agreed to provide data for the project. Their support is invaluable and I can only wish that more will follow their example in the future as they are a great asset to the veterinary profession, beyond clinical practice itself.

Also a special thank you to my current employer, the University of Nottingham, in particular to Prof. Kate White and Prof. Sarah Freeman for being unbelievably understanding and supportive in the past 16 months,

allowing me to complete this work.

Last but not least I would like to thank my loving wife Chiara, sons Paolo and Matteo and my little girl Tosca, for continuing to love me and for providing a new focus in my life! I love you all immensely!

Author's declaration

I declare that, except where explicit reference is made to the contribution of others, that this thesis is the result of my own work and has not been submitted for any other degree at the University of Glasgow or any other institution.

Signature _____

Printed Name ____Marco Duz_____

List of Abbreviations

AI = Artificial Intelligence

AIC = Akaike Information Criterion

BID = Twice daily ("*Bis In Die*")

BSAVA = British Small Animal Veterinary Association

CI = Confidence Interval

COX = Cyclooxygenase

COX1:COX2 = COX1 to COX2 inhibitory ratio

DACVS = Diplomate of the American College of Veterinary

DACVIM = Diplomate of the American College of Veterinary Internal Medicine

DACT = Diplomate of the American College of Theriogenology

DDFT = Deep Digital Flexor Tendon

DSP = Dorsal Spinous Process

EC_{50} = Half Maximal Drug Effect

EIA = Enzyme Immune Assay

EMIS = Egton Medical Information Systems

EMR = Electronic Medical Record

FIR = Firocoxib

FM = Flunixin Meglumine

GCT = Granulosa Cell Tumour

GEP = Group of Equine Practices

HSCIC = Health and Social Care Information Centre

HPLC = High Performance Liquid Chromatography

IC_{50} = Half Maximal Inhibitory Concentration

IT = Information Technology

IV = Intravenously

KDD = Knowledge Discovery in Databases

LPS = Lipopolysaccharide

NHS = National Health Service

NLP = Natural Language Processing

NSAID = Non-Steroidal Anti-Inflammatory Drug

mg/kg = Milligrams Per Kilogram

mRNA = Messenger Ribonucleic Acid

MT3 = Metatarsal 3

NA = Not Available

OCD = Osteocondrosis Dissecans

OR = Odds Ratio

PAC = Public Accounts Committee

PBS = Phosphate-buffered Saline

PBZ = Phenylbutazone

PCV = Packed Cell Volume

PGA = Prostraglandin A

PGE = Prostaglandin E

PGEM = Prostaglandin E Metabolites

PGG = Prostaglandin G

PGH = Prostaglandin H

PMSS = Practice Management Software Sytem

PRIMIS = Primary Care Information Service

SAVSNET = Small Animal Veterinary Surveillance Network

SID = Once Daily ("*Semel In Die*")

TDDA = Term Domain Distribution Analysis

TID = Three times daily ("*Tris In Die*")

TXA = Thromboxane A

TXB = Thromboxane B

USA = United States of America

VeNom = Veterinary Nomenclature

CHAPTER 1 – Review of the Literature

1.1 General non-steroidal anti-inflammatory drug history: from aspirin to coxibs

The earliest recorded usage of a non-steroidal anti-inflammatory drug (NSAID) dates back to over 3500 years ago, from Ebers papyrus, Hippocrates, Celsus, Pliny the elder, Dioscorides and Galen, which all recommended preparations containing salicylate for the treatment of pain (Vane, 2000). The modern era of anti-inflammatory drug usage starts about 250 years ago with the study by Reverend Edward Stone who described the beneficial properties of willow bark (containing "salicine") to treat pain (Stone, 1763). Initially isolated in 1828 by Buchner, salicine was first synthesised in 1853 by Gerhardt and finally in 1897, in Bayer's laboratories, by Felix Hoffman, who also demonstrated its anti-inflammatory efficacy (Botting, 2010; Jerie, 2006). After two years of clinical trials with low doses, Bayer's management decided to start the production and launched aspirin, as an analgesic drug, worldwide in summer 1899 (Vane, 2000). The introduction of aspirin in the market was a major breakthrough economically and socially in human medicine. Recent reports suggest that the average worldwide consumption of aspirin is approximately 80 tablets/person/year (total production of 50,000 tons a year)(Vane, 2000). Although these numbers refer only to aspirin, they suggest how NSAIDs are part of everyday life for most and that they are among the most frequently administered medications in people.

Although aspirin had been marketed from the start as an anti-inflammatory drug, its mode of action remained elusive for 74 years until the break-through discovery that these drugs act by inhibiting prostaglandin (PG) synthesis (Vane, 1971). Whilst researching the actions of the newly discovered prostaglandins, the same group of authors demonstrated that aspirin interferes with their production *in vitro* (Ferreira et al., 1971; Smith and Willis, 1971). The existence of the

cyclooxygenase (COX) enzyme family, also known as prostaglandin endoperoxide synthase enzymes, was for the first time demonstrated by three independent research groups in the late 1980s (DeWitt and Smith, 1988; Merlie et al., 1988; Yokoyama et al., 1988). For a few years the knowledge was limited to a single enzyme (COX-1), until a second, inducible form of cyclooxygenase (COX-2) was identified (Laneuville et al., 1994). As the roles of the constitutively expressed isoform of cyclooxygenase (COX-1), and the inducible isoform (COX-2) were investigated the deleterious effects associated with NSAID use were attributed to the inhibition of COX-1 (Bakhle and Botting, 1996; Vane et al., 1998). Although the mechanism of action will be further discussed in Chapter 2 of this Thesis, the undesirable side-effects resulting from the use of these drugs is considered to be strongly related to the inhibition of prostaglandin production. In people these include gastrointestinal irritation, renal and hepatic toxicity, interference with haemostasis and reproductive tract problems (Bergh and Budsberg, 2005). It has been estimated that in the late 90s more than 30 million people would take NSAIDs daily and that of the approximately 100,000 humans that were treated for adverse gastrointestinal effects induced by NSAIDs 15% died as a consequence of toxicity (Singh, 1998; Singh and Triadafilopoulos, 1999). In 1999 the gastrointestinal side effects of NSAIDs were the 15th most common cause of death in the USA (Wolfe et al., 1999). These figures may explain why both human and veterinary researchers have recently focused on the production of new NSAIDs, which specifically inhibit the COX-2 iso-enzyme, whilst allowing the physiologic regulation of prostaglandin production by COX-1 iso-enzyme. These so called “selective COX-2 inhibitors” may maximize the beneficial anti-inflammatory effects, with reduced potential for development of side-effects. Drugs that emerged from this work include celecoxib, valdecoxib and rofecoxib. These are often referred to as the coxibs and they have been among the top-selling prescription drugs of any category in human medicine (FitzGerald and Patrono, 2001).

Up to just over a decade ago, non-selective NSAIDs (flunixin meglumine and phenylbutazone) were reported as most commonly used veterinary analgesics in Australia and South Africa (Joubert, 2001; Watson et al., 1996). More recently, coxibs have also been evaluated in veterinary medicine for the treatment of pain and inflammation in dog, cat, and horse (Brideau et al., 2001; Giraudel et al., 2009; Kvaternick et al., 2007; Marshall et al., 2011; Orsini et al., 2012; Punke et al., 2008).

1.2 Structure and function of cyclooxygenase enzymes

Despite the different physiologic functions, COX-1 and COX-2 enzymes share a rather similar structure with 61% amino acid homology (Appleby et al., 1994). Their structure constitutes approximately 600 amino acids organized to form a long hydrophobic channel with a hairpin turn at the end (Picot et al., 1994). The change with a valine residue in place of an isoleucine at the position 434 and 523 results in a 25% enlargement of the hydrophobic channel and active site of COX-2 due to a conformational change in phenylalanine 518. This larger diameter allows larger molecules, such as coxibs, to selectively bind to COX-2 but not to COX-1 (Kurumbail et al., 1996; Luong et al., 1996).

Currently, three isoforms of COX enzymes have been identified, namely COX-1, COX-2 and COX-3. The COX-3 enzyme is a variant of COX-1 rather than a distinct isomer, but is expressed mainly in the cerebral tissue and only to a minor extent in other tissues (Chandrasekharan et al., 2002) and will not be mentioned further in this review. The localization and activity of COX-1 and COX-2 enzymes have been investigated intensely since the early 1990s (Vane et al., 1998). COX-1 is present under basal conditions in many cells, including mucosal cells of the gastrointestinal tract, endothelial cells, platelets and the renal medullary collecting ducts and interstitium (Harris et al., 1994; Kargman et al., 1996). The COX-2 isoform

is physiologically present in low concentrations in monocytes, macrophages, smooth muscle cells, fibroblasts and chondrocytes (Smith, 1998), but is also constitutive in some tissues, such as kidney, brain or in canine pyloric and duodenal mucosa (Mitchell and Warner, 1999; Vane and Botting, 1995; Wooten et al., 2008). While COX-1 is considered a constitutive isoform, expressed mainly during physiological conditions, the expression of COX-2 is induced in determinate conditions, such as after stimulation of inflammatory cytokines or exposure to toxins, such as lipopolysaccharide (LPS). Further differences include a TATA box sequence located 25 base pairs upstream of the transcriptional start site, which is present in COX-2 but not in COX-1 enzyme (Appleby et al., 1994). The COX-2 gene also contains several potential transcription regulatory sequences (Appleby et al., 1994). During inflammation and injury, these elements allow COX-2 to respond to a wide variety of stimuli through up-regulation of protein expression. In contrast, the COX-1 gene lacks these elements and possesses the characteristics of a 'housekeeping' gene (Garavito and DeWitt, 1999; Luong et al., 1996). Although the genes, mRNA transcripts, and protein structures of the COX enzymes differ, the COX enzymes both use the same substrate to produce an identical product (Garavito and DeWitt, 1999). The first reaction catalysed by the COX enzymes is the oxidation and cyclization of arachidonic acid to prostaglandin G_2 (PGG_2) at the COX site. This is followed by the reduction of PGG_2 to prostaglandin H_2 (PGH_2) at the peroxidase site (Tazawa et al., 1994). The final product of the COX enzymes, PGH_2, is subsequently metabolized further into products with a wide range of biological activities (Garavito and DeWitt, 1999). Thromboxane synthase in platelets converts PGH_2 into thromboxane A_2 (TXA_2), essential for platelet aggregation (Garavito and DeWitt, 1999; Tazawa et al., 1994).

During inflammation, at the site of injury, the expression of COX-2 leads to increased PGE_2, while inhibition of COX-2 results in a decreased inflammatory response (Vane et al., 1998). However, the perception that

COX-2 is a “bad” enzyme and COX-1 is a “good” enzyme is probably overly simplistic as there is some overlap in the functions of these isoforms and COX-2 has been shown to have physiologic function in certain tissues such as pyloric and duodenal mucosa in the dog (Wooten et al., 2008).

1.3 Evaluation of cyclooxygenase selectivity

Selectivity of COX-2 versus COX-1 is often expressed as the COX-1 to COX-2 inhibitory ratio (COX-1:COX-2), usually expressed as the concentration necessary to inhibit 50% of COX activity (IC_{50}), usually measured by stimulating cells capable of expressing products of these enzymes (Streppa et al., 2002). Recently it has been debated whether using IC_{50} is an accurate reflection of selectivity ratio and of concentrations and activity *in vivo* and that IC_{80} (the concentration that inhibits 80% of COX activity) would be more appropriate for this purpose (Furst, 1999; Hinz and Brune, 2008; Kay-Mugford et al., 2000; Warner et al., 1999). In general terms, the higher the value, the more selective for COX-2 the drug is. In relation to their effect on COX-1 and COX-2 activity, NSAIDs are included in one of four categories (Frölich, 1997). The first of these includes the so called selective COX-1 inhibitors, routinely used in clinical settings to reduce platelet aggregation (e.g. aspirin) (Heath et al., 1994; Warner et al., 1999). At higher doses these drugs may exert an effect at sites other than the vascular bed and may result in severe gastrointestinal and renal toxicity (Frölich, 1997). The second group includes NSAIDs that non-selectively inhibit both isoforms of the COX isoenzymens and include the drugs most commonly used in equine medicine such as flunixin meglumine and phenylbutazone, but also other drugs routinely used in human as well as veterinary patients such as indomethacin, piroxicam, ibuprofen and ketoprofen (Frölich, 1997; Lees et al., 2004b). The third group of NSAIDs includes drugs with a higher affinity for COX-2 but that still induce some degree of COX-1 inhibition (COX-2 preferential inhibitors). Meloxicam, nimesulide and carprofen are the most popular of these drugs. Meloxicam

has been evaluated in double blinded studies with a significant reduction in the number of gastrointestinal complications in comparison to non-selective NSAIDs (Valat et al., 2001; Yocum et al., 2000) and these findings support the increased safety of NSAIDs with increased COX-2 selectivity. However a study on meloxicam showed that this drug might have variable effect on prostanoid production despite serum concentration well within the therapeutic range for this drug (Blain et al., 2002). The last group includes drug with a high affinity for COX-2 and are called selective COX-2 inhibitors (FitzGerald and Patrono, 2001). Drugs of this group include the coxibs and their potent inhibition of COX-2 along with a normal function of COX-1 has been shown to offer significant improvements compared with NSAIDs of the previous three groups in veterinary studies (Beretta et al., 2005; FitzGerald and Patrono, 2001; Lees et al., 2004b; Punke et al., 2008). These NSAIDs are also referred to as COX-1 sparing, COX-2 specific, COX-2 preferential or COX-2 selective without however any true definition of the magnitude of the ratio used to define each term (Papich, 2008).

1.4 Cyclooxygenase selectivity among species, *in vivo* and *ex vivo*

Some disagreement between studies with respect to COX selectivity for different NSAIDs is present in the peer reviewed veterinary literature. Whilst earlier studies using purified enzymes *in vitro* reported a COX-1:COX-2 ratio of 1275 (Gierse et al., 2002) for deracoxib in dogs, subsequent studies performed on a whole blood model had a ratio of only 12 (McCann et al., 2004). For carprofen the measured COX selectivity was ~100 fold different using purified canine enzyme systems compared to canine macrophages (Kay-Mugford et al., 2000; Ricketts et al., 1998) or ~1000 fold difference comparing whole blood with cell culture methods (Wilson et al., 2004). These findings suggest that COX selectivity may vary significantly depending not only on the assay used but also on the model adopted (e.g. *in vitro* or *ex vivo*) (Vane and Botting, 1995). Further, COX

selectivity for various NSAIDs varies widely among species and data regarding one species may not necessarily apply to another (Brideau et al., 2001). Marked difference in carprofen selectivity has been reported in studies using canine cell lines (COX-1:COX-2 IC_{50} ratio of 129) (Ricketts et al., 1998) compared to sheep and rodent cell lines (COX-1:COX-2 IC_{50} ratio of 1) (Vane and Botting, 1995). Further, COX-2 selectivity of etodolac, an acetic acid derivative NSAID, is 10 times greater in people than in dogs (Glaser, 1995; Gierse et al., 2002). While a COX-2 preferential inhibitor (IC_{50} of 16.6) in the dog (Streppa et al., 2002), carprofen is non-selective in the horse (IC_{50} of 1.9) (Beretta et al., 2005). In conclusion these studies evidence not only the importance of assessing species-specific COX selectivity but also that evaluating *ex vivo* as well as *in vivo* models is of pivotal importance to assess the effect that these drugs have on a clinical patient.

1.5 Assays for cyclooxygenase activity determination

It is relatively well accepted that whole blood assays are the gold standard for the determination of COX selectivity *in vitro* (Patrignani et al., 1994) compared to other models using isolated cells or enzyme systems (Li et al., 1995; Noreen et al., 1998; Ogino et al., 1997; Pairet, 1998; Vago et al., 1995). In fact, whole blood assays include most of the components that may normally affect drug activity such as proteins, cells, platelets and circulating enzymes all at physiological concentrations. Whole blood assay measures thromboxane (TXB_2) produced by stimulated platelets which is a product proportional to the activity of COX-1 enzyme, whilst COX-2 activity is inferred from the amount of prostaglandin E_2 produced by leukocytes (Brideau et al., 2001). Non-steroidal anti-inflammatory drugs are usually highly protein bound which in turn means that only a small amount of free active drug is present in blood, which is better expressed in whole blood assays compared to cell cultures or purified enzyme assays (Wilson et al., 2004).

Whole blood *ex vivo* models are considered most clinically relevant as the blood samples for COX activity assays are obtained following drug administration and therefore account for differences in drug metabolism, pharmacokinetic variables and drug accumulation in tissues and are more likely to reflect physiologic and pathologic conditions and predict clinical outcome (Blain et al., 2002).

Pharmacokinetic-pharmacodynamic studies have also been performed to derive the clinically most appropriate dosage for each NSAID (Giraudel et al., 2005; Lees et al., 2004a, 2004b). An inhibition of COX-2 of about 80% (IC_{80}) is necessary to predict a clinical effect and drugs producing 50% of inhibition may not produce a significant therapeutic effect (Hinz and Brune, 2008).

1.6 Non-steroidal anti-inflammatory drug toxicity in horses

Phenylbutazone, an NSAID, is one of the most commonly used drugs for treatment of equine athletes with signs of musculoskeletal pain (Soma et al., 2012), and it is generally believed to be reasonably tolerated in horses when administered at the recommended dosage and dosing interval. In 1979 and again in 1981, two separate research groups (Lees and Michell, 1979; Snow et al., 1981, 1979) demonstrated that administration of this drug to horses (4–8 mg/kg /day) can cause severe adverse effects, including gastric ulceration, renal dysfunction and inflammation and ulceration of the mucosa of the large colon, particularly of the right dorsal colon. These effects have subsequently been confirmed by several other studies (Cohen et al., 1995; Collins and Tyler, 1985, 1984; Hough et al., 1999; Karcher et al., 1990; MacAllister et al., 1993; MacKay et al., 1983; Meschter et al., 1990a, 1990b, 1984; Simmons et al., 1990).

Earlier studies on phenylbutazone used high dosages that today would be beyond what are considered safe. Dosages at 10mg/kg daily for 14 days (Snow et al., 1979) or 13.5mg/kg daily (Meschter et al., 1990a, 1984) were

administered and very frequently resulted in significant renal, gastric and colonic damage. In a study, overdosing phenylbutazone (4.4mg/kg TID) and flunixin meglumine (1.1mg/kg IV TID) and ketoprofen (2.2mg/kg IV TID) for 12 days resulted in gastric glandular ulceration in all treated horses, whilst colonic ulceration was present only in phenylbutazone treated horses (MacAllister et al., 1993). The study by Collins and Tyler (1984) determined that total daily doses up to 8.8mg/kg were generally safe while toxicity was more likely to ensue at higher doses. The findings of this study determined the currently recommended dose for phenylbutazone in horses.

Albeit side effects are generally rare when NSAIDs are administered at labelled doses, concurrent dehydration has also been shown to increase the risk for renal medullary crest necrosis in horses (Gunson and Soma, 1983), which appear to be a species more susceptible to this side effect than others, such as dogs and rabbits (Black, 1986; Faulkner et al., 1984; Read, 1983). Overdosing has also shown to cause renal crest necrosis in horses receiving either phenylbutazone and flunixin meglumine, while horses receiving ketoprofen did not (MacAllister et al., 1993).

The gastrointestinal system is also sensitive to NSAID toxicity and gastric mucosa and the right dorsal colon appear to be most susceptible (Hough et al., 1999). A decrease in mucosal blood flow secondary to prostaglandin production inhibition by COX-1 enzyme has been suggested as the main mode of action (Richter et al., 2002). A study looking at the effect of flunixin meglumine on renal plasma and blood flows failed to find a significant effect (Held and Daniel, 1991), but the concurrent effect on gastrointestinal blood flow remains undetermined. The study by Meschter and colleagues (1984) described that overdosing of phenylbutazone caused wall degeneration of small mucosal veins, with dilation and hyaline degeneration resulting in erythrodiapedesis and leukodiapedesis, submucosal oedema and ulceration. Another study has documented an increase in gastrointestinal mucosa permeability, particularly at gastric

level (D'Arcy-Moskwa et al., 2012). In that study sucrose permeability test (Hewetson et al., 2006; O'Conner et al., 2004) was used to ascertain the permeability of the gastric epithelium and found that the effect of phenylbutazone was significantly greater than that of meloxicam (D'Arcy-Moskwa et al., 2012). Another study comparing the effect of phenylbutazone to that of suxibuzone on the gastric mucosa found that both drugs induced gastric ulcers but phenylbutazone much more so than suxibuzone, compared to a placebo group (Monreal et al., 2004). That study aimed to compare the effect of these two drugs and concluded that suxibuzone is generally safer than phenylbutazone. However, significant flaws in study design make the results of that study difficult to interpret. The study consisted of a comparison of gastric ulceration score in three groups of horses receiving either a placebo, phenylbutazone or suxibuzone (Monreal et al., 2004). Whilst the study found that the majority of horses with gastric ulceration had received phenylbutazone and only a small proportion suxibuzone, none of the horses had undergone gastroscopy before the start of the study. Therefore the prevalence of gastric ulceration in the population before the study commenced was unclear. This was a major limitation since gastric ulceration is often present without clinical signs (Murray et al., 1989). Further, once metabolized by the liver, suxibuzone is converted to active phenylbutazone and released in the blood stream. Ultimately the effect of suxibuzone is similar to that of oral phenylbutazone without the "topical" direct effect on the gastric mucosa after ingestion and could therefore be comparable to intravenous phenylbutazone (Andrews et al., 2009). Some argued that suxibuzone may be more palatable than phenylbutazone, but a study found that a commercial preparation of suxibuzone has similar palatability to that of phenylbutazone (Longhofer et al., 2008) while others found that acceptability was better with suxibuzone (Sabaté et al., 2009). Another later study that compared these drugs at label doses but examined the gastric mucosa before and after the administration of these drugs found no difference between drugs or the placebo (Andrews et al., 2009).

The potential for hepatotoxicity of NSAIDs has also been described in many species (Lee, 2003), including horses although this appears to be less common than gastrointestinal and renal toxicity (Lees et al., 1983). In humans, hepatotoxicity from NSAIDs administration has been described as an intrinsic, dose related reaction but also as an idiosyncratic reaction (Bjorkman, 1998; Tolman, 1998). Most of NSAIDs undergo some degree of hepatic metabolic pathway and severe liver disease might affect drug metabolism. However, the concern of using NSAIDs in patients with liver disease is more focused on the excessive active drug that remains available once the liver is unable to metabolise it effectively (Papich, 2008; Sanchez, 2010).

1.7 Knowledge discovery in databases

Medical record computerisation has been introduced since the mid-1960s (Salton, 1971; Salton and Lesk, 1968). Initially information was stored using one of two main types of systems. The first used a rigid but structured protocol that offered optimal retrievability of the stored information. However, it lacked flexibility and limited the freedom of expression of clinicians storing the information and poorly adapted to unique characteristics of certain clinical cases. The alternative system allowed the storage of free unstructured text in a manner similar to paper records. Although this did not restrict freedom of expression it made data retrieval rather troublesome (De Bruijn and Martin, 2002; Garten et al., 2010).

Systems for the automated retrieval of information from these databases have also been developed for 40 years (Anholt et al., 2014a; Harman, 1996; Heinze et al., 2001; Krallinger et al., 2008; Salton, 1971). For unstructured free-text database most effective systems involve keyword frequency analysis (Anholt et al., 2014a; Kreis and Gorman, 1997; Petrova et al., 2012). The main goal of these retrieval systems is to find the right

index terms so that a query will identify and retrieve the most appropriate documents, cases or records (Kreis and Gorman, 1997).

Tools for text-based data mining have proved useful in human medicine for over a decade in knowledge discovery in databases (KDD) which has been defined as the use of systems to seek and extract similarities of documents in a collection automatically or determine what is unusual about a particular collection (Frawley et al., 1992). A pattern that is interesting according to predefined criteria and certain enough is defined as “knowledge”, whilst “discovered knowledge” is the output of a program that monitors the patterns of a set of facts in a database (Frawley et al., 1992).

Automated KDD uses Artificial Intelligence (AI) to discover useful knowledge from a collection of data. Data mining, referred to as the analysis step of KDD, aims to extract information of interest from a dataset and transform this information into an understandable structure for further use (Piatetsky-Shapiro 2000). A practical application of KDD has been described by Goldman and colleagues (1999). In their study these authors, through the use of Term Domain Distribution Analysis (TDDA), identified that human thoracic lung cancer tumours affect more frequently the right rather then the left lung with a ratio of 3:2 in a small dataset (178 cases, with records of ~250 words/case and a total collection size of 321kbytes). The TDDA used in that study consisted of the automated tracking of the frequencies for specific predefined terms and subsequently highlighting any significant difference from the expected term distribution and frequency (Goldman et al., 1999).

Current research of KDD is focused on methodologies to structure the unstructured textual information to conform free-text information to a predefined format (Garten et al., 2010). Traditional information retrieval systems simply rank documents relevant to a given query, mostly using a threshold function based on keyword frequency analysis with the ultimate

goal of identifying similar documents (Garten et al., 2010). The Electronic Medical Record (EMR) has been a major goal in Health Information Management for decades (Heinze et al., 2001). Heinze and colleagues (2001) reported the findings of LifeCode®, a project using a state-of-the-art Natural Language Processing (NLP) search engine, designed to mine the transcriptions of dictated clinical records from a wide range of medical specialties to facilitate clinical and epidemiological studies on topics of interest for both medical and pharmaceutical industries. The success of previous systems had been seriously limited due to the relative inaccessibility of the information in free-text clinical documentation (Heinze et al., 2001). Attempts to change the documentation habits of physicians have not had significant success largely due to the increased time and inconvenience associated with using computer interfaces that require formatted input. Further, numerous consultations with practicing physicians have taught us that there is a basic inability of fully structured systems to fully adapt to the unique characteristics of each case (Garten et al., 2010). Nowadays the most common applications of these systems in medical research are represented by Medline or Pubmed, which enhance the role of NLP by integrating statistical algorithms to refine search results (Workman and Stoddart, 2012).

The biomedical literature holds almost all our understanding on a wide variety of topics, but remains dispersed across many journals. In order to integrate medical knowledge, connect important facts across publications and generate new hypotheses, authors must organize and encode the contents of the literature. By creating databases that structure the knowledge available on a specific topic, the value of the literature becomes much greater than the sum of the individual reports (Garten et al., 2010).

1.8 Text mining in knowledge discovery in databases

Recent work in biomedical text mining has focused on techniques developed for NLP, and text mining can be thought of as a subset of NLP, which is defined as a conversion of human language into computable formats (Garten et al., 2010). However, text mining also uses techniques developed in the field of machine learning that automatically recognize complex patterns in large text datasets (Garten et al., 2010). Text mining consists of two main steps: identification of documents that may contain the desired information, and subsequent extraction of the information from this set of documents (Krallinger et al., 2008). Text mining is the automated equivalent of reading for people, as just like a reader, the machine selects what they will read, then identifies important entities and relations amongst these entities and finally combines the new information to other parts of the article (De Bruijn and Martin, 2002). The identification process, or "automated reading" can also be subdivided in four general subtasks: text categorization, named entity tagging, fact extraction and collection-wide analysis (De Bruijn and Martin, 2002). The process of text categorisation is an information retrieval process that divides the document into separate subsets belonging to predetermined categories (Iliopoulos et al., 2001). Named entity tagging processes use character-by-character or word-by-word pattern analysis to identify entities fitting a named category (De Bruijn and Martin, 2002). Fact extraction is the identification of entities and their interactions or relationships, which is hardly achievable by an automated process, but can somewhat be obtained by statistical co-occurrence, imperfect parsing or co-reference resolution (De Bruijn and Martin, 2002). Collection-wide analysis includes the processes of integrating information between documents to discover new knowledge (De Bruijn and Martin, 2002; Weeber et al., 2000). This process is summarised in Figure 1.1.

1.9 Electronic medical records in medical practice

The National Health Service (NHS) is the publicly funded health care system in place in the United Kingdom. In April 2004 the United Kingdom department of Health formed NHS Connecting for Health, a group that among it's aims had that of creating a centralised electronic clinical record system that connects practice management systems software (PMSS) of general practitioners and hospitals.

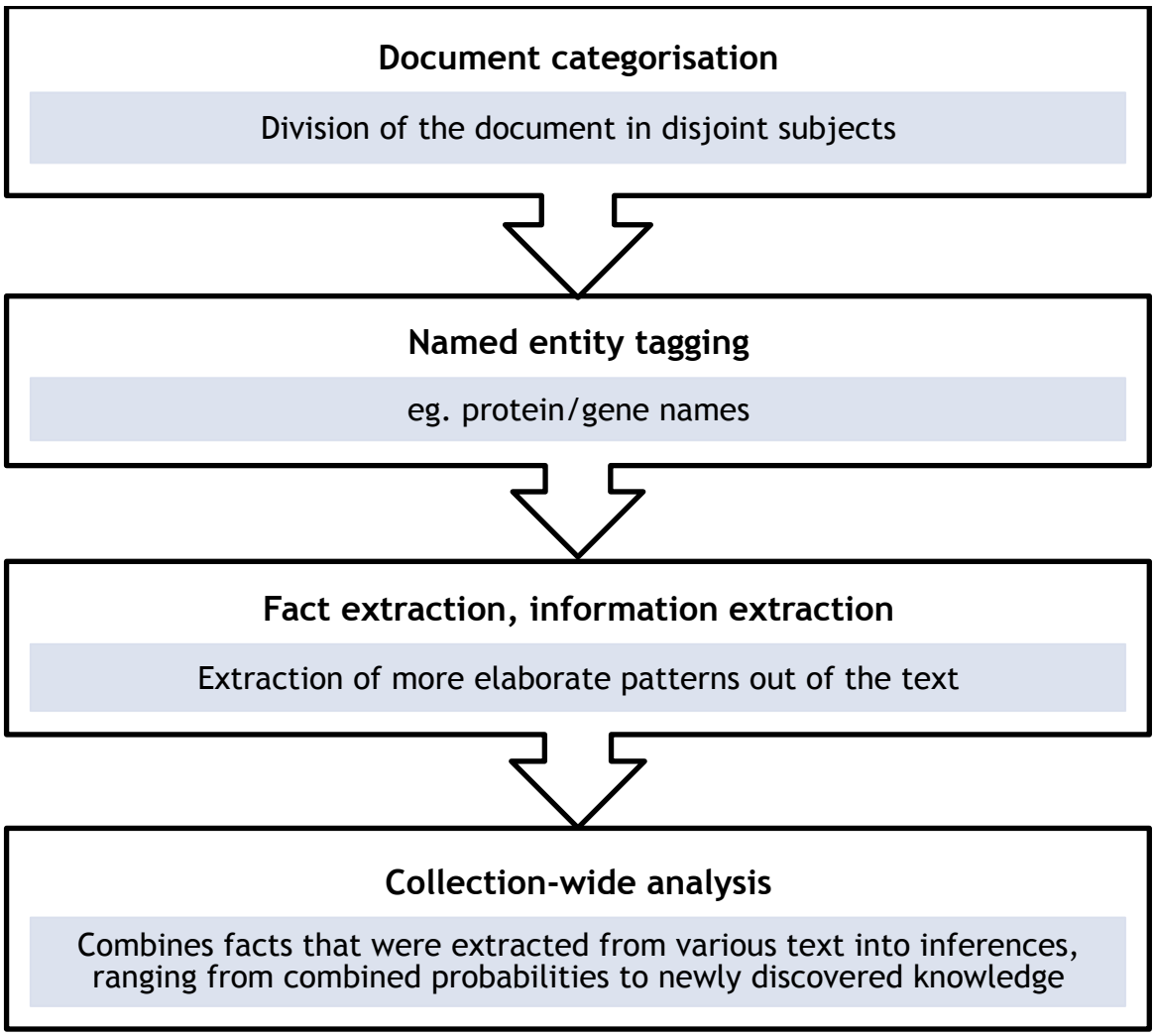

Figure 1.1: Text mining as a modular process (adapted from De Bruijn & Martin 2002)

Among many other benefits, it was envisaged that this could ultimately provide an invaluable pool of information suitable for epidemiological research for authorized health professionals. Due to several political, social and ethical reasons the aims of this project have proved

problematic to achieve and at a great cost as the initial budget of £2.3 billion over three years have been revised to £12.4 billion over 10 years according to the Department of Health's National Program for IT in the NHS of 2006. The project aimed to provide integrated care records services, electronic prescribing and appointments booking, integrating the use of software for access of digital medical imaging and to improve performance management of primary care, but also to provide a central email and directory service. In 2007 the Public Accounts Committee (PAC) expressed some serious concerns that the project would not result in any significant clinical benefit for the patients and in 2011 a lack of significant progress of the project was still noted by PAC (House of Commons Committee of Public Accounts, 2011; House of Commons Health Committee, 2007).

The current record systems adopted by the NHS include entries that are a mixture of text and "Read codes". General practitioners (GPs) are free to adopt whichever PMSS they think would best suit the needs of their practice, which complicates how data can be accessed for research purposes. It is interesting to note that EMR are assessed to see if general practitioners meet NHS targets through qualitative outcome frameworks (QOF) and GPs' good compliance with read codes is followed with a financial incentive. Previously the validity and utility of the information of preliminary electronic patient record systems have been subject to validation against paper records or patient surveys (Pringle et al., 1995; Whitelaw et al., 1996; Wilson et al., 1995). In Britain the Primary Care Information Service (PRIMIS), developed by the University of Nottingham in 2000, was designed to assist general practitioners to effectively use their PMSS to optimize clinical record management and ultimately improve patient care. This software package contains an internal toolkit for measuring reliability of data collection, which offers a form of internal validation system regarding patient clinical and personal data (reference website www.primis.nottingham.ac.uk) (Hassey, 2001). Currently the EMIS system is one of the most common PMSS (53% of general practitioners

in 2011) in use in the United Kingdom that allows storage of data including medical history, acute and repeat medication records and results of blood and radiology investigations posted back to a surgery from a hospital (www.emishealth.com). In September 2014 the system was connected online linking 3750 general practices out of the approximately 9800 present in the United Kingdom and enables other connected medical practices and patients to access their medical records online. The validity of this clinical record system was evaluated by comparing the stored data with information obtained directly from the patients through a questionnaire and results have shown that the information retrieved by this system is valid, complete and accurate (Hassey, 2001). SystemOne is also a commonly used system, available since 2008, which also uses Read codes extensively, but unlike other systems, all data is stored on remote servers. In 2015 the company IMS MAXIMS released a free PMS, which is pioneering open-source software usage in the NHS and might offer a more cost effective alternative for the future. Clinical Practice Research Datalink (CPRD) is a governmental, non-profit research service funded by the NHS National Institute for Health Research (NIHR) and the Medicines and Healthcare products Regulatory Agency (MHRA), a part of the Department of Health. This service provides anonymised primary care records for public hearth research since 1987. In Scotland the Primary Care Clinical Information Unit (PCCIU) was founded in 1999 to collect data from and provide a reporting service to general practice in Scotland.

One of the main issues with these systems relates to the confidentiality of patient data shared by General Practitioners who are obliged to follow the law in the Data Protection Act, but at the same time have the “duty to share” when this is in the best interest of the patient.

In 2013 the Department of Health created the Health and Social Care Information Centre (HSCIC) as an initiative aimed at gathering and analysing data from general medical practice clinical systems in NHS England to help decision makers improve the quality and efficiency of

frontline medical care. The program was called care.data and is managed by Atos, a French company, which has received controversial criticism for the lack of clarity around the ability for patients to opt out the project so that the project was stopped in May 2014, only to be resumed for few months in 2015 before being again paused due to the confidentiality concerns still remaining unresolved. The project was very expensive and data acquisition was mostly suitable for the industry and not for public health research.

1.10 Electronic patients records in companion animal and equine veterinary practice

Compared to the human health sector, the veterinary market lacks a centralised body such as the NHS that attempts to govern patient data management. Although several PMSS are available to veterinary practices, there has been little attempt to centralize the stored information and no attempt has been made to validate these systems for their usefulness in using the stored information for epidemiological research. Further the many available systems seldom allow a simple transfer of patient clinical detail between practices. This offers several challenges to data collection and the gathering of specific clinical information regarding the animal population in a specific country such as the United Kingdom or worldwide.

Recent efforts have focused on the prospective collection of data from veterinary practices selected on the criteria of using a certain PMSS and being willing to participate. The Veterinary Companion Animal Surveillance System, also known as VetCompass, is a project operated by the Royal Veterinary College, University of London in collaboration with the University of Sydney in Australia, to investigate the range and frequency of small animal health problems seen in veterinary practice (http://www.rvc.ac.uk/vetcompass). This project uses a predefined coding system (VeNom – Veterinary Nomenclature) that Veterinary

surgeons from participating practices must utilise to classify each case they deal with into one of these categories. In March 2015 the project included data from over four million small animal patients, from 450 veterinary practices in the UK. The Small Animal Veterinary Surveillance Network (SAVSNET) project is another initiative born from the collaboration between the British Small Animal Veterinary Association (BSAVA) and the University of Liverpool to monitor the current and future disease status, gather data for research and educate public and professionals to improve the management of disease in the small animal population in the United Kingdom (http://www.savsnet.co.uk/). The project in late 2015 involved over 50 small animal veterinary practices. Data is collected using a point-and-click application integrated in the practice's PMSS. Veterinary surgeons from participating practices must then use this application to classify cases they see under a certain category (e.g. body system involved) (Radford et al., 2011). For a random subset of the clinic population a further set of detailed questions are asked to further describe/fit the case into a predetermined category. Both these projects offer the advantage that data are classified in predetermined categories by the veterinary clinicians that input the clinical data to the PMSS and therefore data is already ordered and ready for research. The downside is that clinicians are forced to fit the finding of a clinical case to a certain category, even though the clinical case they are evaluating might subsequently turn out to belong to another category or might just not fit well into any of the categories available. Also these processes require a serious long-term commitment by clinicians that are required to learn these coding systems. Ultimately, to date no validation has been performed comparing the free-text clinical records to that of these ad-hoc categorisations and their sensitivity and specificity remain undetermined.

These projects will offer an excellent support in infectious disease surveillance as they aim to provide veterinary surgeons with real-time information regarding disease outbreaks which would result in prompt

patient testing and effective implementation of preventive strategies (O'Neill et al., 2012a; Radford et al., 2011; Ward and Kelman, 2012). As in human medicine, this could have positive repercussions on both animal welfare and economically for society and veterinary practice (Erstad, 2003). However, these projects suffer significant limitations at the present time, such that only practices using PMSS that allow third parties to access the clinical records in real time can be included.

Other examples utilizing computer-based patient record analysis for retrospective descriptive studies are sparse in veterinary medicine although they have been published in increasing number over the past few years (Boden et al., 2005; Boden and Parkin, 2008; Cameron et al., 2014; Lam et al., 2007a; Ortiz-Pelaez and Pfeiffer, 2008; Oswald et al., 2010).

Another example includes ProActive Insight, a service sponsored by Merial offered to veterinary practices using Merial vaccines since 2013, which uses large datasets from equine practices in the United Kingdom. This service does not aim to use clinical data for epidemiological research, but has the sole intent to maximise income generation while reducing costs for veterinary practices (http://www.proactiveinsight.co.uk). This service is run by a third party company, Veterinary Insights ltd., partly supported by Merial and works by identifying areas that might improve business efficiency and profitability. This is achieved by analysis of key performance indicators aimed at building a picture of how the practice is performing compared to other similar practices throughout the country. This service aims to provide veterinary practices with business intelligence to exploit market trends and support better business decision-making. Since this project is aimed at improving the business aspect of veterinary practice, data has not been used for scientific knowledge advancement to date.

Like in human public health, there are limitations to collection of representative data samples from veterinary practice as it would be

difficult, if at not impossible, to convince the vast majority of veterinary practices to conform to a universal format for data storage. A solution might be to have a ruling institution, such as the Royal College of Veterinary Surgeons in the United Kingdom, to guide this process. Although this may seem logical in general terms, such a process would be impossible to impose as it would effectively limit the freedom of veterinary practices to choose one PMSS system over another. Further, an agreement between research groups would be necessary to decide what the most appropriate system would be in terms of providing data suitable for research. Ultimately, the lack of compliance by veterinary surgeons that often do not wish to share the clinical information of their patients with external investigators is also a major limiting factor. In fact many see disclosing anonymous clinical details to third parties, even if for the simple scope of scientific research, as a breach of client confidentiality. This is why clients enrolled in recent prospective projects are given the opportunity to opt out of these studies when their animal presents for clinical evaluation (O'Neill et al. 2011). Whilst this is going to be gold standard for data acquisition and real time disease surveillance, this is not possible for the studies that use a retrospective analysis of clinical records. However, secure handling of large anonymised datasets is unlikely to ever be a threat to confidentiality between veterinary surgeons and their clients (Balas et al., 2015).

An alternative strategy might consist of convincing veterinary practices of the importance of collecting good quality data suitable for epidemiological research, as the results of such research might in return provide veterinary practice with useful information to improve patient care, owner satisfaction and ultimately increasing client trust and yearly revenues. This approach might pay in the longer term, but its implementation is expected to be slow and would require a significant effort in terms of public engagement and advertising with veterinary practices.

1.11 Non-steroidal anti-inflammatory drugs in veterinary practice

Most information on the preferences of NSAID choice by veterinary surgeons has been obtained through questionnaire-based studies (Dujardin and van Loon, 2011; Hubbell et al., 2010; Joubert, 2001; Watson et al., 1996). Although this may give information on what veterinary surgeons know and think about these drugs, none of these studies provide objective details on what is actually used in practice. Because of their nature, questionnaires may be subject to bias as interviewed subjects are likely to give answers that are more compliant with official recommendations for the topic investigated so that the reliability is usually moderate at best (Helmerhorst et al., 2012). The results of questionnaire-based studies applied to drug usage should therefore be interpreted more as reflection of veterinary surgeon's knowledge rather than an indication of what is actually being administered to the patients. One of the earliest surveys came from Australia from nearly 20 years ago and describes the use of analgesics, including corticosteroids and opioids along with NSAIDs, in dogs and cats (Watson et al., 1996). In that study descriptive statistics were applied to describe preferences by veterinary surgeons to treat a variety of conditions. A similar study was performed in South Africa a few years later and reported similar findings with phenylbutazone and flunixin meglumine being the NSAIDs of choice (Joubert, 2001). It is interesting to note how phenylbutazone was the drug of choice for many musculoskeletal conditions in these studies. It is legitimate to hypothesize that the type of NSAIDs used have largely changed since those studies were performed, as a wide variety of newer NSAIDs with fewer side effects have become available over the past decade, especially for use in small animal practice. Several studies have provided evidence of a lower potential for side effects (King et al., 2010; Moreau et al., 2003; Slingsby and Waterman-Pearson, 2001). Ideally this type of survey should be repeated periodically to ensure that an up-to-date evaluation of drug preferences is provided. In fact, newer drugs such as meloxicam,

carprofen and ketoprofen were reported as those being most frequently prescribed to small animal patients in a survey conducted a few years later among French veterinarians (Hugonnard et al., 2004). However, a similar more recent survey performed among equine veterinary surgeons in the Netherlands reported that phenylbutazone and flunixin meglumine were most used (Dujardin & van Loon 2011). This lack of change in the type of NSAID prescribed is not entirely surprising as the number of new NSAIDs licensed for horses remains limited to meloxicam and only more recently to firocoxib. A list of veterinary products containing NSAIDs in United Kingdom, United States of America and Canada is available in Appendix 1.11).

Despite the significant effort required to obtain data through survey-based research projects, the results obtained should be interpreted as reflection of veterinary knowledge and experience rather that an actual direct assessment of what is actually used. This is where automated mining of digital clinical records has great potential, as it allows a direct objective measurement of amount and type of drug prescribed. A recent study has described a practical application of this methodology to describe the usage of corticosteroids in small animals (O'Neill et al., 2012b). Descriptive statistics were applied to a large dataset of over 30,000 clinical records from three small animal practices in England. The results of that study provided evidence of a significant difference in prescription patterns between practices, which may be a reflection of several factors including personal experience and preference by veterinary surgeons from different practices (O'Neill et al., 2012b). The authors concluded that this was a preliminary study that although including a relatively large number of clinical records, was unlikely to be reflective of the overall population due to the limited number of veterinary practices and veterinary surgeons involved. While these studies include often thousands of medical records it is relevant to consider that, particularly when looking at prevalence of drug usage, there remains a potential effect of practice, which should be

accounted for during analyses. It is therefore important that the number of practices is as large as possible to minimise the intrinsic bias of large veterinary practices.

1.12 Aims of the overall study

This study aims to provide an insightful investigation of the role of COX selectivity in the development of NSAID toxicity in equine clinical practice.

Biochemical investigations in Chapter 2 aim to provide evidence that COX selectivity can affect the potential development of toxic side effects in actual equine clinical patients.

Epidemiological investigations in Chapters 3 to Chapter 5 look at the impact that NSAID usage has on equine practice. This is achieved by interrogating hundreds of thousands of patients' medical records to describe how often NSAIDs are used, for which conditions, and how often toxicity is actually encountered in every day practice and also if there is evidence that administration of COX-2 selective NSAIDs carry a lower risk of side effects.

CHAPTER 2 - Effect Of Flunixin Meglumine, Phenylbutazone And Firocoxib On Cyclooxygenase Activity In The Horse

2.1 Introduction

Non-steroidal anti-inflammatory drugs (NSAIDs) are a common treatment for pain and inflammation in horses undergoing surgery. These drugs exert their effects mostly by regulating cyclooxygenase (COX) enzyme activity and subsequent tissue prostaglandin production. Two main COX isoforms have been identified: COX-1 is the constitutive form and regulates tissue blood flow in physiologic states, while COX-2 is mainly expressed during inflammatory states and its uncontrolled action is considered responsible for many of the undesired effects of the inflammatory process (Vane et al., 1998). In horses, a relationship has been described between the use of non-selective COX inhibitors, including phenylbutazone and flunixin meglumine, and development of significant gastrointestinal and renal toxicity (Hough et al., 1999; Jones et al., 2003; MacAllister et al., 1993; Snow et al., 1979). These adverse effects are the likely result of inhibition of mucosal and renal medullary perfusion by reducing COX-1 activity (Moses and Bertone, 2002). Selective COX-2 inhibitors have been evaluated in horses to ascertain whether they could reduce the risks of side toxic side effects subsequent to COX-1 inhibition (Davis et al., 2011; Doucet et al., 2008; Letendre et al., 2008; Marshall et al., 2011; Tomlinson and Blikslager, 2005).

While traditional NSAIDs, phenylbutazone and flunixin meglumine, are not selective COX inhibitors (Doucet et al., 2008; Marshall et al., 2011), coxibs, such as robenacoxib and firocoxib, have recently been evaluated in horses (Davis et al., 2011; Doucet et al., 2008; Letendre et al., 2008; Marshall et al., 2011; Tomlinson and Blikslager, 2005) and were found to have high COX selectivity ratio *in vitro* (Marshall et al., 2011). Currently,

firocoxib is the only COX-2 selective drug licensed for use in the horse and its licencing covers the alleviation of pain and inflammation associated with osteoarthritis and reduction of associated lameness in horses.

The COX selectivity of an NSAID may be vastly influenced by the assay used as well as the model adopted i.e. *in vitro* or *ex vivo* (Blain et al., 2002; Vane and Botting, 1995). Whilst earlier studies using purified enzymes *in vitro* reported a COX selectivity ratio of 1275 (Gierse et al., 2002) for deracoxib in dogs, subsequent studies performed on a whole blood model had a ratio of only 12 (McCann et al., 2004). Some studies have shown little difference in clinical effect or rate of side effects for NSAIDs with different COX selectivity *in vitro* (Borer et al., 2003) and discrepancies between *in vitro* and *ex vivo* COX inhibition (Blain et al., 2002; Giuliano and Warner, 1999). These studies demonstrate not only the importance of assessing species-specific COX selectivity, but also that evaluating *ex vivo* as well as *in vitro* models is of pivotal importance to assess the effect that these drugs have on clinical patients (Lees et al., 2004a). *In vitro* models allow determination of inhibition concentration curves, which illustrate well the effect of these drugs at different concentrations and provide useful data to predict COX-1 enzyme inhibition at a given concentration. *In vitro* data can be subsequently used to evaluate if any discrepancy exists between expected COX-1 inhibition as calculated from an *in vitro* model and the actual COX-1 inhibition obtained with the drug used at the labelled dose in an *ex vivo* model. The comparison of *in vitro* and *ex vivo* data would provide an objective insight of the reliability of *in vitro* models to assess *in vivo* COX inhibition. To the authors' knowledge published equine studies assess the effects of NSAIDs only *in vitro* or *ex vivo* in experimental animals (Beretta et al., 2005; Cuniberti et al., 2012; Davis et al., 2011) and data obtained from *ex vivo* clinical models is greatly needed.

Several studies have described the pharmacokinetic and pharmacodynamic properties of NSAIDs in horses (Coakley et al., 1999; Gerring et al., 1981; Kvaternick et al., 2007; Lees et al., 2004a; Lees and Higgins, 1985; Letendre et al., 2008; Toutain et al., 1994). For phenylbutazone at the dose of 4.4mg/kg intravenously the serum concentration producing 50% of the maximal effect (EC_{50}) was found to be 3.6±2.2μg/ml (Toutain et al., 1994). For flunixin meglumine at the dose of 1.1mg/kg intravenously the EC_{50} was 0.93±0.35μg/ml (Toutain et al., 1994). To the authors knowledge no data is available of the EC_{50} of firocoxib in horses. However, a study comparing the efficacy of phenylbutazone and firocoxib in managing naturally occurring osteoarthritis in horses found no significant difference at label doses (Doucet et al., 2008). Since *in vivo* the EC_{50} of firocoxib in clinical patients cannot be determined for ethical reasons, as a study protocol would require several serial blood samples collection, describing the serum concentration of firocoxib at 2 and 24 hours would provide an indirect rough estimation of the drug's EC_{50}. These two time points correspond to when the drug has its maximum effect and the lowest effect prior to administration of the subsequent dose respectively. Also, from a practical point of view, residual blood samples could be available as these could be collected for intra- and post-anaesthetic monitoring purposes.

These studies are conducted under controlled research conditions, where animals do not concurrently receive other medications nor undergo procedures that might interfere with drug metabolism. The effect of the concomitant use of other pharmaceuticals and concurrent undergoing of surgical procedures on drug metabolism, which is the typical scenario in clinical patients is unknown. To the best of the author's knowledge the concentration of phenylbutazone, flunixin meglumine and firocoxib administered peri-operatively in actual clinical cases has not been described before.

The aims of this chapter are to:

1. Compare the *in vitro* selectivity of a non-selective COX inhibitor such as flunixin meglumine and the COX-2 selective firocoxib
2. Investigate the effect of phenylbutazone, flunixin meglumine and firocoxib on the activity of COX enzymes in equine clinical patients receiving these drugs for clinical reasons by use of an *ex vivo* method
3. Describe the concentration of these drugs in horses undergoing elective surgery, which are also administered with other peri-operative medications.

2.2 Materials and methods

2.2.1 Evaluation of cyclooxygenase-1 inhibition *in vitro*

2.2.1.1 Animal selection

A convenience population, the teaching and blood donation herd of the Weipers Centre Equine Hospital of the University of Glasgow, was included in the study on the basis of a history of having received no NSAIDs in the previous two weeks and having residual blood available collected for reasons unrelated to the study. None of these horses had a history of systemic disease or had received any medication in the 3 months prior to the study; all horses were up to date with regard to vaccination status, had no abnormal physical examination findings (Byars and Gonda, 2015) and had a blood total protein concentration and packed cell volume (PCV) within the respective reference ranges (Staempfli and Oliver-Espinosa, 2015).

2.2.1.2 Evaluation of cyclooxygenase-1 activity

The effect of the firocoxib and flunixin meglumine on *in vitro* COX-1 activity was determined by measuring coagulation-induced thromboxane

B_2 (TXB_2) as described elsewhere (Beretta et al., 2005; Brideau et al., 2001). Briefly, whole blood (500µl) was added to polypropylene micro-centrifuge tubes containing either firocoxib or flunixin meglumine at a final concentration ranging from 0.01µM and 1000µM for a total of six dilutions for each drug in duplicate. A 500µl sample of heparinized (uncoagulated) blood was collected to serve as a negative control. Whole blood (500µl) was treated with vehicle (positive) control in triplicate. The blood was allowed to clot for one hour at 37°C before being centrifuged at 2000g for 10 minutes. Following centrifugation, 100µl of serum was added to 400µl of methanol, and the resulting solution was centrifuged at 6000g for 10 minutes. A 50µl aliquot of the supernatant was collected and diluted in 150µl of buffer from a commercially available TXB_2 enzyme immunoassay (EIA) kit[1]. This kit was used to determine the amount of TXB_2 in each sample.

Inhibition of COX-1 activity was calculated as the percentage change in TXB_2 concentration compared to the result from the negative control sample. The percentage change was plotted against the corresponding drug concentration, and the best sigmoid curve fit was obtained by use of nonlinear regression using Sigmaplot[2].

2.2.2 Evaluation of cyclooxygenase activity *ex vivo*

2.2.2.1 Animal selection, group allocation and sample collection

Horses presenting to the Weipers Centre Equine Hospital, University of Glasgow, between April and September 2012 with no history of medication including NSAIDs for a minimum of two weeks prior to admission, that were systemically healthy and scheduled to undergo an elective surgical procedure were recruited. Only horses from which sufficient blood (2.5ml)

[1] Thromboxane B_2, Express EIA kit-monoclonal, Cayman chemical Europe, Tallinn, Estonia
[2] Sigmaplot, version 11.2, Systat Software Inc., Chicago, Ill., USA

was remaining from samples obtained prior to surgery, during surgery and the day after surgery for clinical reasons (i.e. pre-, intra- and post-operative monitoring of blood parameters) were included in the study. Horses less than one year of age were excluded to minimise variability due to patient size and diet.

Horses were allocated to one of three groups based on the NSAID chosen by the attending clinician before surgery. The choice of NSAID used was independent from the purposes of the study. These were phenylbutazone[3] (4.4.mg/kg IV BID, n=6), flunixin meglumine[4] (1.1mg/kg IV BID, n=6) or firocoxib[5] (0.09mg/kg IV SID, n=6). There was no standardisation of other medications administered to the horses included in the study.

For each horse, blood samples were collected before NSAID administration (T_0), 2 hours after administration (intra-operatively, T_2) and 24 hours after the first administration (T_{24}) before a further dose of NSAID was give the morning following surgery. The timing of sample collection is represented in Figure 2.1. Residual blood samples were retrieved within one hour of collection and transported immediately to the laboratory for preparation for COX-1 and COX-2 activity determination as previously described (Beretta et al., 2005; Brideau et al., 2001).

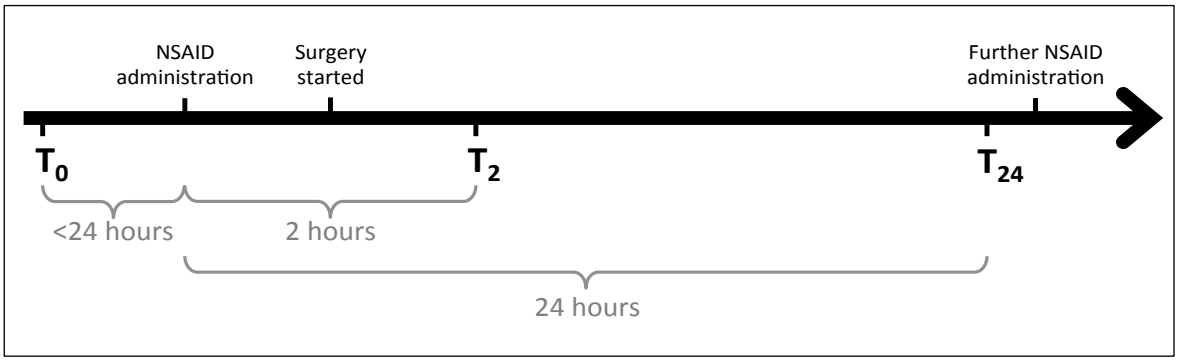

Figure 2.1: Representation of the timing of sample collection for the study evaluating cyclooxygenase activity *ex vivo*.

[3] Equipalazone, Dechra Veterinary Products Ltd, Shrewsbury, Shropshire, England

[4] Finadyne, MSD Animal Health, Milton Keynes, Buckinghamshire, England

[5] Equioxx, Merial Animal Health Ltd, Harlow, Essex, England

2.2.2.2 Determination of cyclooxygenase activity ex vivo

Measurement of *ex vivo* COX-1 activity was performed as described in section 2.2.1.2 by measuring coagulation-induced TXB_2 (Beretta et al., 2005; Brideau et al., 2001).

Measurement of *ex vivo* COX-2 activity was performed using a modification of a previously described technique (Beretta et al., 2005). Briefly, the heparinised blood (500µl) was transferred into 1.5ml polypropylene tubes and LPS (*Escherichia coli 0111:B4*) in 0.1% bovine serum albumin in PBS solution was added to a final concentration of 100µg/ml. A second aliquot of blood was used as the negative (un-stimulated) control specimen. Samples were incubated at 37°C for 24 hours. Following incubation, samples were centrifuged at 2000g for 5 minutes to harvest plasma and 100µl of plasma was added to 400µl of methanol and centrifuged at 6000g for 10 minutes. A 50µl aliquot of the supernatant was collected and diluted in 150µl of Prostaglandin E_2 (PGE_2) enzyme immunoassay (EIA) kit buffer. The amount of PGE_2 in each sample was determined using a commercially available PGE_2 EIA kit[6].

The amount of prostaglandin E metabolites (PGEM) in plasma collected at each time point was measured as an indicator of COX-2 activity using a commercially available PGEM EIA kit as previously described (Cook et al., 2009a). Briefly, all unstable metabolites of PGE_2 were converted to the stable 13,14-dyhydro-15-keto PGA_2 for quantification by means of a commercial ELISA kit [7] following manufacturer's instructions. Each measurement was performed in triplicate on a 96-well plate and the average of the three measurements was subsequently used for the analysis. The R-square of each calibration curve was >99%, indicating the assay was reliable.

---

[6] Prostaglandin E_2 EIA kit-monoclonal, Cayman Chemical Europe, Tallinn, Estonia

[7] Prostaglandin E Metabolite EIA kit-monoclonal, Cayman Chemical Europe, Tallinn, Estonia

2.2.2.3 Data analysis

The relative change in concentration of TXB_2, PGE_2 and PGEM between the baseline sample (T_0) and T_2 and T_{24} samples was calculated. The effect of time and treatment on COX-1 (TXB_2) and COX-2 (PGE_2 and PGEM) activity and drug metabolites concentration was determined by Mann-Whitney test. Bonferroni corrections were used to account for multiple comparisons. The relationship between drug concentration and COX activity overall and at each time point was determined by Spearman's rank correlation (significance set for $p<0.05$). Analysis was performed using Rv.3.0.0 software 2013[8].

2.2.3 Determination of *ex vivo* non-steroidal anti-inflammatory serum concentration

Aliquots of serum obtained at each time point were submitted to an external laboratory[9]. Flunixin meglumine, phenylbutazone and its metabolite oxyphenbutazone and firocoxib concentrations were determined by use of high performance liquid chromatography (HPLC) (Cox and Yarbrough, 2011; Higgins et al., 1987).

2.2.4 Comparison of *in vitro* and *ex vivo* cyclooxygenase inhibition

The relationship between drug concentration and COX activity overall and at each time point was determined by Spearman's rank correlation (significance set for $p<0.05$). On the basis of the drug concentrations obtained by use of HPLC, the predicted percentage inhibition of COX-1 activity was calculated from *in vitro* inhibition-concentration curves and compared with the *ex vivo* inhibition of COX-1 activity. Bland-Altman analysis was performed to determine the level of agreement between the

[8] R, version 3.0.0, R Foundation for Statistical Computing, Vienna, Austria

[9] UNIRELAB srl, Unipersonale, Rome Italy

in vitro and *ex vivo* methods of measuring inhibition of COX activity (Bland and Altman, 1986). Analysis was performed using Rv.3.0.0 software 2013.

All procedures were approved by the Ethics and Welfare Committee of the School of Veterinary Medicine at the University of Glasgow.

2.3 Results

2.3.1 Evaluation of cyclooxygenase-1 inhibition *in vitro*

2.3.1.1 Animal selection

Animals included in the *in vitro* study were two mares and two geldings; two Thoroughbreds, one Warmblood and one pony; ages were 8, 23, 19 and 28 years and each animal weighed 563kg, 534kg, 615kg and 296kg respectively.

2.3.1.2 Evaluation of cyclooxygenase-1 inhibition

The equation that best predicted the relative inhibition of COX-1 activity expressed as the change in TXB_2 concentration relative to the concentration of the drug was as follows:

$$Percentage\ inhibition = y_0 + a/[1 + e^{-e(x-x_0)/b}]$$

where y_0 is the minimum COX activity, a is the difference between the maximum and minimum COX activity, e is the base of the natural logarithm, x is the concentration of the drug, x_0 is the drug concentration required for 50% inhibition of COX activity, and b is the slope.

For flunixin meglumine, the best fit (r^2 = 0.999) of the inhibition-concentration curve was obtained by use of a 4-point sigmoid function represented by the following equation (Figure 2.2):

$$Percent\ inhibition_{flunixin} = -11.4541 + [111.509/\ (1 + e^{-(x\ -\ 18.841)/11.7442})]$$

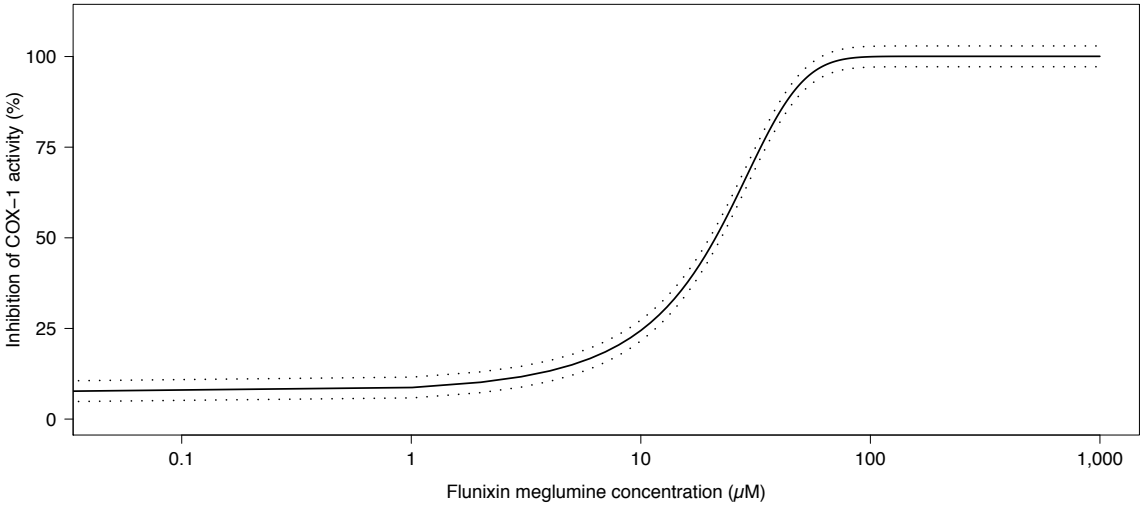

Figure 2.2: 4-point sigmoid curve representing the best fit of the inhibition-concentration curve of COX-1 activity measured as TXB_2 concentration for flunixin meglumine calculated from *in vitro* data for concentrations ranging from 0.01μM and 1000μM; dotted lines represent 95% confidence interval limits.

For firocoxib, the best fit (r^2 = 0.952) of the inhibition-concentration curve was obtained by use of a 3-point sigmoid function represented by the following equation (Figure 2.3):

$$Percent\ inhibition_{firocoxib} = \ 46.7088/(1 + e^{-(x\ -\ 6.61021)/2.6457})$$

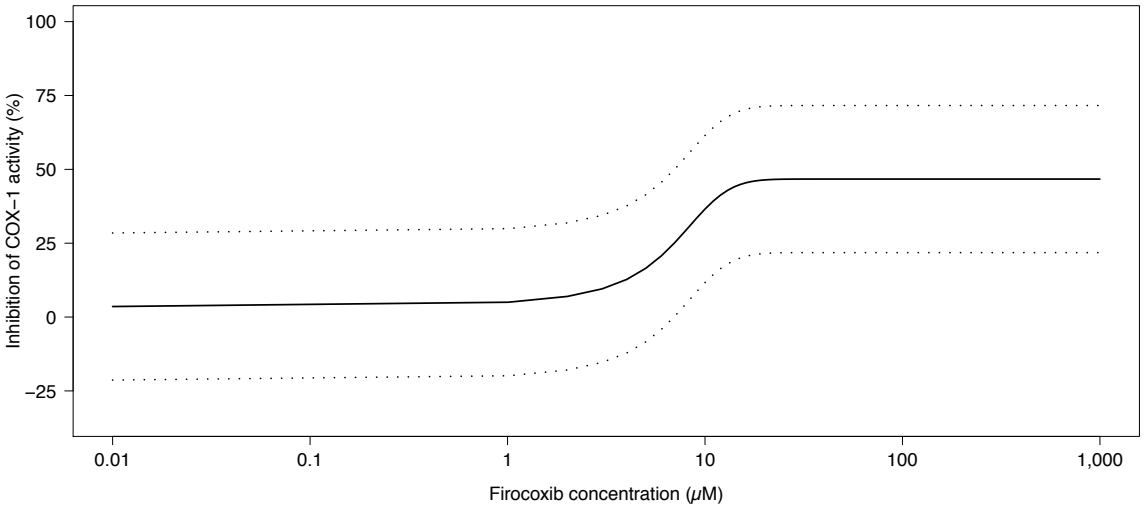

Figure 2.3: 3-point sigmoid curve representing the best fit of the inhibition-concentration curve of COX-1 activity measured as TXB_2 concentration for firocoxib calculated from *in vitro* data for concentrations ranging from 0.01μM and 1000μM; dotted lines represent 95% confidence interval limits.

2.3.2 Evaluation of cyclooxygenase activity *ex vivo*

2.3.2.1 Animal selection and group allocation

Horses in the phenylbutazone group had a median age of seven years (mean: nine years) and a median weight of 536kg (range: 260-759kg) and included two Warmbloods, one Thoroughbred and three ponies of which there were three geldings, one mare and two stallions. Horses in the flunixin meglumine group had a median age of eight years (range: 1-17 years), median weight of 588kg (range: 350-636kg) and included three Warmbloods and three Thoroughbreds (four geldings, one mare and one stallion), which underwent elective soft tissue (n=4) or orthopaedic surgical procedures (n=2). Horses in the firocoxib group had a median age of four years (range: 1-10 years), median weight of 538kg (range: 323-651kg) and included two Warmbloods, two Thoroughbreds and two ponies (five mares and one stallion), which underwent elective orthopaedic surgical procedures (n=6).

A more detailed description of horses and procedures is included in Table 2.1.

Table 2.1. Summary of the demographic information of the study population and procedures performed for horses in each group.

Group	Horse	Age	Breed	Gender	Weight	Diagnosis	Procedure
PBZ	1	5	WB	G	580	DDFT tear	Tenoscopy
	2	16	Pony	G	468	DDFT tear	Tenoscopy
	3	5	WB	F	573	OCD Hock	Arthroscopy
	4	9	Pony	S	499	-	Castration
	5	14	TB	G	759	PSD	Neurectomy fasciotomy
	6	5	Welsh B	S	260	Cryptorchid	Castration
FM	7	1	TB	S	350	-	Castration
	8	4	WB	G	625	Sarcoids	Surgery+Cryo+Chemo
	9	8	TB	F	-	GCT	Ovariectomy
	10	8	WB	G	636	Over-riding DSP	DSP resection
	11	17	TB	G	588	Echinococcosis	Cyst lavage
	12	9	WB	G	566	Suspensory Desmitis	Neurectomy
FIR	13	7	Highland	S	651	OCD	Arthroscopy
	14	1	Trakhener	F	323	MT3 Cyst	Arthroscopy
	15	4	WB	F	528	OCD	Arthroscopy
	16	1	WB	F	323	OCD	Arthroscopy
	17	4	TB	F	549	OCD	Arthroscopy
	18	10	TB	F	578	OCD	Arthroscopy

PBZ: phenylbutazone; FM: flunixin meglumine; FIR: firocoxib; WB: warmblood; TB: thoroughbred; G: gelding; F: female; S: stallion; DDFT: deep digital flexor tendon; OCD: osteocondrosis dissecans; PSD: proximal suspensory desmitis; GCT: granulosa cell tumor; DSP: dorsal spinous processes; MT3: metatarsal 3; Age in years; Weight in kilograms.

2.3.2.2 Determination of cyclooxygenase activity ex vivo

The concentrations of PGE_2 and TXB_2 varied widely among horses. Therefore, the results are expressed also as the percentage change from the baseline concentration. At T_2 and T_{24}, COX-1 activity was reduced compared to baseline in horses receiving phenylbutazone or flunixin meglumine (Fig 2.4). At T_2 and T_{24}, the relative COX-1 activity was significantly greater in horses receiving firocoxib compared to horses receiving phenylbutazone ($p=0.008$) or flunixin meglumine ($p=0.005$) (Figure 2.4). The effect on COX-2 activity was not significantly different between drugs ($p=0.5$; Figure 2.5 and Figure 2.6).

Considering the samples as a whole without dividing by time point the results of Spearman's rank analysis evidenced one significant correlation that was between phenylbutazone and absolute TXB_2 concentrations ($p=0.03$). No other significant correlations were present when considering the overall samples without dividing in time-points.

When subdividing the samples by time-point at T_2 the relative concentration of PGE_2 was significantly correlated to the concentration of oxyphenbutazone ($p=0.03$). When considering the absolute TXB_2 and PGEM concentration they were significantly correlated to flunixin meglumine concentration ($p=0.02$) and firocoxib concentration ($p=0.02$) respectively.

At the third time-point the relative phenylbutazone concentration was significantly correlated with the absolute TXB_2 and PGEM concentrations ($p=0.01$). Flunixin meglumine was significantly correlated with absolute TXB_2 ($p=0.03$) and PGEM ($p=0.04$) concentrations.

Spearman's rank analysis of all samples revealed no significant correlation between flunixin meglumine, phenylbutazone or firocoxib concentration and either TXB_2, PGE_2 or PGEM relative or absolute concentrations. At T_2, the absolute metabolite concentration TXB_2 and PGE_2 were significantly correlated to flunixin meglumine concentration ($p=0.02$) and firocoxib concentration ($p=0.02$) respectively. At the third time-point flunixin meglumine concentration was significantly correlated with absolute TXB_2 ($p=0.03$) and PGEM ($p=0.04$) concentrations.

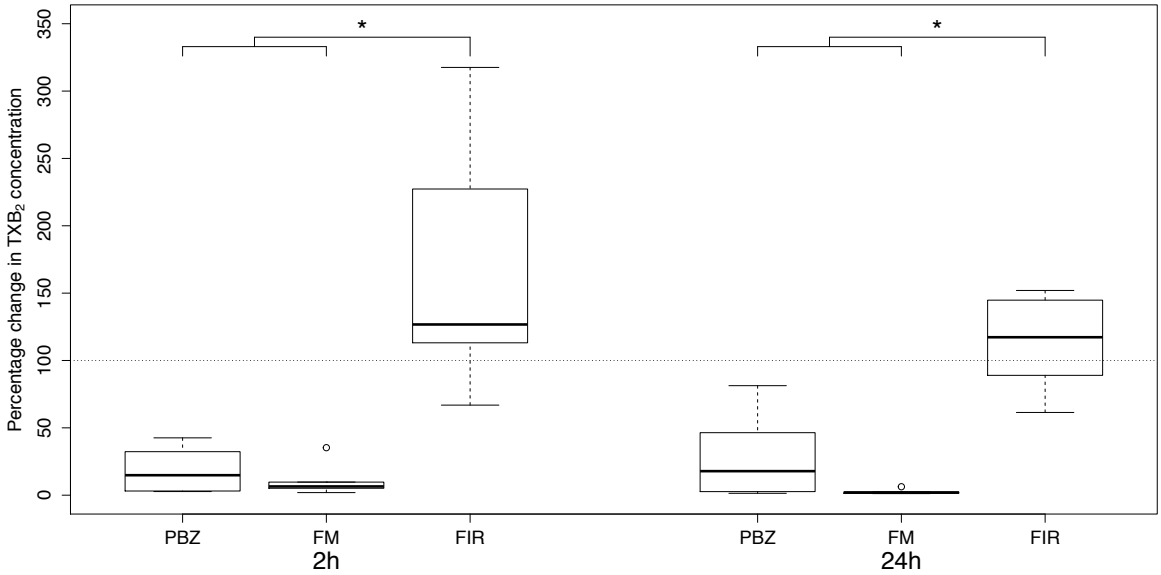

Figure 2.4: Box-and-whiskers plots of the percentage change in the amount of coagulation-induced TXB_2 representing COX-1 activity, compared with the value before NSAID administration (baseline; value set at 100% - horizontal dotted line), in horses at 2 and 24 hours after treatment with phenylbutazone (n=6), flunixin meglumine (n=6) or firocoxib (n=6).

Each box represents the second and third quartile ranges, the solid horizontal line in each box represents the median, the whiskers represents first and fourth quartile ranges, and the circles represent outlier data points. *: relative COX-1 activity differs significantly ($p<0.05$) between treatment groups. PBZ: phenylbutazone; FM: flunixin meglumine; FIR: firocoxib

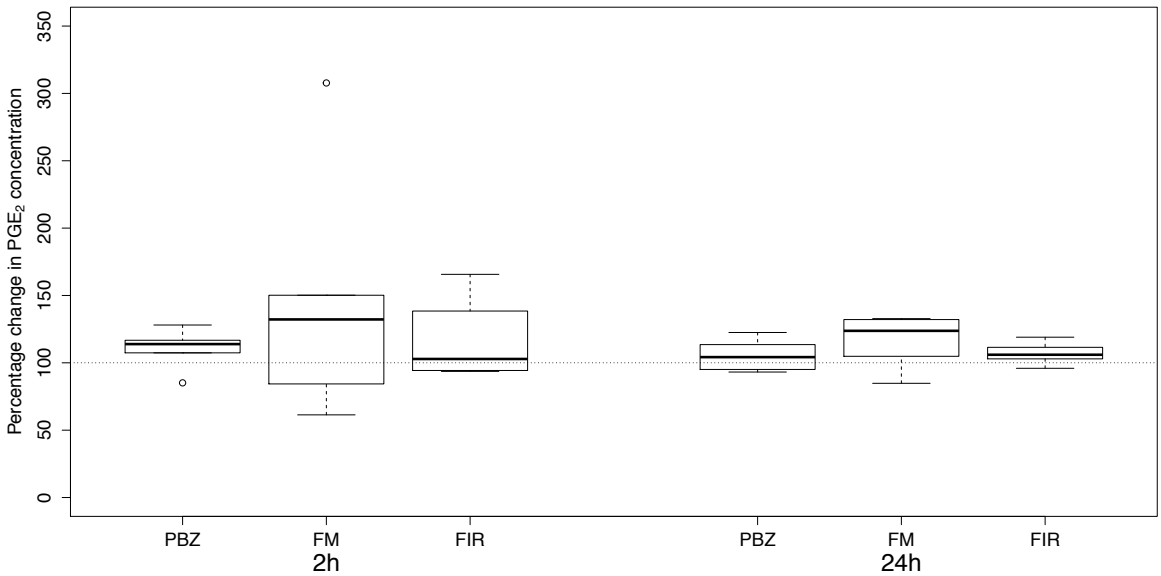

Figure 2.5: Box-and-whiskers plots of COX-2 activity expressed as the percentage change in LPS stimulated PGE_2 concentration, compared with the baseline value (set at 100% - horizontal dotted line).

PBZ: phenylbutazone; FM: flunixin meglumine; FIR: firocoxib

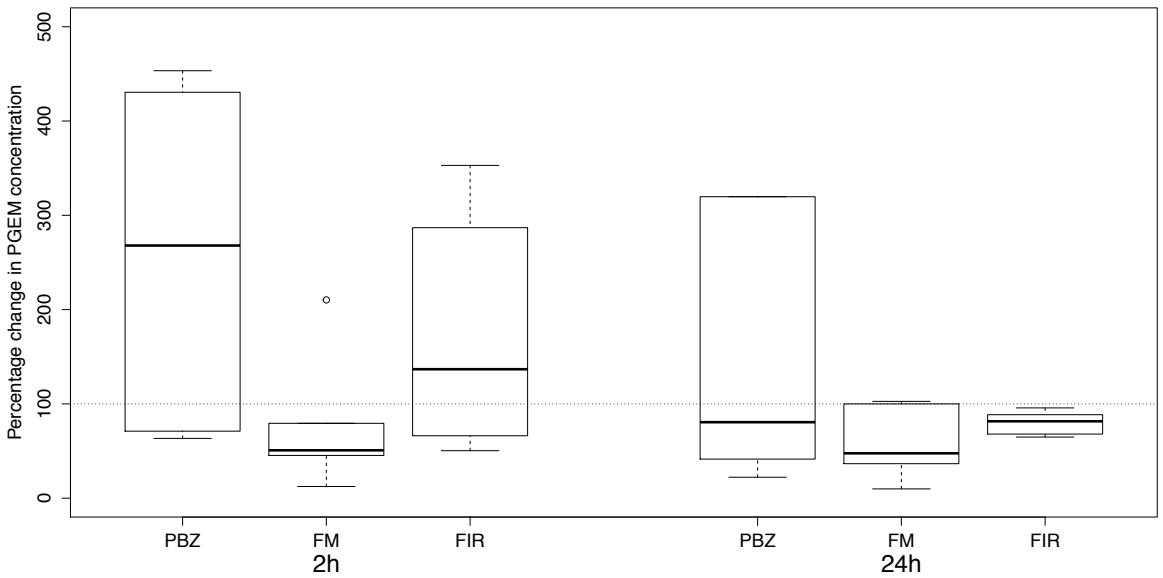

Figure 2.6: Box-and-whiskers plots of COX-2 activity expressed as the percentage change in plasma PGE metabolite concentration, compared with the baseline value (set at 100% - horizontal dotted line).
PBZ: phenylbutazone; FM: flunixin meglumine; FIR: firocoxib

2.3.3 Determination of *ex vivo* non-steroidal anti-inflammatory drug serum concentration

The concentration of phenylbutazone at T_0 was 0µg/ml for all but one horse that had a concentration of 1.30µg/mL. The median phenylbutazone concentration at the second time-point two hours after drug administration was 28.88µg/mL (mean: 29.05µg/ml; range: 21.35-37.40µg/mL). At the third time-point the median concentration of phenylbutazone was 24.02µg/mL (mean: 22.29µg/ml; range: 12.03-34.26µg/mL).

The baseline concentration of oxyphenbutazone prior to phenylbutazone administration was 0µg/ml for all but one horse that had a concentration of 0.18µg/ml. At the second time-point (2h after administration) the median oxyphenbutazone concentration was 1.19µg/mL (mean: 1.15µg/ml; range: 0.59-1.66µg/mL). At the third time-point the median concentration of oxyphenbutazone was 3.32µg/mL (mean: 3.45µg/ml; range: 2.42-4.42µg/mL).

For flunixin meglumine the baseline concentration (before drug administration) was 0µg/ml for all horses. At the second time-point (2h after administration) the median flunixin meglumine concentration was 2.55µg/mL (mean: 2.84µg/ml; range: 2.45-3.82µg/mL). At the third time-point the median concentration of flunixin meglumine was 1.29µg/mL (mean: 2.60µg/ml; range: 0.18-8.74µg/mL).

For firocoxib the baseline concentration (before administration) was 0ng/ml for all horses. At the second time-point (2h after administration) the median firocoxib concentration was 53.00ng/mL (mean: 65.17µg/ml; range: 30.8-134.8ng/mL). At the third time-point the median concentration of firocoxib was 35.4µg/mL (mean: 33.33µg/ml; range: 21.6-40.7µg/mL).

These results are summarized in Figure 2.7, Figure 2.8 and Figure 2.9.

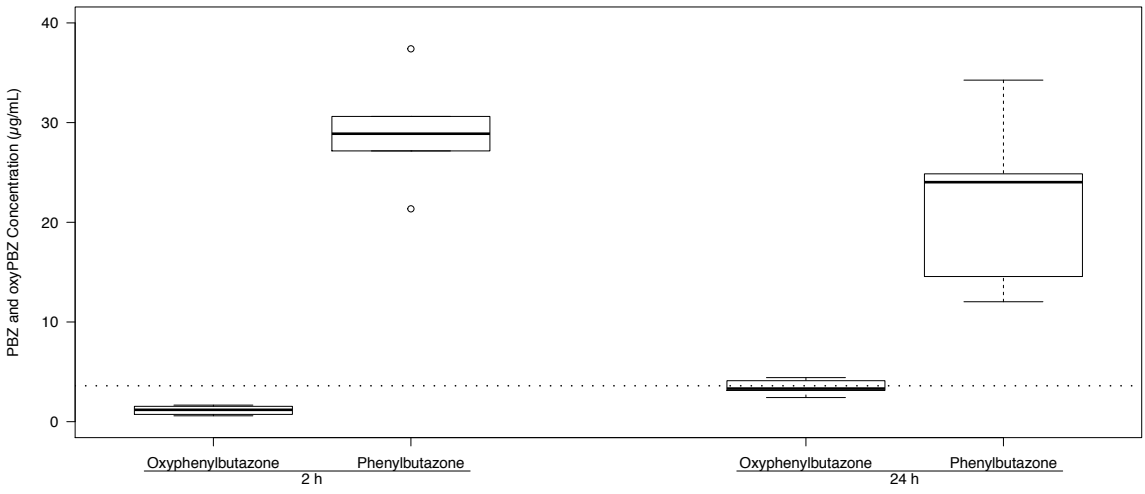

Figure 2.7: Box-whiskers plot depicting the concentration of phenylbutazone and its metabolite oxyphenbutazone after 2 and 24 hours from intravenous administration of phenylbutazone at 4.4mg/kg BID. The horizontal dotted line represents the serum concentration of phenylbutazone at EC_{50} (Toutain et al., 1994).

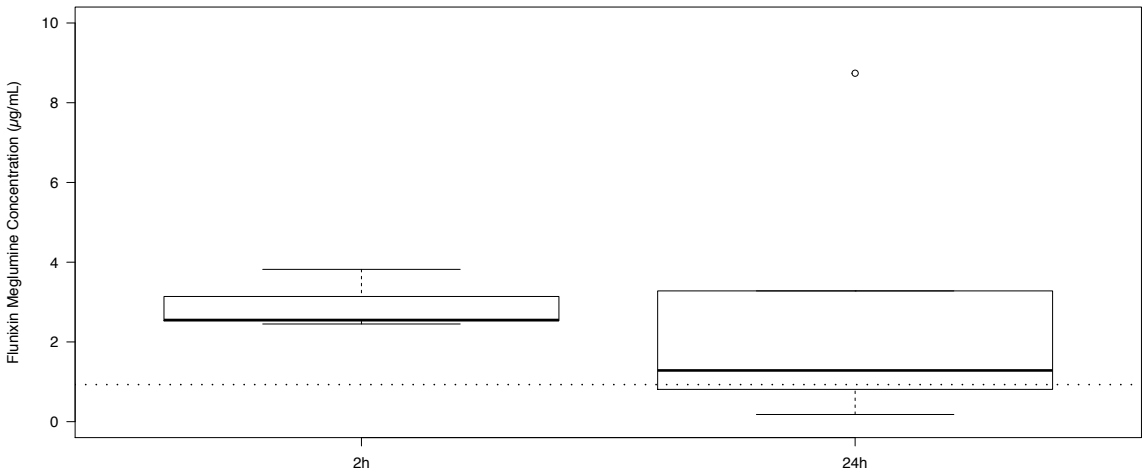

Figure 2.8: Box-whiskers plot depicting the concentration of flunixin meglumine after 2 and 24 hours from intravenous administration at the dose of 1.1mg/kg BID. The horizontal dotted line represents the serum concentration of flunixin meglumine at EC_{50} (Toutain et al., 1994)**.**

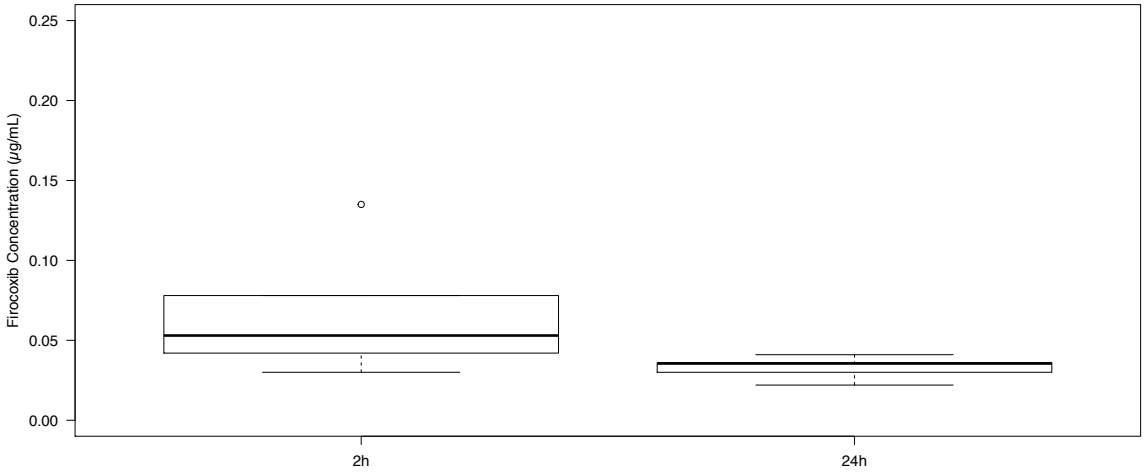

Figure 2.9: Box-whiskers plot depicting the concentration of firocoxib after 2 and 24 hours from intravenous administration at 0.1mg/kg SID.

2.3.4 Comparison of *in vitro* and *ex vivo* cyclooxygenase inhibition

In vitro and *ex vivo* COX-1 inhibition was significantly different for both flunixin meglumine ($p<0.001$, Figure 2.10) and firocoxib ($p=0.04$, Figure 2.11).

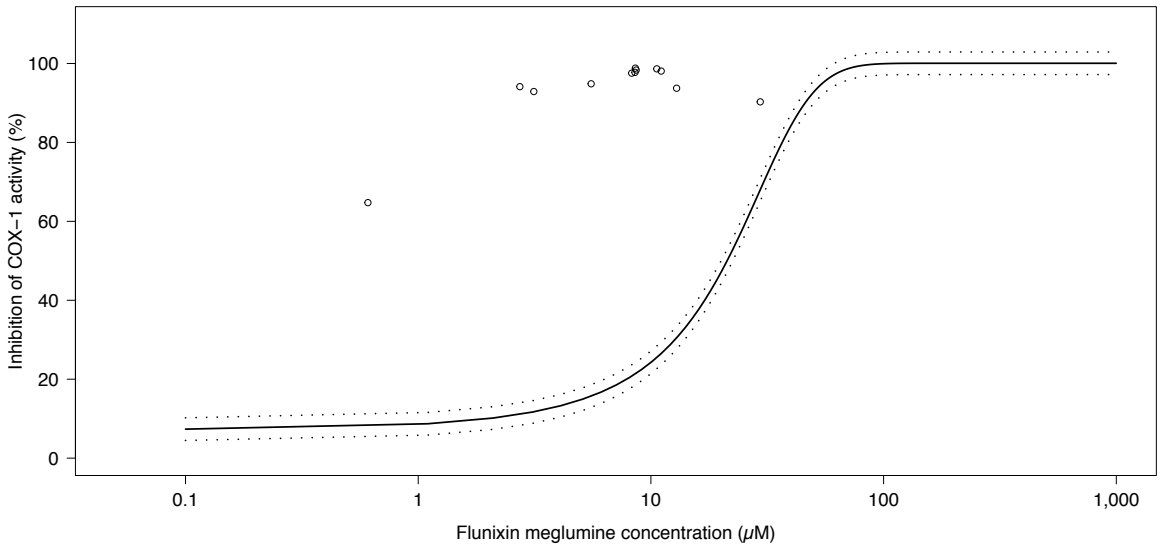

Figure 2.10: Comparison of flunixin meglumine concentration and *in vitro* and *ex vivo* inhibition of COX-1 activity. The sigmoid curve represents the calculated *in vitro* concentration-inhibition curve and the dotted lines represent the 95% CIs. Circles represent *ex vivo* measurements of inhibition of COX-1 activity at known drug concentrations. Notice that inhibition of COX-1 activity is higher *ex vivo* than *in vitro* (p<0.001).

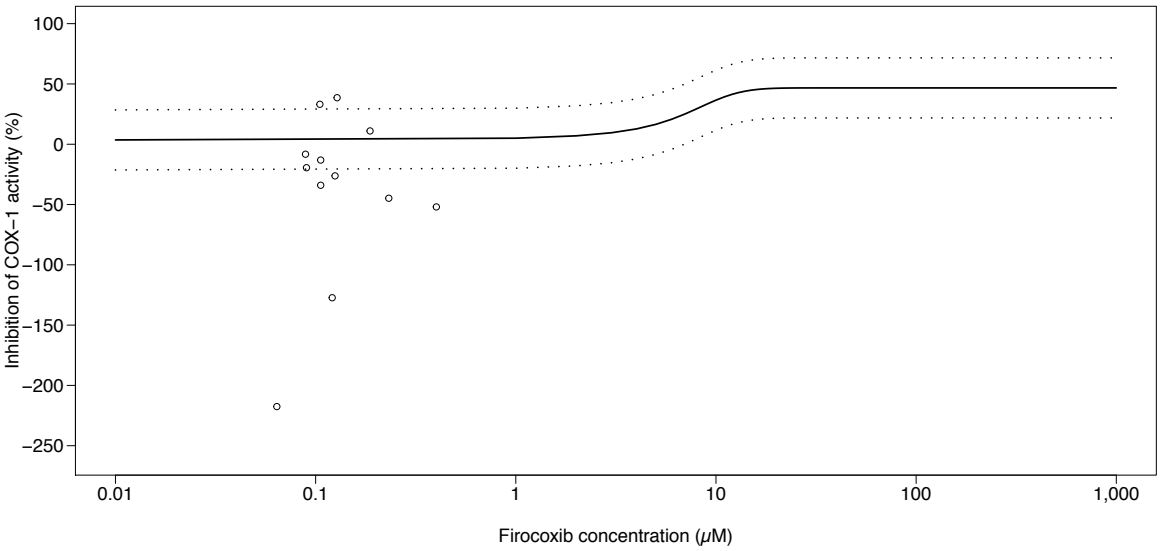

Figure 2.11: Comparison of firocoxib concentration and *in vitro* and *ex vivo* inhibition of COX-1 activity. The sigmoid curve represents the calculated *in vitro* concentration-inhibition curve and the dotted lines represent the 95% CIs. Circles represent *ex vivo* measurements of inhibition of COX-1 activity at known drug concentrations. Notice that in some cases the *ex vivo* inhibition of COX-1 activity corresponds to inhibition expected from *in vitro* data, whereas in the remaining cases, COX-1 activity is not inhibited by firocoxib. Overall data obtained *in vitro* and *ex vivo* were significantly different (p=0.04).

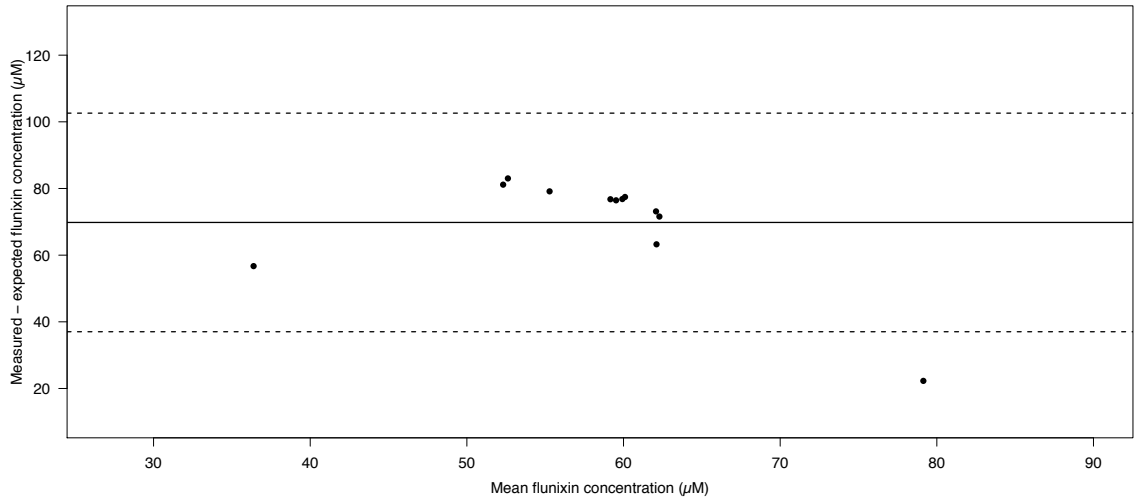

Figure 2.12: Bland-Altman plots of *in vitro* and *ex vivo* COX-1 activity after administration of flunixin meglumine to 6 horses. The mean bias (solid line) is high and 95% CIs (dotted lines) is >0% which suggests poor agreement. Flunixin meglumine concentration was determined for *ex vivo* samples collected from 6 horses and calculated from the inhibition concentration curve determined *in vitro*.

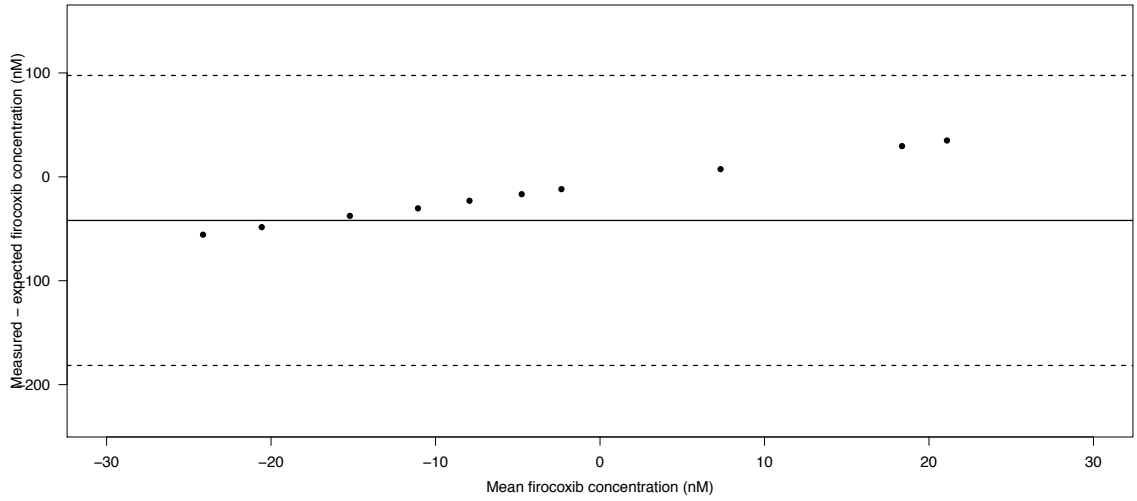

Figure 2.13: Bland-Altman plots of *in vitro* and *ex vivo* COX-1 activity after administration of firocoxib to 6 horses. The mean 95% CIs (dotted lines) is extremely wide, which suggests that the *in vitro* curve for inhibition of COX-1 activity is not useful for predicting *ex vivo* inhibition of COX-1 activity. Firocoxib concentration was determined for *ex vivo* samples collected from 6 horses and calculated from the inhibition concentration curve determined *in vitro*.

Bland-Altman analysis revealed a mean difference between *in vitro* and *ex vivo* COX-1 activity. The mean difference was 69.8% (95% CI, 37.0% to 102.6%) for flunixin meglumine (Figure 2.12) and –42.0% (95% CI, –181.5% to 97.6%) for firocoxib (Figure 2.13).

2.4 Discussion and conclusions

The investigations in this chapter provide a biochemical perspective of the potential for toxicity of NSAIDs in horses. Clinically, most severe side effects from their use mainly involve the gastrointestinal and urinary systems as these are more susceptible to changes in physiologic COX activity, which might affect mucosal blood flow (Cohen et al., 1995; Jones et al., 2003; Read, 1983; Warner et al., 1999).

This chapter determined the effect of flunixin meglumine and firocoxib on COX activity *in vitro* as well as that of flunixin meglumine, phenylbutazone and firocoxib *ex vivo* in horses undergoing elective surgery. Comparison of *in vitro* data of COX-1 inhibition and *ex vivo* data highlights the importance in following up *in vitro* research with studies on *ex vivo* models, as highlighted in other species, as well as for other NSAIDs (Blain et al., 2002; Gierse et al., 2002; Giuliano and Warner, 1999).

Section 2.3.1 describes the inhibition of COX-1 activity by flunixin meglumine and firocoxib in horse's blood *in vitro*. These data were obtained to calculate inhibition-concentration curves used to determine whether any significant discrepancy was present between *in vitro* and *ex vivo* COX-1 activity for the two drugs. The calculation of COX-2 activity *in vitro* was not performed. Assessing the effect on COX-2 activity would have been useful to calculate the IC_{50} or IC_{80} of these drugs (Beretta et al., 2005). However, this was not the goal of the study. This study was primarily to evaluate the potential for toxic side effects of these drugs arising from the excessive inhibition of COX-1. Further only IC_{50} and IC_{80} were also not determined to reduce costs.

Similarly no *in vitro* study on phenylbutazone was performed because of the similar COX inhibitory activity to flunixin meglumine, as illustrated in section 2.3.2 and to reduce the costs associated with the analysis.

The results of the study in section 2.2.2 show that firocoxib administration induced no significant inhibition of COX-1 activity in clinical patients, while flunixin meglumine and phenylbutazone administration induced profound inhibition of COX-1 activity. This supports the findings of another study (Cook et al., 2009a) in which investigators found less inhibition of COX-1 activity by firocoxib than by flunixin meglumine in small intestinal ischemia-reperfusion injury. No significant change in indicators of COX-2 activity, LPS-stimulated PGE_2 concentration, and PGEM concentration after surgery in either treatment group was present. Given that this was a clinical study, ethically it was not possible to include a control group without NSAID administration to quantify the effect of surgery on COX-2 activity. However, our findings indicated that the effect of firocoxib on COX-2 activity was comparable to that of phenylbutazone and flunixin meglumine and all drugs prevented an increase in COX-2 activity in the 24 hours after surgery.

Whether firocoxib would have reduced COX-2 activity also in a population of clinical patients undergoing more invasive procedures remains undetermined. Horses in each group of this study were comparable for age, gender, weight and breed characteristics and the only difference was the type of elective procedure that each horse was undergoing. However, the effect of the type of procedure was not tested with formal statistics because the group sizes were so small that the likelihood of failing to identify a true significant difference would have been high. A limitation of the present study was the inability to match surgical procedures between groups, given that blood samples from clinical equine patients were used. Group allocation of horses was chosen independently from the scope of the study and no clinician was directly involved in performing the study. Veterinary surgeons chose which NSAID to administer independently, using their clinical judgment and following label recommendations.

All horses had no evidence of severe disease affecting more than one body system, and no horse included in the study had abnormal results for physical examination or hematologic evaluation. All procedures performed were considered elective. The commercial preparation of firocoxib, is licensed in the United Kingdom for the alleviation of pain and inflammation associated with osteoarthritis and alleviation of lameness in horses. Because of the narrow spectrum of conditions licensed for treatment with firocoxib, only horses with evidence of joint disease undergoing arthroscopy were included in the firocoxib group. This procedure is minimally invasive and unlikely to induce extensive systemic inflammation. In comparison, the flunixin meglumine group included horses undergoing soft tissue procedures that could potentially have resulted in greater COX stimulation than for arthroscopy (Jacobsen et al., 2009). However, there was no significant difference in preoperative prostanoid concentrations between groups, and postoperative prostanoid concentrations were not increased in the flunixin meglumine group. A horse in the flunixin meglumine group underwent exploratory laparotomy to evaluate a hepatic mass that was confirmed to be a hydatid cyst, which likely constituted an incidental finding (Barton, 2010). A study conducted to examine the effect of surgery on the early inflammatory response (< 24 hours) found a significant difference in serum amyloid A concentrations between procedures with minimal tissue injury (e.g., arthroscopies) and procedures with intermediate tissue injury (e.g., laryngeal surgeries and castrations), but no significant difference in serum amyloid A concentrations was found between surgeries involving minimal and major tissue injury (Jacobsen et al., 2009). In addition, investigators in that same study found no significant effect of tissue injury on other variables of inflammation, including white blood cell count and serum iron concentration (Jacobsen et al., 2009). These findings suggest that although there may be a difference in the degree of inflammation stimulated by surgery, it is minimal during the first 24 hours after surgery. In the study reported here, horses had no abnormal physical examination

findings or results for hematologic evaluation before surgery and did not develop complications after surgery. Therefore, this difference between groups was considered a minor issue. Further, when a more invasive procedure, such as midline laparotomy, was performed in healthy research horses, the effect of firocoxib on COX activity was not significantly different from that of flunixin meglumine (Cook et al., 2009a). In light of these findings and the limitations imposed by the clinical study design, enrolling patients undergoing procedures that involved mild to moderate tissue damage was considered acceptable.

Another limitation of the current study is that drug concentration was tested only for the NSAID used in the horse's group. In theory, horses in the flunixin meglumine and firocoxib group could have received phenylbutazone shortly prior to admission. One horse in the phenylbutazone group had low but detectable serum phenylbutazone and oxyphenbutazone concentration suggestive of administration few days before presentation to the hospital, in face of a history of no drug being administered. Measuring concentration of each NSAID in each animal at the baseline sample was not performed as it would have been costly and at the time the samples were collected it appeared reasonable to rely on history collection to determine if a horse matched the inclusion criteria of no recent NSAID administration. It is also relevant to highlight that the concentration of phenylbutazone was 10-fold smaller than the lowest concentration of the drug at the third time point. Therefore it was decided that data from this horse could be included in the study.

The present study was not designed to compare analgesic effects of these drugs. Despite the short half-life of flunixin meglumine (2.1 to 4.2 hours) (Pellegrini-Masini et al., 2004; Toutain et al., 1994) this drug can significantly reduce tissue production of prostaglandins in exudates for up to 24 hours, which is well after the elimination of flunixin meglumine from the blood stream (Higgins et al., 1987; Toutain et al., 1994). The

persistence of firocoxib at a peripheral site of inflammation remains undetermined. However, the long half-life (29 to 31 hours) after once-daily administration should guarantee persistence of the inhibition of COX activity in peripheral tissues throughout the course of treatment (Cox et al., 2012; Kvaternick et al., 2007). Evidence suggests that firocoxib is effective for controlling orthopaedic pain in both experimental and clinical settings (McCann et al., 2002; Orsini et al., 2012). None of the clinical patients in the firocoxib group required further administration of analgesic by attending clinicians, who were not involved in the study, during the first 24 hours after surgery (i.e., after a single dose of firocoxib). The author's subjective clinical impression was that firocoxib might offer adequate analgesia for minimally invasive elective surgical procedures such as arthroscopy in horses, but further studies would be required to enable objective assessment of the efficacy of firocoxib as a perioperative analgesic. Whether firocoxib would control inflammation and the associated pain from more severe conditions, remains undetermined and should be evaluated in future studies in patients with septic synovitis, colic and endotoxaemia. Results for experiments in one study revealed that firocoxib was as effective as flunixin meglumine for managing signs of pain in healthy horses undergoing ventral midline coeliotomy and an experimental model of jejunal ischemia-reperfusion injury without enterectomy (Cook et al., 2009a). In that study, firocoxib was also as effective as flunixin meglumine at inhibiting prostaglandin production driven by ischemia-reperfusion injury (Cook et al., 2009a). Whether firocoxib would control inflammation and the associated pain from more severe naturally occurring disease or colic remains undetermined and should be evaluated in ad hoc studies.

The findings of this study supported the hypothesis that firocoxib has *ex vivo* COX-1–sparing effects. However, further studies with more homogeneous groups would yield more conclusive evidence. Firocoxib did not inhibit *ex vivo* COX-1 activity in equine patients that underwent

elective surgery and could potentially offer an alternative for the treatment of pain and inflammation in patients for which the use of non-selective NSAIDs is contraindicated. Further studies are required to evaluate the use of firocoxib in patients with clinical conditions for which its use is not currently licensed, including intestinal ischemia, right dorsal colitis, or pre-renal or intrinsic acute renal failure, before it can be recommended for use in these conditions.

The data combining the peri-operative serum concentrations of phenylbutazone, flunixin meglumine and firocoxib to COX activity in actual equine clinical cases undergoing elective surgery is also novel. As drugs such as anaesthetics, intravenous fluids and antimicrobials concurrently administered in surgery might share metabolic pathways with NSAIDs, the way pharmacodynamic and pharmacokinetic data obtained in research settings relate to real-life clinical scenarios remains undetermined. The findings in this study show that the drug concentration for phenylbutazone and flunixin meglumine at two hours is much higher than the expected EC_{50} (Toutain et al., 1994) and that confirms the expectation that these drugs used at the label dose maintain therapeutic concentrations for at least two hours. At 24 hours the concentration of these drugs was generally still above EC_{50}, with the exception of two cases where the concentration of flunixin meglumine had dropped below the EC_{50} threshold of 0.93μg/ml for this drug. The reason for this finding in these two horses remains unclear. It is possible that another drug was administered in place of flunixin meglumine for the second dose. While there is no record of this happening, this occurrence cannot be entirely discounted. Alternatively, it is also possible that in some clinical cases flunixin meglumine undergoes faster metabolism and clearance than expected. This finding requires further evaluation as a better understanding of drug clearance in these cases might affect the dose regimen administered and would ultimately enhance drug efficacy and patient welfare. At 24 hours, therefore after the second dose given at 12

hours after the initial dose, phenylbutazone and flunixin meglumine concentrations were generally still similar to the concentration at two hours. The higher variability at 24 hours is likely a reflection of the variable time the third sample was obtained. Sample collection was unrelated to the study and entirely clinician dependent. Horses were included if samples were available at the three time-points. The first sample was collected prior to NSAID administration and the second invariably two hours after NSAID administration (eg. during surgery, during anaesthesia monitoring). As surgery started at different times of the day (between 8am and 3pm) and it was hospital practice that from the day following surgery drugs for twice daily administration (such as phenylbutazone and flunixin meglumine) were to be given at ~8am and 8pm, the "24 hour" sample could have been obtained anywhere between 17 and 24 hours from the first NSAID administration. This variability might explain greater variability at the third time-point. While this was a shortcoming of the study it was not possible for the investigators to control drug administration or sample collection on real clinical cases. None of the horses was systemically ill following surgery and all samples were obtained for post-operative monitoring. Inclusion of the actual time between drug administration and third sample could also have been recorded to be included in the analysis, but addition of this variable would have required a much larger group size. Ultimately the overall aim of the study was to compare the effect of these drugs on COX activity and the data collected was adequate to achieve this goal. Extending this to a larger number of cases would have been economically beyond the funds available for this study.

The lower concentration of firocoxib at 24 hours is not unexpected as this drug is administered only once a day and no additional dose was given before the last time-point. To the author's knowledge no study exists to determine the EC_{50} of firocoxib in horses. However a study comparing phenylbutazone (4.4mg/kg PO BID) and firocoxib (0.1mg/kg PO SID) found

similar clinical efficacy between the two drugs to manage pain associated with osteoarthritis in horses (Doucet et al., 2008). Although the present study did not look at the clinical efficacy of these drugs the doses used were comparable to that study. A translational comparison between that and the present study would suggest that the EC_{50} of firocoxib is at least as low as the median firocoxib concentration of 53µg/ml. However, this statement is simply speculative and further studies on firocoxib efficacy are required. Significant correlations between indicators of COX activity and flunixin meglumine concentration were identified at the 2-hour and 24-hour time points, and between indicators of COX activity and firocoxib concentration at the 2-hour time point only. The correlation was present only in a few of the combinations tested and this was likely a result of the effect of multiple factors including short metabolite half-life, difference in drug half-life between groups, and individual patient differences. It is possible that the lack of recognition of correlation in some cases was a consequence of the heterogeneous small group sizes. Group size was determined based on the predicted difference in the drugs COX selectivity. Involving more horses might allow detection of other significant relationships between drug and metabolites concentrations and enzyme activity, disease states and age or breed differences and warrants future investigations.

Section 2.3.4 describes the relationship between *in vitro* COX inhibition and *ex vivo* inhibition in horses. Available *in vitro* data indicate an approximately 265-fold difference in selectivity between firocoxib and non-selective COX inhibitors (McCann et al., 2002). Drug concentrations in the present study were in the range expected on the basis of pharmacokinetic parameters reported for these drugs (Letendre et al., 2008; Toutain et al., 1994). Pharmacokinetic studies are performed on healthy animals and the applicability of these parameters to clinical patients is debatable. Patients often have serious chronic or acute disease or receive multiple drugs (e.g., anaesthetics, antimicrobials, or

prokinetics) simultaneously. In the study reported here, all horses received antimicrobials peri-operatively and anaesthetics (general anaesthesia). These could potentially have interfered with metabolism and efficacy of the drugs of interest. No difference was expected between the predicted drug concentrations calculated from the pharmacokinetic parameters and the actual drug concentrations, considering that these patients had no abnormal findings for physical examination or results for hematologic evaluation. This would suggest that pharmacokinetic parameters for these NSAIDs determined in experimental animals are applicable to clinical patients undergoing elective surgery.

Studies conducted to examine the inhibitory effects of NSAIDs on COX in horses are based on *in vitro* methods (Beretta et al., 2005; Davis et al., 2011). In the present study, we examined the ability of *in vitro* methods to predict *ex vivo* inhibition of COX-1 activity by flunixin meglumine and firocoxib in horses. The function best fitting the in-vitro concentration-inhibition curve was used to predict the relative inhibition expected for the actual drug concentrations measured by use of HPLC in the samples obtained *ex vivo*. Results of Bland-Altman analysis revealed a high bias, particularly for flunixin meglumine (95% CIs for flunixin meglumine were both > 0 and indicated that *ex vivo* inhibition of COX activity was more efficient than *in vitro* inhibition), which suggested very poor agreement between the *ex vivo* and *in vivo* methods (Bland and Altman, 1986). The wide 95% CIs (> 100%) for firocoxib suggested that estimation of *ex vivo* inhibition of COX-1 activity by use of *in vitro* data was highly variable. Other authors have described the manner by which *in vitro* data on inhibition of COX activity in humans differs between NSAIDs (Blain et al., 2002). Investigators of that study found that the overall value of *in vitro* assays to predict *ex vivo* inhibition of COX activity differed among drugs: the *ex vivo* effect of diclofenac was reliably predicted *in vitro*, whereas *ex vivo* and *in vitro* inhibition differed significantly for ibuprofen on COX-2 activity and meloxicam on COX-1 activity (Blain et al., 2002). Investigators

of another study found that eltenac was preferentially selective for COX-2 *in vitro* but not *ex vivo* in horses (Cuniberti et al., 2012). In veterinary species, studies have detected significant differences in the *in vitro* and *ex vivo* COX selectivity ratio (concentration required to inhibit enzyme activity by 50%) of NSAIDs (Cuniberti et al., 2012; Schmid et al., 2010). Calculation of the *in vitro* COX selectivity ratio has been reported previously and was not an objective of the present study; calculation of an *ex vivo* COX selectivity ratio is not possible in clinical patients. The significant difference between calculated and actual measurements in COX-1 activity further supports the need to assess NSAID efficacy *ex vivo* as well as *in vitro* in horses. However, it is also possible that variability in procedures performed in the flunixin meglumine group could have caused variable degrees of inflammation and might have contributed to the bias to some extent. In addition, the *in vitro* and *ex vivo* experiments were conducted in separate populations of horses. However, the comparison of *in vitro* and *ex vivo* methods warrants further investigation and comparison for COX-2.

In conclusion firocoxib behaves as a selective COX-2 inhibitor *ex vivo*. This might provide the prospect of a safer profile for this drug when compared with flunixin meglumine and phenylbutazone and might provide an alternative for the treatment of inflammation in patients for which the use of non-selective NSAIDs is contraindicated. Further studies are required to evaluate the use of firocoxib in clinical cases for which its use is not currently licensed, including intestinal ischaemia, right dorsal colitis or pre-renal or intrinsic acute renal failure before it can be recommended for use in these conditions.

While the safer profile of firocoxib compared to non-selective COX inhibitors has been demonstrated in this chapter the actual clinical relevance of this information needs to be assessed further. A different approach is necessary to describe how frequent NSAID toxicity is and if horses receiving non-selective COX inhibitors are more at risk of

developing clinical signs of toxicity compared to those receiving COX-2 selective NSAIDs. This could be performed by assessing the clinical records of a large population of equine patients to identify the prevalence of toxicity, compare different NSAIDs and account for usage of other drugs and comorbidities (Chapter 5). Such type of analysis is unknown on a large scale in equine medicine and therefore the technique would require to be thoroughly validated (Chapter 3).

A study looking at the overall effect on the population from the usage of NSAIDs will provide a different perspective on severity and frequency of toxicity in the horse population.

CHAPTER 3 - Text Mining Big Data From Equine Medical Practice

3.1 Construction of a large equine electronic medical records database

3.1.1 Introduction

The number of projects that have been developed in the last few years to collect data from veterinary practice to look at large EMRs datasets both retrospectively as well as prospectively is on the increase (Jones et al., 2014; Lam et al., 2007b, 2007c; Mattin et al., 2014; O'Neill et al., 2012a, 2012b, 2013; Oswald et al., 2010; Radford et al., 2011). Much of these data are obtained from first opinion veterinary practice for disease surveillance but also to evaluate disease prevalence, risk factors and inform survival analysis. Those that obtain data prospectively are mostly focused on small animal practice and require variable commitment to produce reliable data as a conscious effort is required by veterinary surgeons to classify clinical cases for further analysis. In the case of SAVSNET (Small Animal Veterinary Surveillance Network), an initiative by the British Small Animal Veterinary Association and the University of Liverpool, a point and click application allows a reasonably quick classification by syndrome/body system and detailed information is only obtained for a random subsample of cases (Jones et al., 2014; Radford et al., 2011). With this system the effort is minimal for the operator, but as only a small subsample of cases (10%) are classified in detail the potential to draw conclusions, that may be non-representative of the wider dataset or population, is significant. The VetCompass project run by the Royal Veterinary College, is based on the use of a predetermined coding system, VeNom (Veterinary Nomenclature), which allows precise classification of most conditions (Mattin et al., 2014; O'Neill et al., 2012a). The VeNom system contains several hundreds of possible codes, which requires

considerable compliance from clinicians who need to learn to use the VeNom coding system in order to provide accurate and complete data. This might induce errors as it is foreseeable that clinicians might learn to use a set of the most commonly used terms and not commit to use the most appropriate code in the nomenclature for each case or not classify conditions that they see less commonly as they might not have the inclination to look up the most appropriate code. The use of a restricted coding system might limit the freedom of expression of the clinician and should probably be combined with free-text mining techniques (Heinze et al. 2001). Whilst providing a way of validation of a coding system, free text mining would also be a complementary and/or supplementary alternative for data extraction to semi-structured systems such as SAVSNET and VeNom. These systems rely on veterinary practitioners to be committed in learning these coding systems in order to use the most appropriate term and a validation process would allow understanding how classification by the coding system fits the clinical data described in the text.

Free-text mining techniques are suitable for analysis of prospective as well as retrospective data as they do not require any specific co-operation by clinicians other than the completion of the EMR for each of their patients (Lam et al., 2007c). This is usually done for management and billing reasons so the data used are what would be produced by each practice regardless of involvement in a research study. On the other hand, data analysis becomes more laborious as the data are less structured and come in different forms from different practices, making the combination of data from multiple practices cumbersome.

Data for this study originates from two main areas: United Kingdom and North America (United States of America and Canada). Each dataset was obtained retrospectively using two different strategies from each main continental area. This section of this chapter describes the two strategies

used to create a multi-centre equine EMR database and compares the data obtained by each strategy.

3.1.2 Materials and methods

3.1.2.1 Strategy in the United Kingdom

A total of 86 veterinary first-opinion equine practices based in the United Kingdom were contacted. For each practice the goal was to get in touch with at least one of the partners to discuss and clarify the aims of study and details concerning client confidentiality and data anonymisation. A single partner (representing the views of his or her co-partners) was targeted as the ultimate decision-makers as to whether their practice could take part in the study.

In all cases a leaflet to briefly explain the aim of the study was sent by email along with a draft of the confidentiality agreement (Appendix 3.1). Veterinary surgeons of local Scottish equine first-opinion practices were also contacted in person at a local veterinary continuing professional development (CPD) meeting organised by the Weipers Centre Equine Hospital at the University of Glasgow in April 2012. For non-local veterinary practices contact details were obtained from each practice's web-site, the initial contact was made with the administrative staff of the practice (both by email and phone), who were relied upon to forward the message to one of the partners (previously selected from their web-site).

Of the 86 practices that were approached, 51 were a convenience sample selected from the list of practices that had referred at least one case to the Weipers Centre Equine Hospital in the previous decade. Also 35 were members of an established UK-based group of equine veterinary practices (GEP) spread throughout the country committed to work together to achieve the highest standard of veterinary care and to provide continuing

professional education to their members. Member practices were initially approached by the GEP group director and data was subsequently obtained directly from those member practices that agreed to participate. All practices willing to participate were subsequently included in the study.

Of the 51 practices approached directly, a partner could be contacted directly in 23 practices. For non-replying practices it is not known whether there was a lack of interest from the partner in participating or whether the message ever reached the partners (the administrative staff might have not passed the message on as requested).

For those 12 practices that agreed to participate, after the initial contact, subsequent communication was with a member of the administrative staff designated by the partner, who would then help to extract data from the practice management software system. Instructions on how to extract the information in a format suitable to the aims of the study were obtained directly from the PMSS provider of each practice. Where the software could not produce anonymous data, instruction on how to substitute patient details with a unique numeric anonym using Microsoft Access[10] was provided to the practice (Appendix 3.2). Once each dataset was created, it was then shared with our research group via a free web-based file hosting service.

The minimum inclusion criteria for the database from the United Kingdom were as follows:

- A column with an anonymous unique identifier for each individual animal was required in order to track EMRs for each individual animal throughout the dataset over time.

[10] Microsoft, Thames Valley Park, Reading, Berkshire, U.K. RG6 1WG

- A date of data entry for each EMR was required to assess temporal relationships, for example between drug administration and certain clinical signs that could indicate toxicity.
- Clinical notes (including free text notes and laboratory results where available).

Other desirable, but not essential, information included:

- Demographic or signalment information (including anonymous id, species, breed, gender, date of birth).
- Codes to differentiate first opinion and referral work for practices where both were present.
- Details of the quantity as well as type of drug administered or dispensed.

3.1.2.2 Strategy in North America

Data collection in North America was performed by approaching PMSS providers directly. Initially three main companies were approached to obtain data, only one of which was open to discuss participation.

The standard contract of this company with its client practices specified that the company had the right to use/sell EMRs information for other purposes (marketing and research) as long as patient/client details remained anonymous. This company agreed to anonymise, clean and format the data in order to be compatible with our research. It was agreed that the dataset should contain at least 150,000 equids from North America and that this should represent at least eight equine practices with a relevant first opinion workload with good geographical spread through the North American continent.

The minimum and desirable inclusion criteria for the North American database were the same required for the data from the United Kingdom described in section 3.1.2.1. Further desirable information required also:

- Approximate practice location (i.e. State)
- Approximate owner location (i.e. State)

We were able to include more required criteria in the North American datasets because they were coming from the same PMSS provider and because we were paying for the provision of these data.

3.1.2.3 Data formatting

Where data format provided by each practice did not match that of a pre-established format suitable for analysis, dataset manipulation was performed using Rv.3.0.0. The pre-established format included a datasheet organised into seven columns (anonymous patient identification number, practice, date of birth, breed, gender, date, clinical text) with a row of data for each EMR entry. The date column included the date data was entered in the system. The clinical text column included all the text present in the EMR entry in one single text cell, including details of clinical examination, procedures, laboratory results, prescribing and invoicing.

The study was approved by Ethics and Welfare Committee of the School of Veterinary Medicine at the University of Glasgow.

3.1.3 Results

3.1.3.1 United Kingdom dataset

Of the total of 86 practices contacted initially, twelve agreed to participate in the study. Of these, five practices were included through GEP and the remaining seven practices had been included by directly

approaching the practice. For the vast majority of the 63 practices contacted directly that did not participate (42), no partner Veterinary Surgeon responded to any of the initial and following contact efforts (a total of three phone calls and emails over a period of two to three weeks). Of the practices approached indirectly through GEP, 29 out of 35 did not wish to participate.

Each dataset included the minimum database described in section 3.2.2.1. The format was very variable between systems and also within the same system, resulting in practices using the same PMSS providing datasets with different structure. Data from some PMSS required significant manipulation of variable technical difficulty to become suitable for analysis as highlighted by the following examples.

The first example is from the PMSS of one practice, which stored all veterinary reports and discharge instructions in a sequence of sub-folders each named with a number and the concatenation of number in the folder's name would provide the unique numeric identification for the animals. This numeric identification matched other data (e.g. day to day hospitalisation findings or billing information such as drugs dispensed) for that animal stored in other spreadsheets. Further, some of the first opinion cases (as well as most of the referral cases of this practice) had veterinary reports and discharge instructions that were stored as MS Word documents and were converted as unformatted text strings and added into a spreadsheet with their unique identification obtained from the concatenation of sub-folders names. This dataset included also patients from the small animal branch of the practice combined with the equine practice and a large amount of time and effort had to be spent in the identification and removal of non-equine patients. This was achieved by searching for documents that included other animal species. Further the date was automatically extracted from each document's name where available as a proportion of these include the date in the document's

name. For those where the date was not available in the file name, information about date was extracted from within the documents' as the date was usually included in the first row of data in most of the remaining cases. The remaining few hundred cases had to be edited manually individually as the date was present randomly within the text of the document so the process could not be easily automated.

Another first opinion equine practice included also data from the farm animal branch, which was removed by mining rows where words from other species were identified (the PMSS of that particular practice had no column to identify the species).

Data from two practices, using different PMSSs, was organised in multiple tables but a cross-referencing column was available to merge the two tables.

For two practices using the same PMSS there was the need by one of the technicians of the PMSS Company to write a script to specifically extract the information of interest and attach it to the demographic patient details.

Two practices had both first opinion and referral services included within the same dataset. For one of these the PMSS allowed differentiation between referral and first opinion cases. For the second practice, differentiation of first opinion and referral cases was performed by identification of the primary first opinion and referral veterinary surgeon. For both practices first opinion and referral services were well separated and although some veterinary surgeons from the referral service might have examined some first opinion case, this would have been a rare occurrence.

Data from two practices, that had agreed to participate, was unsuitable, having been provided in an unstructured spreadsheet. Ultimately data from 10 veterinary practices was available for use during the rest of the project.

These examples highlight the complexity of the process required for data cleaning and preparation for the subsequent analysis. This process required a significant effort and time, although could have been somewhat faster if performed by an experienced programmer. The remaining practices provided data that was readily usable.

The complete final dataset from the United Kingdom included a total of 2,653,695 rows (overall size 462.1MB) of data from 141,543 equine patients attended by the first opinion service of 10 equine practices between 1987 and 2013. The data is summarised in Table 3.1. Figure 3.1 represents the distribution of the amount of data (actual rows of data) entered in the respective PMSS each year by each practice. Figure 3.2 represents the number of new patients (i.e. patients not previously identified within the dataset) added to the dataset by practice each year. Figure 3.3 represents the total number of animals in the dataset by practice each year.

Table 3.1: Summary of the contribution of each practice to the dataset from the United Kingdom.

Practice	Region	Rows	Animals	Period	Gender (F-M-U)
A	Scotland	30677	3828	1993-2012	1676/291/61
B	Scotland	Unsuitable	-	-	-
C	Scotland	7805	215	2007-2012	0/0/215
D	South East	779328	70487	1987-2012	16780/23895/123
E	East Midlands	662484	9401	2007-2012	0/0/9401
F	North East	Unsuitable	-	-	-
G	North West	54044	3248	1995-2012	681/149/0
H	East of England	202514	9745	1997-2013	2445/6013/1287
I	North West	15431	1552	2010-2012	626/209/0
J	York & Humber	59927	3273	1994-2008	1260/1402/611
K	South West	566083	26059	2007-2013	6002/9372/10651
L	East of England	275405	13735	2005-2013	3226/3875/6634
Total			141,543		

Rows: rows of data; F/M/U: Female/Male/Unknown

Where the demographic details were available the dataset included 1,349 rows from 94 Donkeys and 664,197 rows from 34,559 horses. Of the remaining records 1,122,465 rows of data were from 87,710 animals classified as equines. The remaining 865,684 rows of data from 20,698 animals included no details of their species. In terms of breed 2,324 types of breed and breeds crosses were reported. These included also misspellings and different variations of abbreviations (eg. Thoroughbred, TB, Tb, tb, t/b, T/b, etc). The dataset included a total of 180,840 rows of data from 7618 Thoroughbreds (6.9% of the animals for which a breed was recorded), 44,520 rows of data from 1878 Warmbloods (1.7% of the animals for which a breed was recorded), 14,879 rows of data from 599 Arabian horses (0.5% of the animals for which a breed was recorded), 62,927 rows of data from 2941 draft horses (2.7% of the animals for which a breed was recorded), 117,738 rows of data from 4122 ponies (3.7% of the animals for which a breed was recorded) and 10,146 rows of data from 960 miniature breeds (0.9% of the animals for which a breed was recorded). No data of breed was available in 566,796 rows of data from 31,280 animals (22.2% of all animals).

The remaining 1,655,849 rows of data from 91,663 (83.5% of the animals for which a breed was recorded) animals were a mixture of combinations of breed crosses or misspellings or unclassifiable abbreviations. Gender was recorded for 79704 (56.5%) animals. This dataset included 32,697 females (41.0% of those for which gender was recorded; 541,366 rows of data), 27,778 geldings (34.9%; 410,318 rows) and 5,767 entire males (7.2%; 95,062 rows).

A further 13,462 animals (16.9%; 267,635 rows) was recorded as “male” with no specification to whether the patient was neutered or not. For 61,357 animals (43.5%; 1,339,317 rows) no gender was recorded.

Age was calculated as the difference in number of days between date of birth and the date the record was entered in the system. Date of birth was available for 77,754 (55.1%) animals (1,308,001 rows of data; 49.3%). The maximum age recorded was 2013 years and the minimum age was -23 years. As these were clearly mistaken dates entered in the system age was only included for those 1,115,060 (42.0%) entries where age was in the range 0 to 40 years. Of these the median record (or row of data) age was 9 years and the mean age was 10.8 years (1st quartile five years, 3rd quartile 15 years). A realistic age was not available in 1,538,635 (58.0%) rows of data of which 192,941 rows of data from 17,916 animals included an age that was likely wrong (less than 0 or greater than 40 years).

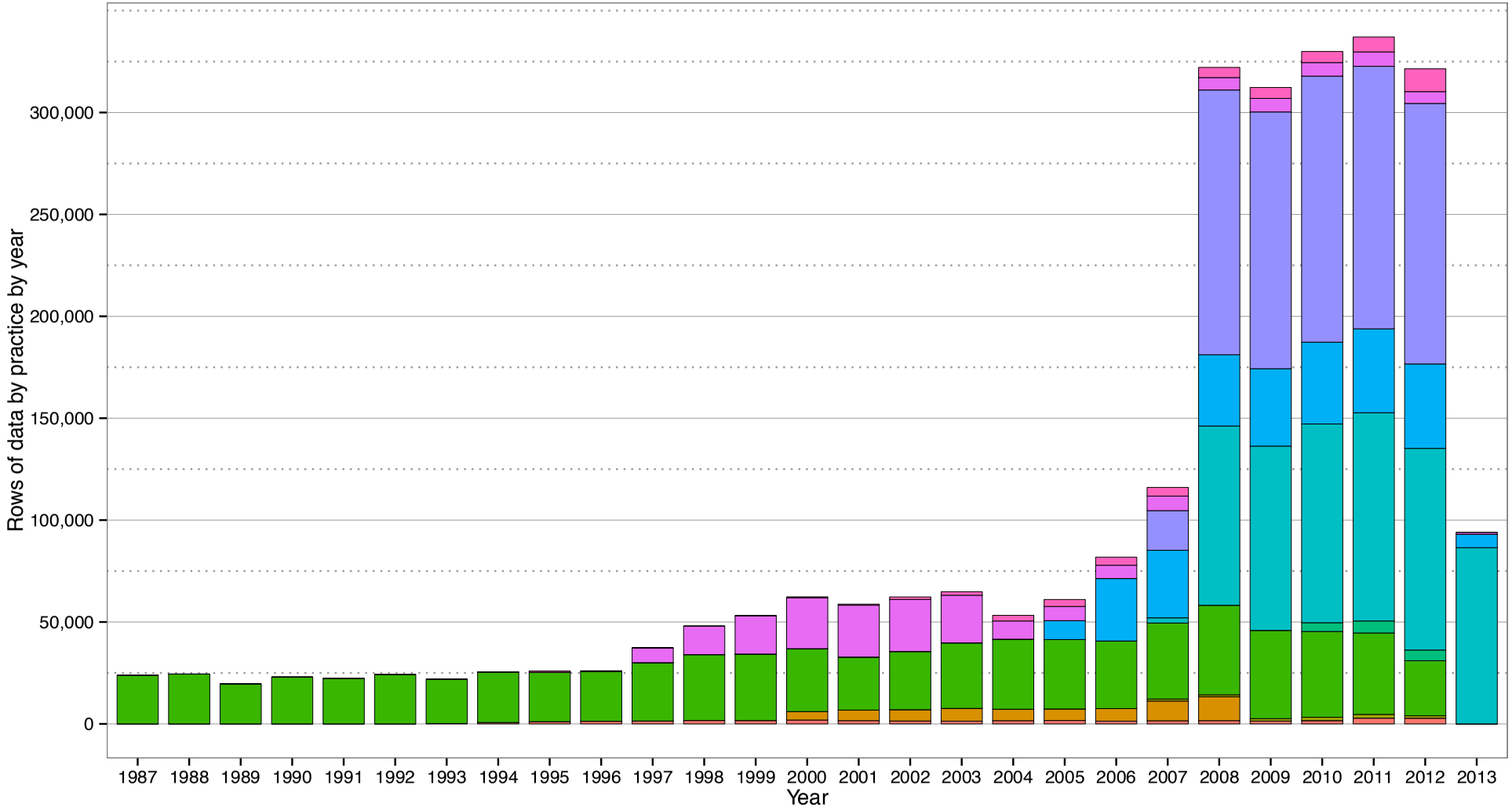

Figure 3.1: Barplot illustrating the number of rows of data provided each year by each of the 10 practices in the United Kingdom between 1987 and 2013 (each different colour represents a different practice).

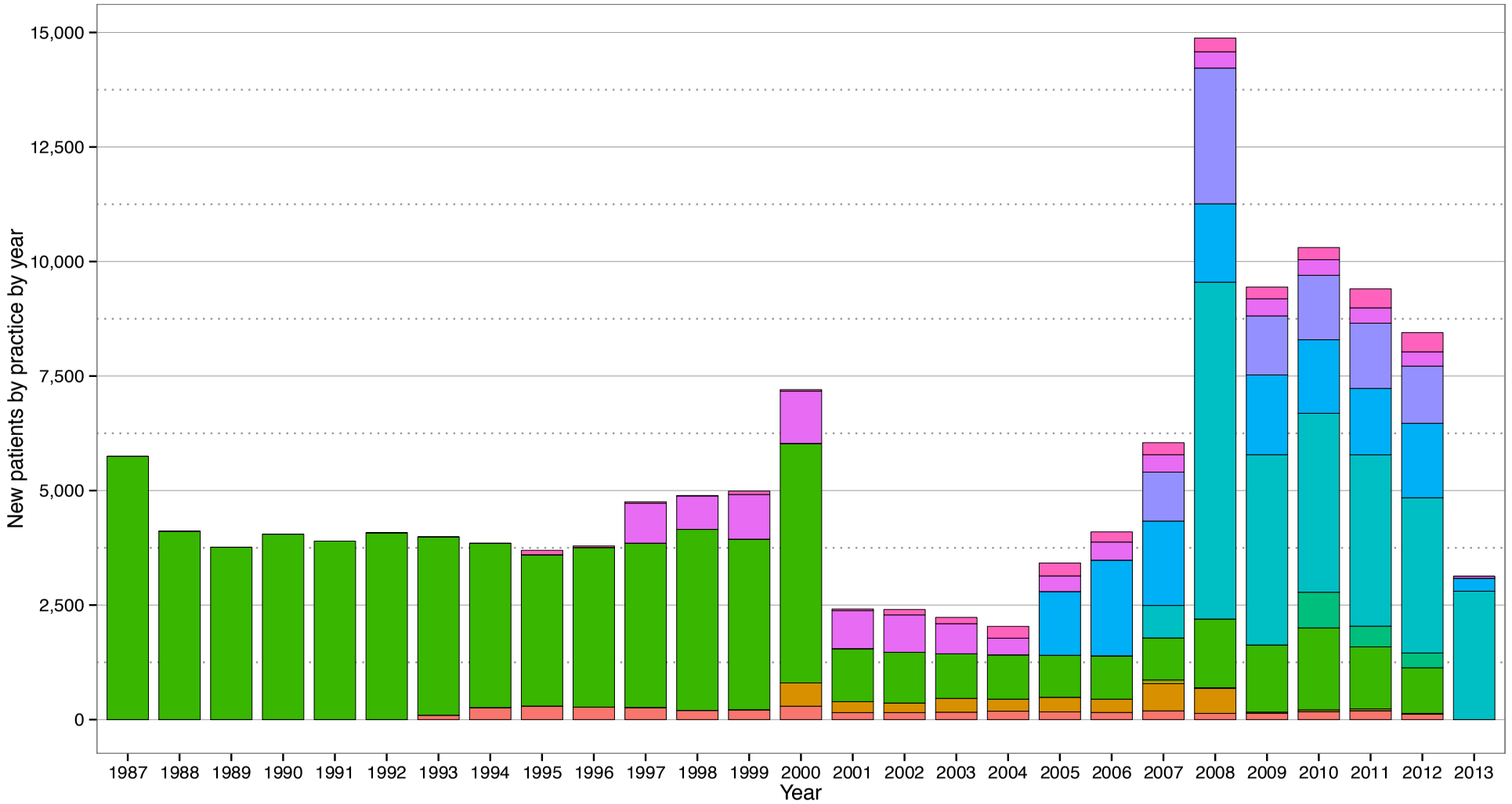

Figure 3.2: Barplot illustrating the number of new patients in each year by each of the 10 equine practices from the United Kingdom between 1987 and 2013 (each different colour represents a different practice).

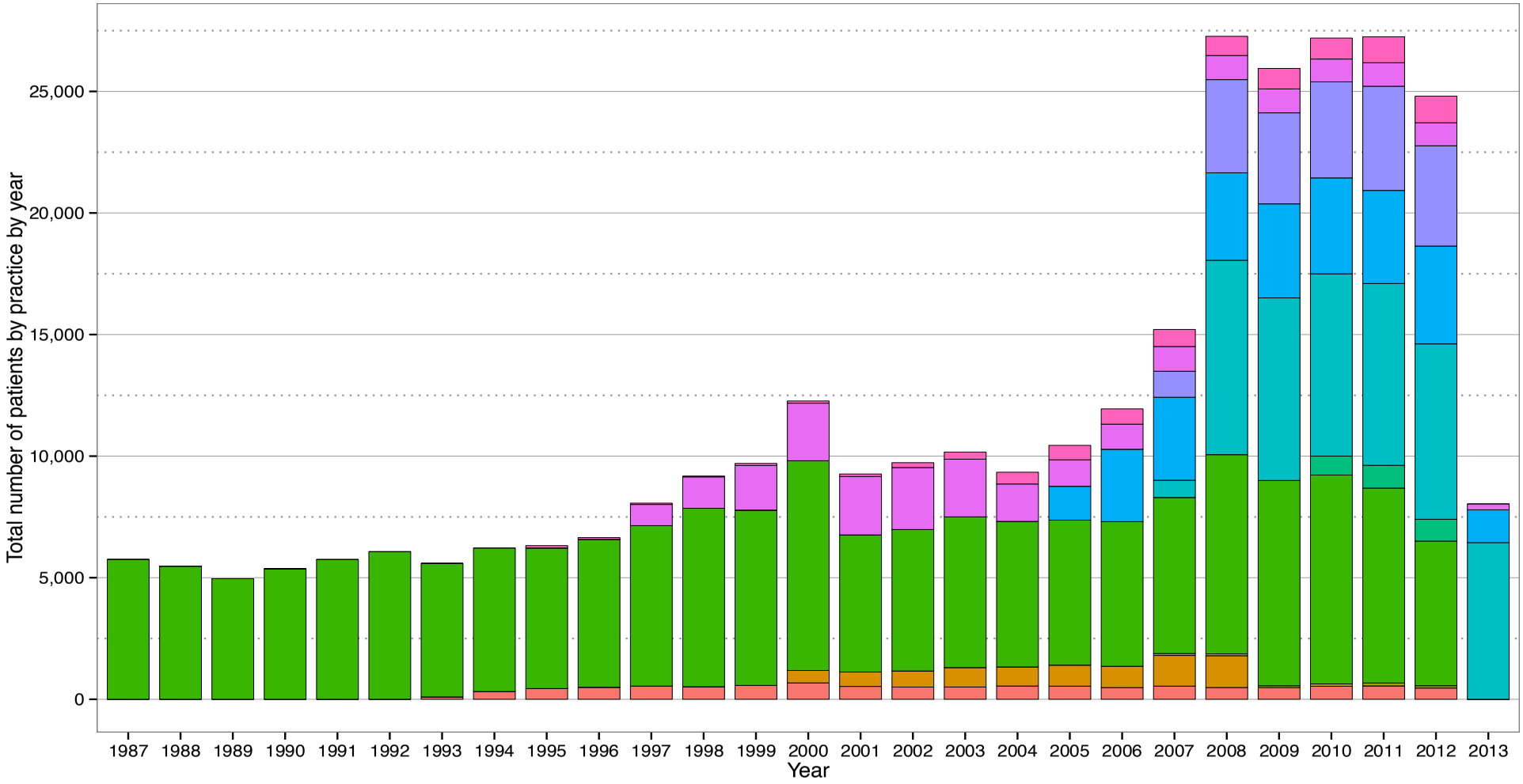

Figure 3.3: Barplot illustrating the number of animals in each year by each the 10 equine practices from the United Kingdom between 1987 and 2013 (each different colour represents a different practice).

3.1.3.2 North America dataset

The dataset from North America included data from nine practices including a total of 27 branches distributed throughout 16 states of Canada and the United States of America. The overall dataset included a total of 11,699,875 rows of data (overall data size 5.54Gb). Of the total of 312,634 equids in the dataset, 256,069 (81.9%) equids had first opinion records only, 15,941 (5.1%) equids had referral records only and 40,624 (13.0%) equids had records from both first opinion and referral services. The data spanned between 1994 and 2013. Figure 3.4 represents the distribution of the amount of data entered each year by each practice. Figure 3.5 represents the number of new patients by practice in each year and Figure 3.6 represents the number of horses in the dataset by practice each year. A proportion of data from three practices in North America prior to 2005, 2006 and 2007, respectively was known to have a wrong date of data entry to the system as a consequence of having imported data from a previous PMSS not compatible with the current PMSS.

As data was prepared directly from the PMSS Company, it was organised in a consistent structured manner. Each of the nine datasets included a comma separated value spreadsheet with 50 columns with the following information: two Anonymous Patient Identification (one for the practice and one general id), demographic details (species, breed, gender, colour, date of birth), date of data entry, and 42 columns to structure the clinical notes (including free text notes, note type, lab results and reference ranges, drugs dispensed and amounts, diagnosis, owner and practice state). The detailed columns of the dataset are summarised in table 3.2.

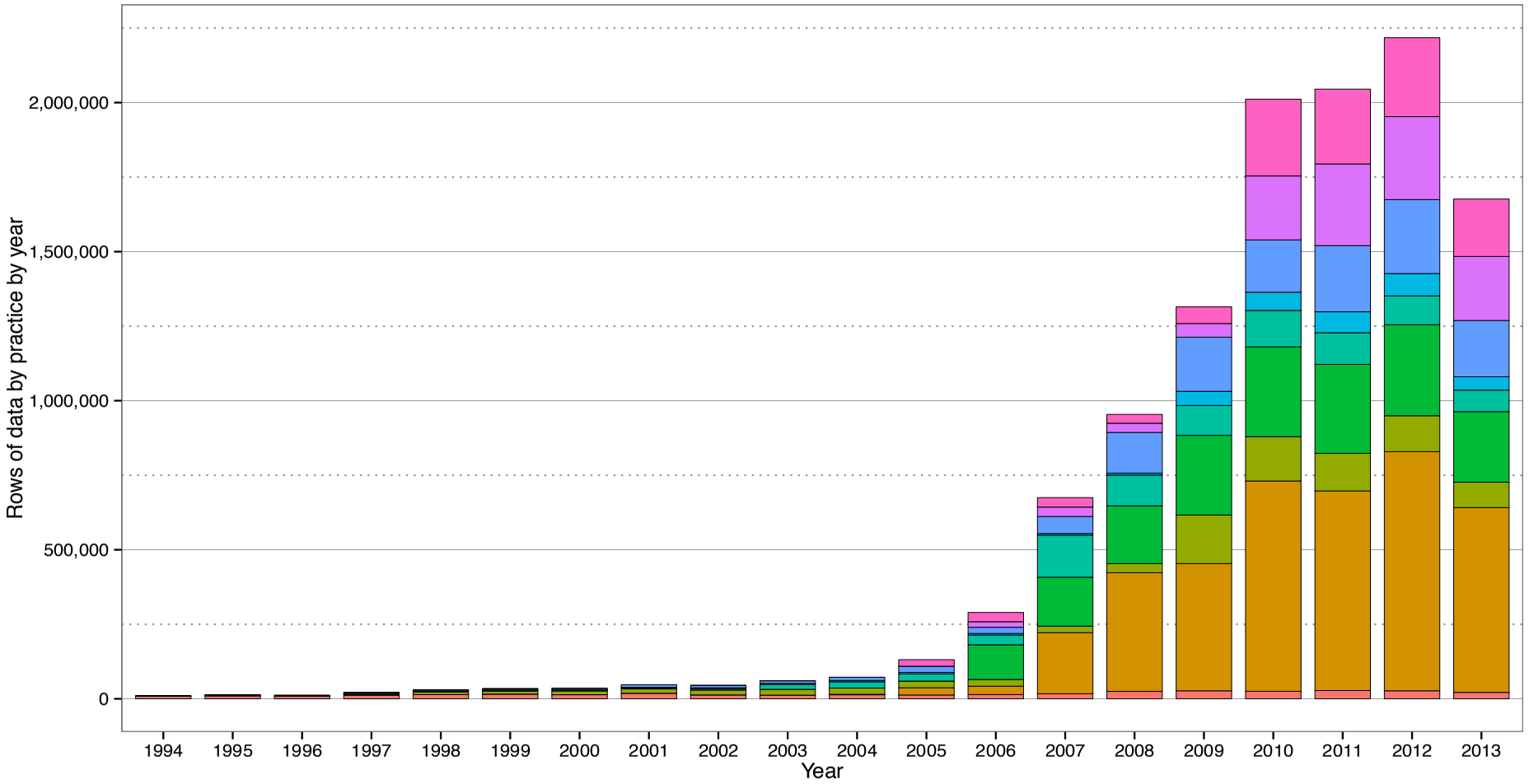

Figure 3.4: Barplot illustrating the number of rows of data provided each year by each of the 9 practices from North America (U.S.A. and Canada) between 1994 and 2013 (each different colour represents a different practice).

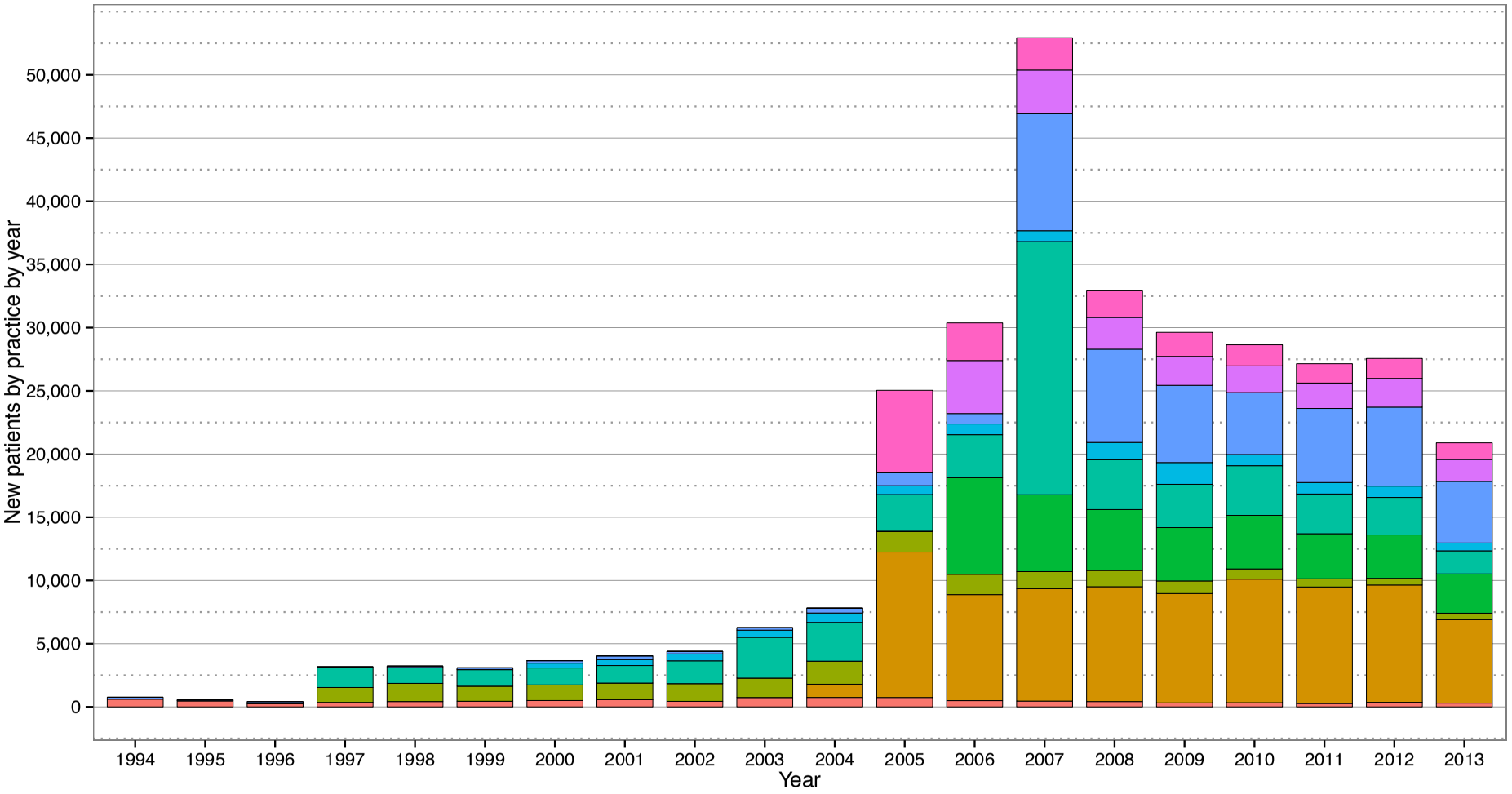

Figure 3.5: Barplot illustrating the number of new patients in each year by each of the 9 practices from North America (U.S.A. and Canada) between 1994 and 2013 (each different colour represents a different practice).

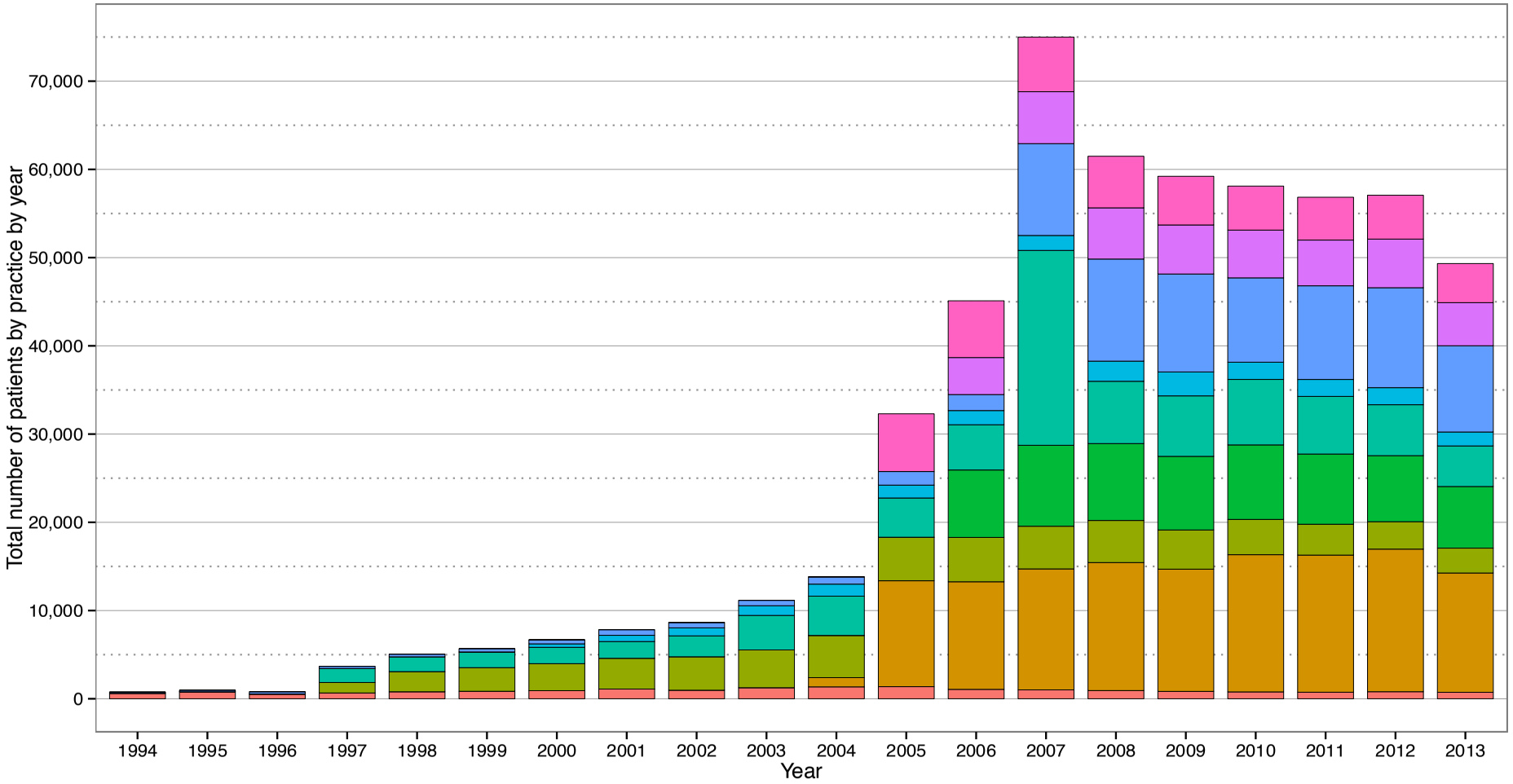

Figure 3.6: Barplot illustrating the number of animals in each year by each of the 9 practices from North America (U.S.A. and Canada) between 1994 and 2013 (each different colour represents a different practice).

Table 3.2: Table summarising the contribution of each practice to the dataset from North America.

Practice	Region	Br	Rows	Animals	Period	Gender (F/M/U)
L	Colorado	1	339476	9329	1994-2013	3569/4341/1419
M	7 States	14	3880795	82431	2004-2013	20198/25204/37029
N	Virginia	1	847674	20483	1997-2013	7510/10924/2049
O	Alberta	2	1884163	37117	2006-2013	9789/13085/14243
P	Ontario	4	857020	60551	1996-2013	7977/10761/41813
Q	California	1	334532	11537	1999-2013	2184/2406/6947
R	2 states	2	1311646	48369	1994-2013	9678/13627/25064
S	Tennessee	1	1108298	20580	2006-2013	3875/7637/9032
T	Wisconsin	1	1136271	22237	2004-2013	9057/10721/2459
Total	**-**	**27**	**11699875**	**312634**	**117 years**	**73837/98742/140055**

Br: Branches; Rows: rows of data; 7 States: Texas, Arizona, N Mexico, Oklahoma Montana, Iowa and Florida; 2 States: Florida and New York; F/M/U: Female/Male/Unknown

The last column included also the identification of the primary veterinary surgeon. This was requested to identify those veterinarians with high post-graduate clinical qualifications (DACVS, DACVIM and DACVT) or that worked as part of the referral service. In fact, this PMSS had no inbuilt facility to differentiate between first opinion and referral caseloads in those practices where both services were available. Opposed to the dataset from the United Kingdom, for the dataset from North America the population was a mixture of first opinion and referral as it was not possible to distinguish between these two populations.

While the PMSS data was well structured, the content of the clinical notes was extremely variable and reliant on veterinarians/veterinary technicians. For example a single visit would typically include data over several rows for which demographic information was repeated in each row. The clinical notes could be included in a single cell per row or in multiple cells per row. For cells where no data was available or applicable or entered in another row for that visit the cell would be filled as Not Available (NA). For example, if a horse had an orthopaedic examination and received some phenylbutazone and some blood work was performed then the clinical findings were included on one row and the lab-work results and reference ranges on another row or on the same row. Details of the drug dispensed would usually be reported in separate columns on

the same or on a different row such as “Invoice Item Description”, “Dispensed” or “Quantity”, but sometimes these details were included also, or just, in the free-text clinical notes columns. All these rows shared the same demographic information and date of data entry, which allowed identification of that particular examination on a patient. A list of the most relevant columns included in the dataset from North America is summarised in Table 3.3.

Table 3.3: List of the most relevant columns including the clinical notes in the dataset from North America.

Column name	Content	Rows with empty cell
Note	Free-text notes	9,518,669
Result type	Description of Notes/Value column	8,505,189
Value	Free-text notes	8,529,218
ResultDataType	Invoice vs Appointment vs Result	635,672
Result Units	Units of parameter	11,333,279
ResultRefRangeHigh	Higher end of reference range	11,390,356
ResultRefRangeLow	Low end of reference range	11,390,356
ResultComments	Free-text notes	3,325,997
InvoiceItemDescription	Free-text notes	5,469,218
ItemDescription	Free-text	5,469,207
ContainerDescription	Dispensing code	8,754,044
DiagnosticCategoryName	Administrative code	11,446,396
DiagnosticCodeName	Administrative code	11,686,190
DiagnosticSubCodeName	Administrative code	11,697,864
DiagnosisName	Diagnosis	11,699,857
DiagnosisPathString	Pathological diagnosis	11,699,864
IsPresentingComplaint	Presenting complaint	All Empty
DiagnosisDifferentalDate	Date differential diagnosis added	11,699,871
DiagnosisTentativeDate	Date tentative diagnosis added	11,688,874
Physical	Quantity dispensed	6,230,050
Dispensed	Quantity dispensed	6,230,443
Quantity	Volume administered	5,470,720
MeasurementName	Administrative code	5,469,897
DiagnositcClassName	Administrative code code	11,375,708
OwnerState	Owner state	45,251
PracticeState	Branch location	1700
RecordProviderName	DVM identification	82,854

Other columns missing are anomymous identifiers for who input the data, under who the data is invoiced to.

As a significant degree of variability was present in the way practices and their veterinarians stored the information in the system and to account for

typing errors and data input mistakes (data added to the wrong columns) all the columns including clinical information other than identification, demographic information, date of data entry and practice location were merged in a single column of text.

Where the demographic details were available the horse population dataset included 2,116 rows of data (0.02% of total) from 118 Donkeys, 147 rows of data (>0.01% of total) from eight mules, 273 (<0.01% of total rows of data from two zebras and 11,697,339 rows of data (99.98% of total) from 312,506 animals classified as equines. In terms of breed 646 types of breed and breeds crosses were reported. These included also different abbreviations and misspellings (eg. Warmblood, warm blood, Dutch WB, Begian Warmblood, Trakhener, VTrakhener, etc.). The dataset included a total of 3,817,733 rows of data from 66,960 quarter horses (34.5% of the animals for which a breed was recorded), 1,244,730 rows of data from 26,374 Thoroughbreds (13.6% of the animals for which a breed was recorded), 1,653,924 rows of data from 20,748 Warmbloods (10.7% of the animals for which a breed was recorded), 729,650 rows of data from 15,444 horses of American breeds (Appaloosa, Paint, Peruvian Paso, Tennessee walking horse, etc.) (7.9% of the animals for which a breed was recorded), 260,330 rows of data from 5444 Arabian horses (2.8% of the animals for which a breed was recorded), 201,483 rows of data from 5632 ponies (2.9% of the animals for which a breed was recorded), 80,121 rows of data from 1664 draft horses (Clydesdale, Percheron, etc.) (0.9% of the animals for which a breed was recorded), 37,912 rows of data from 849 cobs (0.4% of the animals for which a breed was recorded) and 80,402 rows of data from 2098 miniature breeds (1.1% of the animals for which a breed was recorded); 30,142, 12,866 and 26 rows of data were from 1038 donkeys, 469 mules and 4 zebras respectively (0.5%, 0.2% and 0.002% of the animals for which a breed was recorded respectively). No data of breed was available in 2,326,650 rows of data from 118,487 animals (37.9% of all animals). The remaining 1,223,906 rows of data from 47,423

(24.4% of the animals for which a breed was recorded) animals were a mixture of combinations of breed crosses or misspellings or unclassifiable abbreviations.

Data on gender was available for 172,579 animals (55.2% of total) and included 4,125,743 rows from 73,837 females (42.8% of those for which gender was recorded), 4,554,301 rows from 85,483 geldings (49.5% of those for which gender was recorded) and 876,618 rows from 13,237 entire males (7.7% of those for which gender was recorded). A further 22 (>0.1%) animals (447 rows) were recorded as "male" with no specification to whether the patient was neutered or not. For 140,055 animals (44.8% of total) no gender was recorded (2,142,766 rows).

Age was calculated as the difference in number of days between date of birth and the date the record was entered in the system. Date of birth was available for 198078 (63.3%) animals (9,357,112 rows of data; 80.0%). The maximum age recorded was 253 years and the minimum age was -101 years. As these were clearly mistaken dates entered in the system age was only included for those entries 9,235,185 rows from 189,311 animals (60.6% of the total) in the range 0 to 40 years, resulting in the exclusion of age in 121,927 rows of data (1.3% of those with a date of birth available) from 8767 animals (4.4% of those with a date of birth). Of these with a plausible age the median age was eight years and the mean age was 9.1 years (1^{st} quartile four years, 3^{rd} quartile 13 years) and this reflected the age at the time each row of data was entered in the system.

3.1.4 Discussion

The aim of this work was to assemble databases compatible with text mining. The practice-by-practice approach implemented in the United Kingdom offered no practical advantage other than being more economical. Data collection was completed at virtually no cost as data

was voluntarily provided free of charge by participating veterinary practices. These practices pay significant annual fees to their PMSS Companies, which often include basic technical support as part of the standard assistance contract. Data extraction can often be obtained using in-built search functions to create a comprehensive spreadsheet including all the information in the EMRs. However, assistance was required on two occasions from the PMSS Company to create queries to obtain the data required. In both cases the cost was affordable (£200) and provided data of suitable quality.

The greatest shortcoming of this approach was related to the time and effort required to gather data. A large number of practices had to be contacted at the start to include only few (17% of the 86 initially approached provided data) in the end and the response rate was similar to that of another similar study in small animals (Cameron et al., 2014). Previous studies requiring private equine practices to share client data reported a comparable low response rate of 23% (Hotchkiss et al., 2007). The overall process, including data cleaning, required more than 18 months to complete.

Approaching GEP proved helpful as partners of veterinary practices appeared more likely to reply to a message from a known contact such as the GEP group director, particularly when they had not previously been in touch with a member of our research group from the University of Glasgow. Collaboration with GEP worked as a mutual benefit as provision of data was obtained in exchange for free-of-charge presentations at one of their CPD events. Provisional results of data analysis were presented at GEP meeting in October 2013.

The variability in dataset format provided by each practice as well as the difference in PMSS used by participating practices also required significant efforts to obtain a dataset suitable for analysis. This required also a

significant amount of time and effort, including lengthy communication with some practices and their PMSS Company to clarify technical characteristics of their datasets and avoid data format misinterpretation.

The dataset obtained from North America was provided in a format ideal for analysis as cleaning and formatting was performed directly by the PMSS provider company. The dataset obtained in this manner was costly (about £9,000), but the cost was reflected in quality and quantity of data provided. This approach proved faster as data was obtained within weeks of finalising an agreement with this PMSS Company and was immediately ready for analysis.

Both United Kingdom and North American approaches produced final datasets with structures meeting the minimal requirements for our research. However, a large amount of desirable data was missing from both dataset, as much of the demographic details were not available as they were not recorded in the system by administrative staff, veterinary technicians/nurses or veterinary surgeons. Missing data is often a significant issue in retrospective studies and little (if anything) can be done to retrieve missing information, particularly where patient and owner's identities are masked. Nevertheless the amount of information available in the dataset was still sufficient for significant analyses.

A major limitation of both datasets was that there was no way to check correctness of data added to the PMSS due to the retrospective data collection. Although the vast majority of the information contained is assumed to be correct, errors are bound to be present for any of the information entered either automatically or manually. Correctness of patient identification is of pivotal importance for PMSS as this allows longitudinal tracking of patients through time and it is therefore created automatically. However, animals sharing first and last names might be confused by a member of staff entering the information in the PMSS.

While this is possible in theory when adding new information to the system, the amount of this type of incorrect data is likely to be minimal as the correctness of this information is essential to correct billing and the heart of practice-client trust and the importance of allocating the information to the correct patient would be emphasised to all staff working at these practices. Data referring to date of examination were added to the PMSS automatically so mistakes might arise through two mechanisms. The main mechanism is by importing old data from previous PMSS not compatible with the current system. A proportion of data from three practices in North America prior to 2005, 2006 and 2007, respectively was known to be wrong due to the incorrect importing of data from a previous not compatible PMSS. For one practice this was reflected by a very large number of cases seen in 2007 (~20,000) while the following years this practice had a caseload of roughly 5,000 cases per year. For all remaining practices from North America there was little change in the number of cases between years prior and following the introduction of the current PMSS. The second mechanism that might lead to recording an incorrect date may be due to systems failure where the date of the system is reset. Because it is a legal requirement for practices to keep accurate medical records, it is likely that any effect of system dates would be short lived and would have a minimal impact on the overall correctness of the data.

Date of birth, species, breed and gender were entered manually in the system and the amount of incorrect data is unknown. Age at the time of the veterinary consultation was calculated from the record date and date of birth. In some instances, the recorded date of birth was obviously erroneous (e.g. after the date of data entry or even of the year 1899), leading to ages of less than zero or more than forty years. These age values were removed before analysis, however an unknown proportion of retained ages (between zero and forty years) may have also been incorrect for the same errors in manual entry. Unfortunately there is no

way of retrospectively confirming the validity of these dates and related age, therefore age-related findings from the data should always be interpreted with some caution. However, there is no reason to suspect a systematic bias in manual errors in that manual errors would have been likely to cancel each other out and therefore have minimal impact on the overall results. Only if the error were systematic (eg. repeated shifting of decimal points) would the results have been seriously compromised.

As to the free-text clinical notes, these included any information deemed worth recording by the attending veterinarian. It is likely that some detail is missing due to forgetfulness or lack of commitment to keeping good records under different circumstances. Finally errors in clinical judgment (wrong diagnoses, diagnoses not supported by appropriate evidence) are also possible and simply reflect the nature of equine practice, particularly for ambulatory first opinion equine practice.

In conclusion, two possible strategies to create a large dataset of EMR have been described. While one method is cheaper, the other provided a larger amount of better quality data in a much more timely fashion. A limitation of both methods reflects the bias of using only practices willing to participate (United Kingdom data) or only using the one PMSS providing the data (North American data). Future efforts should aim to provide equine practices with good quality studies aiming to improve decision-making in equine practice as this might in return raise awareness in veterinary practice of the need for more good quality data to perform more studies, which would benefit both research and veterinary practice.

3.2 Text-mining electronic medical records from equine medical practice: methodology validation

3.2.1 Introduction

3.2.1.1 Free-text mining in the veterinary literature

The widespread use of electronic medical records (EMR) by private veterinary practices could support large epidemiological studies by providing large datasets of clinical records without the manual labour required to access traditional paper records. Automation of this process should significantly increase speed of data extraction. However, the technique used to extract the information of interest should be thoroughly validated to illustrate the reliability of the automated process. Electronic medical records are often stored as free-text with minimal structure and practice management software systems (PMSS) seldom offer means to search these records adequately (Anholt et al., 2014b; Erstad, 2003).

The potential applications of text mining in clinical research are multiple and include retrospective analysis of data to describe changes in disease prevalence, relationships between treatments and side effects, treatment outcomes, risk factor analysis and survival analysis. Prospective studies could combine the use of coding systems with text mining for syndromic surveillance (Anholt et al., 2014b), as has been successfully accomplished in human medicine (Brossette et al., 1998; Gerbier et al., 2011).

Text mining to retrieve targeted electronic medical records has been used in some veterinary studies in recent years (Anholt et al., 2014b; Lam et al., 2007a, 2007b; Oswald et al., 2010). Lam and colleagues (2007) have described a methodology to mine free-text clinical records using the software package SimStat/WordStat by Provalis Research. In that study the authors describe the reasons for retirement from racing for racehorses

in a dataset from the Hong Kong Jockey Club. This software has subsequently been used in risk factor analysis for retirement following tendon injuries or to determine the prevalence of cervical vertebral stenotic myelopathy in a thoroughbred breeding farm (Lam et al., 2007b; Oswald et al., 2010). A study by Anholt and colleagues (2014) reported its use in small animal veterinary practice in Canada to illustrate the frequency of antimicrobial usage in pets with diarrhoea demonstrating the potential of this methodology for disease surveillance.

Technique validation is extremely important for any assay or analysis in research. This applies also to text mining of EMRs. A recent study aimed to validate the use of WordStat for text mining veterinary EMRs against manual classification (Anholt et al., 2014a). This study describes in detail how analysis is performed using the software manufacturer's recommendations. The dataset was imported as a comma separated value file format and included 25,000 records from 12 first opinion small animal practices in Canada. The dataset included a column for the anonymised identification number of each animal, patient species, breed, gender and date of birth plus a rough indication of the owner post-code area and also a column with the free-text clinical notes (including chief complaint, history, physical examination findings, procedures performed and relevant results) added by the attending clinicians. The dataset did not include any standardised diagnostic coding or fixed vocabulary. The analysis was performed to automatically characterise the text contained within the free-text notes and resulted in high sensitivity (87%) and specificity (99%) proving that WordStat can be a useful tool in clinical epidemiologic research. These authors elected to sacrifice the identification of some true positives in order to minimise the number of false positives. However, while their specificity was excellent, sensitivity might be improved further with a substantial change in the methodology to improve identification and exclusion of falsely positive and falsely negative cases. A similar approach is described by Lam and colleagues (2007) with the

addition of a review process of the data incorrectly classified and update of the search dictionaries in an iterative manner. However, this study did not report sensitivity and specificity of this technique (Lam et al., 2007a)

3.2.1.2 Text-mining software

Documents to be searched can be imported into WordStat[11] after being processed in SimStat[12]. Simstat will support files of several formats including csv, xls, xlsx and xml. For the purposes of this study data was imported as csv. Import was achieved by following the path "file -> Data -> Import". The import process requires a variable amount of time depending on the file size (number of columns and rows imported) and machine power.

Once imported in SimStat, the menu driven command: "Statistics -> Choose X and Y" opens a "Choices Dialogue" window that will allow the user to determine which columns of the dataset are to be used as reference (e.g. row numbers) and which contain the text to be mined. The reference column is to be entered in the tab labelled "Independent:" and the column or columns containing the text to be mined are to be entered in the tab labelled as "Dependent:". Once each column of interest has been added, WordStat can be launched by selecting "Statistics -> Content Analysis".

WordStat is a linguistic based program that allows identification of predetermined words or compound phrases included in a user-defined categorisation dictionary within a given free-text, returning all rows of data ("cases") matching any word in said dictionary. The software includes a feature to exclude terms of little semantic value (561 terms between pronouns, conjunctions and adverbs) unless these are included in the categorisation dictionary. This feature simplifies and speeds up the

[11] WordStat v6.1.20, Provalis Research, Montreal, Canada
[12] SimStat v2.6.2, Provalis Research, Montreal, Canada

process by removing a large number of terms repeated throughout the text and that likely have little, or no significant, bearing on the interpretation of the text.

The “Frequency tab” in WordStat then produces a list of all remaining words within the dataset and this list can be used to further define the categorisation “inclusion dictionary” and increase the sensitivity including misspellings and abbreviations actually included in the dataset. This list of words can be exported as a spreadsheet and evaluated using other software such as MS Excel or Numbers. The software also offers some syntax shortcuts to include words with a similar root by adding the character “*” at the end of the common root. For example “abd*” would identify all cases including a word starting as “abd”, which includes “abdomen”, “abdominal”, “abd.”, etc. Nevertheless unique or rare spellings that do not follow a specific rule would still need to be included individually as specific words. The word list is a very useful tool as it provides the user with the means to produce a dictionary that includes all relevant terms and their variations that are actually present in the dataset.

As identification of a specific word within the text might not always correctly classify a case as part of the appropriate category, negations must be taken into account. For instance if the characterisation dictionary includes the word “diarrhoea”, a sentence such as “the horse does not have diarrhoea” would be classified as positive, whilst clearly the horse “does not have diarrhoea” and therefore should be classified as negative. Efficient identification of such false positives maximises the specificity of the analysis. WordStat supports the creation of semantic rules using boolean functions (“and”, “or”, “not”) or other modifiers (“no”, “if”, “watch”) and proximity operators (“near”, “before”, “after”) to further categorise cases. For example an operator might specify that any case for which the word “not” is present within three words of the word “colic”

than the case is to be classified as a “not colic” rather than in the “colic” group. This is done using a specific but simple syntax. For this example the rule to obtain this result should be coded as “not BEFORE colic/C 5”. A list of rules to exclude false positive cases could be included in an “exclusion dictionary” that could then provide a list of those cases that should be removed from the more comprehensive “inclusion dictionary”. All rules in the exclusion dictionary are created starting from words or combinations of words included in the inclusion dictionary. Creating these rules in the exclusion dictionary will remove false positive cases ultimately optimising specificity. However, the linguistic variability of free-text records is immense and creating rules that fit all data is often not possible and a degree of error is likely to be present, so that in the end finding the right balance between cases to be included or removed becomes a subjective process partly based on trial and error, running analysis, examining the output, updating dictionaries and re-running the output (Lam et al., 2007a). For example the rule “not BEFORE colic/C 5” might identify as false negative the sentence “do not call if the horse does not colic again” as the horse is likely to have shown abdominal discomfort at some point. However, the sentence “the horse did not colic again in this occasion, but has foot pain” similarly would also be incorrectly identified if the rule were removed. Therefore if the methodology used for text-mining includes pre-set rules to identify false positive cases, it is appropriate to report sensitivity and specificity of the inclusions-exclusion dictionary combination against the dataset being mined.

An alternative, which may be more time-consuming, but might ultimately offer better results, is that of searching for relatively complete sentences identifying false positive cases and adding those directly to the exclusion dictionary. This is laborious as the output search from the inclusion dictionary is to be evaluated manually. However, this would allow extraction only of truly false positives improving sensitivity, specificity and positive and negative predictive values.

This study describes the validation of a text-mining method depicting exclusion terms individually rather than by rules and compares the results to the literature (Anholt et al., 2014a)

3.2.2 Materials and Methods

3.2.2.1 Data used for validation

Lifelong clinical records were extracted from a random sample of patients from the United Kingdom dataset described in the Chapter 3 using R v3.0.2. Some patients were randomly selected from a group for which the records mentioned colic (50 animals). Patients for which conditions with an expected very low prevalence such as right dorsal colitis (3 animals) and renal failure (17 animals) were mentioned at some point during their life were identified using the search feature of a text editor software[13] and were subsequently added to the population to ensure that cases with a very low prevalence were included in the validation process. Finally records from a subset of 265 animals randomly selected from the overall population 141,543 animals from the United Kingdom, excluding those 70 already included, were also included. The random selection of patients was programmed so that the final number of rows of data from each practice would reflect in the validation dataset the overall proportion of rows of data from each practice as closely as possible. The obtained dataset is from now referred to as "validation dataset". Columns for patient identification, date and free-text clinical notes were also included. There was no standardised diagnostic coding, fixed vocabulary in the dataset.

[13] TextEdit, version 1.9, Apple Inc., Cupertino, CA, USA

3.2.2.2 WordStat settings

The validation dataset was imported into SimStat by classifying the patient identification and note columns as the independent and dependent variables, respectively in the “Statistics -> Chose X-Y” tab.
WordStat was launched by clicking on “Content Analysis” under the “Statistics” tab. Wordstat first view is at the tab named “Dictionaries”. In this, the “Exclusions:” option is to be unselected while the tab “English - Lemmatization” is selected in the scroll down menu by “Substitution:”.
Under the “Options” tab the following boxes were selected. Under the sub-tab “Text Processing” no option was ticked other than “Accept numeric character” under the “Characters:” section. In the box “Add characters appearing:” box, a string including all punctuation characters such as: !?-+:”;’,.<> was added to both boxes labelled as “Anywhere”. Under the “Speller/Thesaurus” tab, in the box “Active spell checking dictionaries:” the “American.adm” and “British.adm” options were selected, whilst “Ignore words containing numbers” and “Ignore words in uppercase” were deselected.

3.2.2.3 Inclusion dictionary

After selecting the WordStat settings highlighted in the previous section, clicking on the tab “ Frequencies” WordStat created a list of all terms included in the dataset, which was then exported into a MS-Excel spreadsheet to be evaluated manually word-by-word. Any term that might have been of interest was then included in an appropriate categorization dictionary. This included terms spelled correctly as well as words that identify terms of interest spelled incorrectly or abbreviated. Also words which could be part of combinations of words identifying conditions of interest (eg. “right” as part of “right dorsal colitis”) were included.

3.2.2.4 WordStat search

After each categorization dictionary was created from the list of words produced in the "Frequency" tab, the rows of data (cases) that included the terms contained in the categorization dictionary were identified by clicking on the "keyword-in-context" tab. The result produced a spreadsheet with five columns including the position (row number) of that row of data in the original dataset, three columns with text including all text immediately before the searched term, the searched term and all text immediately after the searched term, respectively and the last column identifying the animal identification number. The result of the search was then saved as a ".csv". This procedure was performed for each of the dictionary terms of interest selected in the "Keyword:" scroll down menu under the "Keyword-in-context" tab.

3.2.2.5 Exclusion dictionary

The output of the search obtained from the characterization dictionary (inclusion dictionary) was subsequently evaluated manually to identify false positive cases. Each exclusion dictionary was created with combinations of words identifying false positive terms.

Once a comprehensive exclusion dictionary had been created, the cases that contained the exclusion terms were identified by scanning the dataset against the exclusion dictionary and the result was then exported as a ".csv" file. This output included the original data row number, three text columns and the animal identification number.

3.2.2.6 Removal of false positive terms

Both search results from inclusion and exclusion characterisation dictionaries were imported into R v3.0.2 so that the false positive terms

identified by the exclusion dictionary could be removed from the search results of the inclusion dictionary. This was achieved in R v3.0.2 by the one-line command:

true.pos<-inclusion[!inclusion$rowames%in%exclusion$rownames,]

where “inclusion” is the dataset including the rows of data extracted from the original dataset as containing terms in the inclusion dictionary, and “exclusion” is the dataset including the cases matching terms within the exclusion dictionary.
The row numbers (“rownames”) in the resulting “true.pos” dataset were then used to extract the row of data from the original dataset with the command:

result<-orig.data[orig.data$rownames%in%true.pos$rownames,]

where “orig.data” is the original dataset including all the records. The dataset “result” included all the rows of data from the original dataset that included terms in the inclusion dictionary but that also excluded the cases containing terms specified in the exclusion dictionary.

3.2.2.7 Re-inclusion dictionary

The output search of the exclusion dictionary was also evaluated manually to identify whether it included any row of data identifying a false negative term. Combinations of words uniquely identifying these false negative rows of data were included in the re-inclusion dictionary. Mining for terms in the re-inclusion dictionary within data obtained from the exclusion dictionary identified truly positive terms, which were subsequently re-added to the “result” dataset obtained in section 3.3.2.6.

The whole mining procedure is summarised in Figure 3.7.

3.2.2.8 Characterisation dictionaries used for validation

For technique validation four dictionaries were searched. The first characterisation dictionary included a class of drugs such as “NSAIDs”. The second dictionary included a condition of an expected relatively high prevalence, such as “colic”, while third and fourth dictionaries included conditions at expected low prevalence such as “renal failure” and “right dorsal colitis”. Preparation of inclusion, exclusion and re-inclusion dictionaries was performed as described in sections from 3.2.2.2 to 3.2.2.7.

3.2.2.9 Manual classification as “gold standard”

The validation dataset was classified manually only after the automated classification process as described between sections 3.3.2.1 and 3.3.2.7. Manual classification was performed using MS Excel on the same file that had been imported into WordStat for analysis, including two columns, one for the animal identification and one for the free-text records. A third column was then used to encode each row of data according to one of the categories: “NSAIDs”, “colic”, “renal failure”, “right dorsal colitis”. Rows of data were included in each of the four categories if an NSAID was administered/prescribed, a horse exhibited signs of abdominal discomfort, or a diagnosis of renal failure or right dorsal colitis was made, respectively. The dataset including the manual classification column was saved as a “.csv”.

The text-mining process is summarised in Figure 3.7.

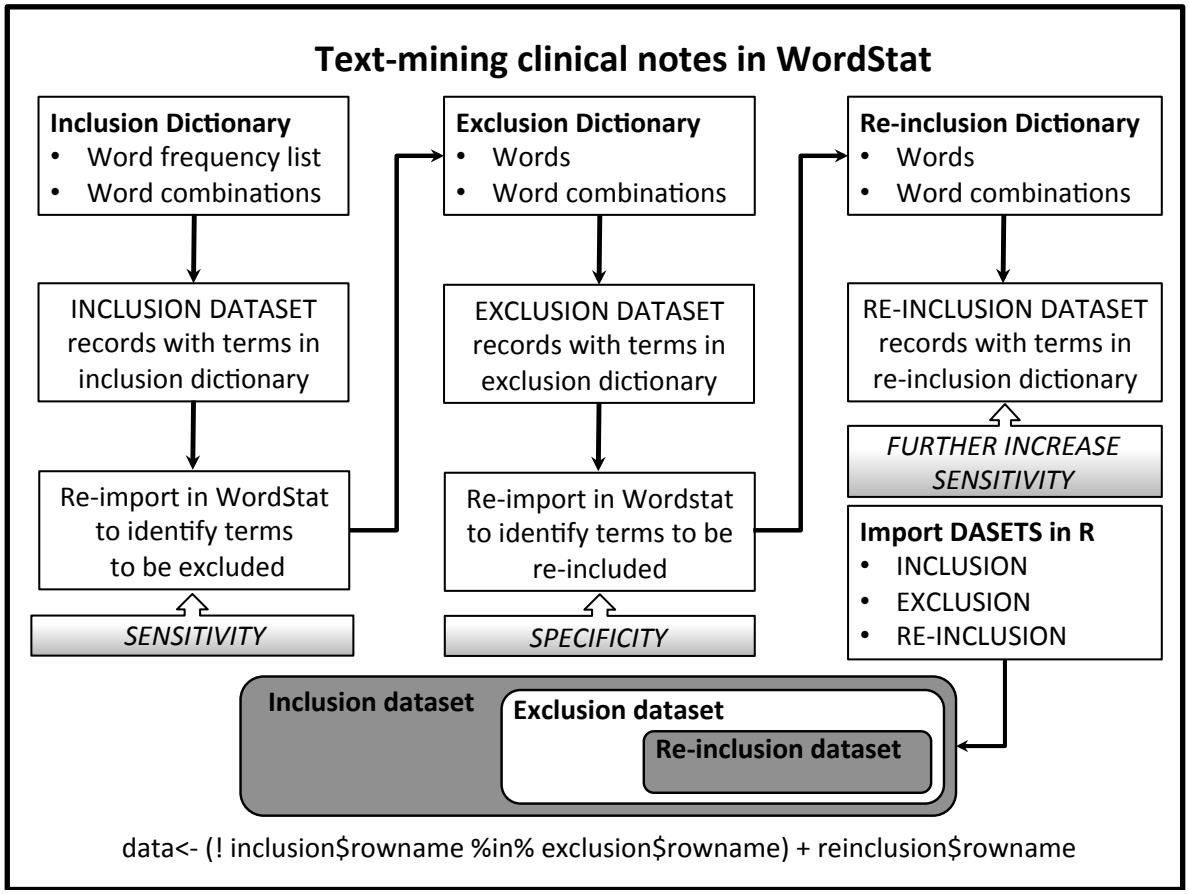

Figure 3.7: Flow chart summary of the text-mining process adopted in the study. Inclusion and re-inclusion dictionaries defined analysis sensitivity; Exclusion dictionary defined analysis specificity. The tree datasets are then imported in R v3.0.2 **to remove rows from the Exclusion dataset from the Inclusion dataset and to then re-include those in the Re-inclusion dataset. The lower portion of the picture summarises how the final dataset (dark grey) resulted from the subtraction of the exclusion dataset from the inclusion dataset and the final addition of re-inclusion dataset obtained from the exclusion dataset.**

3.2.2.10 Comparison between manual and automated classification

The search output for each of the four characterization dictionaries as well as the validation dataset including the column with the manual classification were imported into R v3.0.2. The row number obtained from automated and manual classification was compared and any discrepancy recorded and subsequently re-evaluated manually to investigate the source of the disagreement. A note was made as to whether the mistake was made by the automated or manual analysis.

3.2.3 Results

3.2.3.1 Data

A subsample of clinical records from 335 animals was selected with a total of 17,561 rows of data. The number of cases and rows of data per practice are summarised in Table 3.4.

Table 3.4: summary of cases used for validation from the original dataset for the validation dataset. Data obtained from the United Kingdom.

Practice	Total patients (%)	Total rows of data (%)	Validation patients (%)	Validation rows of data (%)
A	3828 (2.7%)	30,677 (1.2%)	6 (1.8%)	89 (>0.5%)
C	215 (0.2%)	7,805 (0.3%)	3 (0.9%)	100 (0.6%)
D	70487 (49.8%)	779,328 (29.4%)	115 (34.3%)	2960 (16.9%)
E	9401 (6.6%)	662,484 (25.0%)	79 (23.6%)	10513 (59.9%)
G	3248 (2.3%)	54,044 (2.0%)	12 (3.6%)	492 (2.8%)
H	9745 (6.9%)	202,514 (7.6%)	26 (7.8%)	826 (4.7%)
I	1552 (1.1%)	15,431 (0.6%)	4 (1.2%)	40 (0.2%)
J	3273 (2.3%)	59,927 (2.3%)	8 (2.4%)	215 (1.2%)
K	26059 (18.4%)	566,083 (21.3%)	51 (15.2%)	1608 (9.2%)
L	13735 (9.7%)	275,405 (10.4%)	31 (9.3%)	718 (4.1%)
Total	**141,543**	**2,653,698**	**335 (0.2%)**	**17561 (0.7%)**

Values in brackets refer to proportion of animals and rows of data from that practice from total and of the respective proportions of animals and rows of data from each practice in the validation dataset compared to the total dataset. The validation dataset included 0.2% of all animals and 0.7% of all rows of data of the data from the United Kingdom.

3.2.3.2 Inclusion Dictionary

The inclusion dictionary for “NSAIDs” included 53 terms, 58 for “colic”, 13 terms for “renal failure” and 6 terms for “right dorsal colitis”.

Following data extraction of the total of 17,561 rows of data in the validation dataset, terms in the NSAIDs inclusion dictionary were present 1562 times in 1181 rows for NSAIDs, 356 times in 295 rows for colic, 23 times in 23 rows for renal failure and seven times in seven rows for right dorsal colitis.

3.2.3.3 Exclusion Dictionary

The exclusion dictionary for “NSAIDs” included four terms, 131 for “colic”, four terms for “renal failure” and no terms for “right dorsal colitis”.

Following data extraction of the total of 17,561 rows of data in the validation dataset, terms in the NSAIDs exclusion dictionary were present 125 times in 112 rows, 63 times in 57 rows for colic, twice in two rows for renal failure and no term for right dorsal colitis.

3.2.3.4 Re-inclusion Dictionary

The re-inclusion dictionary for “NSAIDs” included 4 terms, five for “colic” and no terms for “renal failure” and “right dorsal colitis”.

Following data extraction of the total of 17,561 rows of data in the validation dataset, terms in the NSAIDs exclusion dictionary were present 79 times in 78 rows, twice in two rows for colic. No term was present for both renal failure and right dorsal colitis.

The full list of terms in these dictionaries is reported in Appendix 3.3.

3.2.3.5 Result dataset

Rows of data including false positive terms consistent with an erroneous classification of unaffected patients as affected were removed as described in section 3.2.2.6 and the few false negative terms identified by the re-inclusion dictionary were re-included in the “result” dataset. This resulted in 1149 rows for NSAIDs, 239 rows for colic, 23 rows for renal failure and seven rows for right dorsal colitis.

3.2.3.6 Comparison between manual and automated classification

There was good agreement between manual and automated search methods for each of the four dictionaries evaluated in the study.

Manual search identified 1132 cases out of 17,561 rows of data that had included a record referring to administration of NSAIDs, while automated analysis identified 1149 cases. Sensitivity, specificity and positive and negative predictive values were 99.8%, 99.9%, 98.3% and 100.0%, respectively. Manual classification missed 19 instances where NSAIDs had in fact been administered that were correctly classified by automated analysis. Two further cases were false negative cases incorrectly classified by the automated process, but correctly identified in the manual processing. This included words within the dictionary that had also been included in the exclusion dictionary as part of the sentence referred to a possible future use of the drug (so drug was not administered yet), while a previous sentence in the same case referred to the current use of the drug. For example a horse "*has been evaluated for lameness. Horse also has copd with dry hay and has diarrhoea after bute*." In this example a horse might be included as it has had diarrhoea with an inclusion dictionary for diarrhoea. However, the exclusion dictionary might include lameness terms, such as "*lameness*" as many cases that had receive phenylbutazone might contain several variations of a sentence intimating that if the horse develops diarrhoea whilst being administered an NSAIDs to manage a lameness then the owner should call immediately. The re-inclusion dictionary would include again the specific word combination that identifies this row of data "*Horse also has copd with dry hay and diarrhoea after bute*" so this row is not misclassified by the exclusion dictionary alone, without including other cases referring only to the hypothetical development of diarrhoea.

Manual search identified 226 rows of data referring to colic out of 17,561, while automated analysis identified 239 cases. Sensitivity, specificity and positive and negative predictive values were 100.0%, 99.9%, 94.6% and 100.0% respectively. Similarly, manual classification missed 13 cases identifying a colic episode that were correctly classified by automated analysis. The automated process did not produce any false negative result.

Regarding the conditions of expected low prevalence there was perfect agreement between manual and automated analysis. The dataset included 22 cases referring to renal failure and seven cases referring to right dorsal colitis and were all identified correctly by both methodologies. Sensitivity, specificity and positive and negative predictive values were all 100.0%. All the results are summarised in table 3.5.

Table: 3.5: Sensitivity, specificity, positive and negative predictive values (PPV and NPV respectively) of automated analysis compared to manual analysis reported as per cent values.

		Manual		Sensitivity	Specificity	PPV	NPV
		+	-				
Colic	+	226	13	100	99.9	94.6	100
	-	0	17322				
NSAIDs	+	1130	19	99.8	99.9	98.3	100
	-	2	16410				
Ren fail	+	22	0	100	100	100	100
	-	0	17539				
RDC	+	7	0	100	100	100	100
	-	0	17554				

In the 2x2 tables comparing automated and manual classification, rows are conditions identified by the software and columns correspond to manual classification. +/- in the manual "Manual" column identifies the number of positive and negative terms classified manually in each category (Colic, NSAIDs, Ren Fail and RDC). The +/- of each category identifies the number of positive and negative terms classified automatically. NSAIDs: non-steroidal anti-inflammatory drugs; Ren fail: renal failure; RDC: right dorsal colitis

3.2.4 Discussion

The findings of this study highlight that the free-text mining methodology here described can achieve excellent results, comparable to that of the current "gold standard" which is manual analysis. It is arguable whether

manual analysis is really the “gold standard” as it has not been validated being for years the only method available to classify free-text medical records. While this method yielded acceptable results for its intended purpose so far, one must acknowledge that there are some limitations in manual evaluation of medical records as this method is based on the assumption that an operator will limit classification mistakes to a minimum. Nevertheless evaluation of human/operator error should be an integral part of any validation process (Huber, 1998). Furthermore, one might expect that operator induced errors might increase with operator fatigue, which increases along with the length of the dataset. In the current study a total of 32 errors were produced by the operator, by missing the presence of a term, which had otherwise been identified by the automated analysis. Even though these errors were present after a single manual analysis, likely consequence of operator fatigue, the difference with automated analysis was minimal and most importantly, the automated analysis yielded a better result than manual analysis. Manual classification also has considerable room for improvement. In the present study one operator read and classified the validation dataset once. Employing two or more operators to read the same text and reading the text more than once could improve significantly sensitivity of manual analysis. This however yields the drawback of becoming then more time consuming and to require twice the manual effort with doubling of the costs required.

The technical time required to automatically mine the information of interest from the dataset is negligible in comparison to that of manual analysis (few seconds with the automated process, depending on machine power, compared to ~80 man/hours for the manual analysis for a dataset of 17,561 rows of data). However, it is important to point out that a large amount of time was necessary initially to create adequately comprehensive characterisation dictionaries. In fact dictionary creations required a much longer time (~300man/hours) than manual classification

for this small dataset of 17,561 rows of data. This highlights that the benefits of using automated analysis are directly proportional to the size of the dataset as the time required for analysis would mostly then be dependent on computer processing power. Further the dictionary could be used as a starting point to analyse different dataset examining the same conditions; however, ideally one should create a dictionary starting from the word frequency list and ensure that no new term is lost. Also a dictionary from a previous dataset used for a new dataset could include several terms not present in the new dataset, affecting the time required for analysis as several terms not in new dataset would be searched, even if not there. The analysis results would be unaffected if a dictionary contained terms not present in the data, however.

Excellent specificity and sensitivity were expected as each dictionary was created including all possible words that would have identified a certain category starting from the list of words actually present in the dataset. This included misspelled and abbreviated terms. Although browsing through the word list was a time consuming but pivotal step of the analytic process, it also means that the dictionary is very dataset-specific and if new data is added then the dictionary may need to be updated to include new terms that might be only present in the new data.

Sensitivity was higher than that of the study by Anholt and colleagues (2014a) preserving similar specificity. In that study specificity was as high as 99.3%, but the authors commented that this high specificity was obtained only by sacrificing sensitivity (87.6%). This discrepancy originates in the different approach used to create the exclusion dictionaries. Anholt and colleagues (2014a) defined the characterisation dictionary by setting semantic rules to identify false positive terms as described in section 3.3.1.2, but recognised that when numerous rules were developed to correctly classify all of the possible variations of a sentence, the process would improve either only sensitivity or specificity at the expense of the

other. False positive terms included sentences where the operator meant to report that a particular term was not present, for example that “the patient did not have colic”. Alternatively false positive terms could occur for single words with multiple meanings; for example “displacement” could identify a “colonic displacement” in the dictionary “colic” or “fracture displacement” in the dictionary “fractures”, or “dorsal displacement of the soft palate” in the dictionary “respiratory”, etc. For the example of colic, comprehensive sentences including negative expressions identified from the inclusion dictionary search output (“doesn’t have colic”, “not have colic”, “no colic”, “no sign of colic”, etc.) were included in the exclusion dictionary. In the case of words compatible in more than one inclusion dictionary, comprehensive word combinations were used for the exclusion and re-inclusion dictionaries. For example, the “exclusion colic” dictionary included terms such as “dorsal displacement of the soft palate”, “fracture displacement”, etc, whilst the re-inclusion dictionary included combination of words (down to the complete sentence) to identify terms that where truly positive enlisted in the exclusion list search.

As that study focused on syndrome surveillance, the authors elected to maximise specificity to ensure that the largest proportion of cases identifying a particular syndrome were correct (Anholt et al., 2014a). The authors also commented that their study’s findings were strictly setting specific and clarified how the process should be re-evaluated on a different dataset (Anholt et al., 2014a). For example, linguistic differences may reflect different geographical regions as well as temporal differences from where and when data had been generated. The methodology used in the current study differs in that false positive cases were identified manually by the definition of specific word combinations in place of construction of automated rules. Further, the method by Anholt and colleagues (2014a) identified a subset of cases that were positive in some instances and negative in others. This difficulty was

overcome also by adopting the re-inclusion dictionaries to ultimately correctly classify those ambiguous terms. This process was more laborious as it added several steps to the analysis process and was ultimately more time consuming but was justified by the resulting excellent sensitivity and specificity.

In the current study inclusion of all terms and their variations, abbreviations and misspellings was of pivotal importance to maximise sensitivity of the analysis process. Despite the effort to include all appropriate terms actually present in the dataset, sensitivity was not 100% as some positive cases where still misclassified after subtraction of terms in the exclusion characterisation dictionary. Mostly this occurred when true positive terms were present along with terms included in the exclusion dictionary in the same case, as described in the results section. For example a sentence that said: “this horse suffered abdominal pain today, but did not colic after administration of a NSAID”, would be initially detected by the inclusion dictionary as including the words “abdominal pain” and “colic”, but would be then excluded as containing the word combination “did not colic”, whilst it is clearly a colic case. Fortunately this was a rather rare occurrence in the dataset and the use of a re-inclusion dictionary provided an efficient solution to this problem. The use of an exclusion and re-inclusion dictionary appeared more useful for dictionaries identifying clinical syndromes, such as colic. Clinicians might discuss a differential diagnosis so exclusion and re-inclusion dictionaries might be helpful to differentiate between true and false positive cases. However, for the identification of drug usage the adoption of exclusion and re-inclusion dictionaries is of arguable benefit. Drugs are recorded when they are administered or dispensed for billing purposes or when the clinician wishes to record that treatment has commenced. In the present study, adoption of exclusion and re-inclusion dictionaries for “NSAIDs” was necessary only because of the ambiguity intrinsic to Buscopan® (Buscopan Compositum and Buscopan 20), since this name

identifies two commercial preparations, one with and one without an NSAID. So these dictionaries allowed identification of terms referring to the formulation actually containing the NSAID. Since this ambiguity is unusual with pharmaceutical products, in the investigator experience the use of an inclusion dictionary alone might suffice and yields excellent specificity and sensitivity to identify drug usage. The investigator experience for drug categories other than NSAIDs is that no false negative terms are generally present therefore only the inclusion dictionary could be used for the analysis.

In the present study it was noticed that in most cases a relatively small number of word combinations identified the vast majority of false positive cases, which made exclusion dictionary definition somewhat faster. This is explainable by the fact that the person entering the text records might tend to use similar word combinations to describe similar scenarios. Nevertheless, great effort was required to include a large number of false positive cases in the exclusion dictionary and false negative cases in the re-inclusion dictionary. The vast majority of discrepancies between automated and manual classification was for terms classified as false positives, which were in fact found to be correctly classified by the automated analysis and had been missed on the first manual evaluation. The results of this study show that automated analysis of free-text clinical records is possible and can offer results at least as good as manual classification. In fact, our analysis shows that automated analysis has slightly better sensitivity than manual analysis as operator errors due to tiredness are minimised by the automation, which prevents missing some truly positive terms. However, sensitivity and specificity of automated analysis are still greatly dependent on an operator related process in the process of dictionaries' creation.

In terms of data handling, R v3.0.2 was used to remove false positive cases and to re-include truly positive cases that were initially excluded.

This was achieved by subtracting the rows of data identified from the exclusion dictionary search from the inclusion dictionary search output and to subsequently re-include those in the re-inclusion dictionary. This procedure could have been performed using other commercial software packages, such as MS Excel or MS Access, but R v3.0.2 was chosen because this software is an open source software and is not limited by the size of the dataset and therefore the methodology tested is also suitable also for larger datasets, exceeding the limits of 1.4 million rows of data or the 2Gb file size imposed by MS Excel or MS Access. Ultimately, the code written for R v3.0.2 was relatively simple and fast and produced the desired result in very little time for a dataset of this size. Nevertheless other software packages might have been used such as SQL or Stata. The decision of using R v3.0.2 was fundamentally based on the investigator's preference.

When comparing the efficiency of manual classification to that of the automated analytic process this study does not account for time and effort required to develop the skills necessary to set up and run the analysis in WordStat and R v3.0.2. A significant effort was put in place before starting the study to understand in depth how these software packages work and how they might assist this type of analysis. However, section 3.3.2 of this chapter describes in detail how the analysis has been done and should assist others wishing to use this technique on a different dataset. While following these instructions the use of WordStat is relatively straight forward, using a programming language like R v3.0.2 remains more challenging and would require further training in this programming environment, which is outwith the scope of this manuscript.

In conclusion, the automated process is significantly faster, once inclusion, exclusion and re-inclusion classification dictionaries are prepared on a dataset of this size. As all words present in the dataset are used, sensitivity does not appear to be an issue for this method of analysis. In terms of optimised specificity, the use of exclusion and re-

inclusion dictionaries is useful in situations where there are many false positive and subsequently false negative cases. This is achieved simply by evaluating the output search of the inclusion dictionary to identify any significant proportion of erroneous classifications. False positive and negative cases appeared proportionally more common when trying to identify general syndromes, such as colic, but less common when focusing on specific diagnosis or when looking at drug administration.

In the future this type of analysis may aid several applications, from the retrospective analysis of EMRs, to prospective studies for syndromic surveillance or to monitor changes in caseload in veterinary practice. Alternatively, EMRs can offer clues to compare treatment outcomes and efficacy or identify the prevalence and significance of side effects following drug administration on large patient populations.

CHAPTER 4 – Non-Steroidal Anti-Inflammatory Drug Usage In Equine Practice

4.1 Introduction

Knowing the prevalence of drug usage might provide useful information to understand the behaviour of veterinary practitioners. Despite economical and research efforts necessary to demonstrate safety and efficacy of new drugs that undergo an arduous licensing process years might pass by before practitioners embrace regular use of new pharmaceuticals. This is understandable as practitioners are responsible for using these drugs and some time is required to develop a personal experience with their use, switching from a well-established practice with older, known substances to new ones.

Further, every country has its own official body responsible to define appropriate drug usage within its jurisdiction. Our data was obtained from the United Kingdom, United States of America and Canada and in each of these countries veterinarians must, by law, prescribe drugs licensed for use in the species they are treating, as these drugs have been tested for safety and efficacy in these species. However, clinical practice often requires veterinary staff to treat patients when no drug is licensed for use in that species, but alternative drugs are licensed for use in other species with a similar condition or even in human patients.

In the United Kingdom, veterinary surgeons are allowed off-label use of some drugs under the “Cascade”, which is an official legislative provision in the Veterinary Medicine Regulations that allows veterinary surgeons to prescribe unauthorised medicine that would not otherwise be permitted for use in some species (www.vmd.degra.gov.uk).

The principle of the Cascade is that if no suitable veterinary medicine, authorised in the United Kingdom, is available to treat a condition in a determined animal species, the veterinary surgeon responsible for the animal may still be able to treat that animal by using drugs licenced in other species or for human patients if no veterinary product is available, to avoid causing unnecessary and unacceptable suffering. Similar legislations are enforced in the countries of North America. Describing the prevalence of off-label use of a certain drug in a certain species might also highlight the need for a licensed product containing that drug in that species. Ultimately, analysis of prevalence of drug usage in veterinary practice might assist governing bodies to prioritise drugs that require more urgent evaluation under the legal licensing process.

Analysis of the context of drug usage might also provide information for clinical research providing evidence to support current practice where such evidence is currently lacking. For example, in horses phenylbutazone is commonly used to treat pain and inflammation associated with orthopaedic disease and flunixin meglumine is generally used for inflammation and pain associated with soft tissue conditions (Sanchez and Robertson, 2014), despite convincing evidence of a very similar mode of action, both being nonselective COX inhibitors (Beretta et al., 2005).

In the United Kingdom, as well as United States and Canada, phenylbutazone is licensed exclusively for the treatment of musculoskeletal disorders, while flunixin meglumine is licensed for treatment of both musculoskeletal disorders and visceral pain. Therefore, use of phenylbutazone for the treatment of colic would be an off-label use in these countries, particularly when a suitably licensed alternative, such as flunixin meglumine, is readily available. If the practice of using phenylbutazone to treat patients with visceral pain is somewhat frequent, legislative bodies might consider extending authorization to treat visceral

pain with this drug should they consider it appropriate, as long as no evidence of unwanted side effects is identified.

The aims of this study are to describe the prevalence of usage of the most commonly used non-steroidal anti-inflammatory drugs (NSAIDs) in equine practice in the United Kingdom as well as in North America. Further, the NSAID choice for colic and orthopaedic disease will also be described.

The final section of this chapter will include an overall discussion and conclusion, comparing differences in NSAID use in veterinary practices between the United Kingdom, USA and Canada.

4.2 Non-steroidal anti-inflammatory drug usage in the United Kingdom

4.2.1 Introduction

The prevalence of NSAID usage has not been previously described in a multicentre large population of horses in the United Kingdom. Several pharmaceutical substances containing NSAIDs licensed for use in horses are available in this country. These include phenylbutazone, suxibuzone, flunixin meglumine, ketoprofen, carprofen, vedaprofen, metamizole, meloxicam and firocoxib. Flunixin meglumine, meloxicam and ketoprofen are licensed in horses to treat pain and inflammation associated with musculoskeletal disorders and alleviation of visceral pain associated with colic. Metamizole is available in the United Kingdom in a formulation with a butylscopolamine (Buscopan Compositum), a spasmolytic, for the treatment of pain associated with colic. Phenylbutazone, suxibuzone, carprofen are licensed for the treatment of pain and inflammation associated with musculoskeletal disorders in horses. Firocoxib is only licensed for treatment of pain and lameness associated with osteoarthritis for up to 14 days. Vedaprofen is available as an oral gel preparation, licensed for treatment of pain associated with musculoskeletal and soft-

tissue disorders and trauma, including preventative treatment before surgical trauma.

Other NSAIDs currently not licensed in horses but seldom used in equine practice include meclofenamic acid, eltenac and aspirin. Meclofenamic acid and eltenac were once available for use in the horse but their licenses have now been expired for over a decade. There is no licensed equine product containing aspirin, but acetylsalicylic acid is often used for treatment of hypercoagulative conditions which result in intravascular thrombus formation. Common conditions include thrombophlebitis following catheterisation and laminitis and therefore aspirin is used by some to prevent platelet aggregation, one of the mechanisms that lead to clotting.

The present study describes the use of these types of NSAIDs in a population of equids from the United Kingdom. The overall prevalence estimate and 95% confidence intervals (95% CIs) are determined and the yearly and mean prevalence of use for each drug is reported for the 5-year period between 2008 and 2012.

4.2.2 Materials and methods

The dataset of EMRs from 141,543 animals (between 1987 and 2013; 2,653,695 rows of data) was analysed with the methods described in detail in Chapter 3. Briefly, the list of the words included in the dataset was used to identify terms that might identify a NSAID. This included commercial brand names of NSAIDs as well as pharmaceutical names and colloquial names. The NSAIDs inclusion dictionary included a total of 232 words (Appendix 4) covering both brand and pharmaceutical names as well as various misspellings and abbreviations. The words included in the dataset were selected to identify NSAIDs in general and also in particular the following drugs: flunixin meglumine, phenylbutazone, suxibuzone,

ketoprofen, carprofen, vedaprofen, metamizole, meloxicam, firocoxib, meclofenamic acid, eltenac and aspirin.

Prevalence of NSAID usage and 95% CIs were then calculated as previously described (Wilson, 1927) for the equine population in the whole dataset while the yearly prevalence of drug usage was calculated between 2008 and 2012, inclusive.

Finally, NSAIDs usage was also investigated concurrently with colic and orthopaedic disease, to describe which NSAIDs are most commonly used with these conditions. The dataset was subsequently searched using the methodology described in Chapter 3 to identify records referring to colic and orthopaedic disease. A colic episode was defined as such when colic was reported on a given day for a give animal. When colic was reported in two consecutive days in the same animal, these were counted as two colic episodes as these could have required a repeated administration of NSAIDs. Administration of each drug was classified as related to the relevant disease episode if given on the same day that the disease was reported. Similarly an orthopaedic episode was defined as such when an orthopaedic condition or procedure was reported for a given animal. When the orthopaedic condition was reported in two consecutive days in the same animal, these were counted as two episodes as these could have required a repeated administration of NSAIDs. Administration of each drug was classified as related to the relevant orthopaedic episode if given on the same day that the disease was reported.

The study was approved by Ethics and Welfare Committee of the School of Veterinary Medicine at the University of Glasgow.

4.2.3 Results

4.2.3.1 General prevalence of non-steroidal anti-inflammatory drug usage

The prevalence of horses receiving a NSAID at least once in the records, during the 27 year period, was calculated. A total of 40,350 (28.6%; 95% CIs: 28.4-28.8%) animals received NSAIDs at least once out of the total of 141,543 individual animals included within the dataset.

The NSAIDs most frequently used were phenylbutazone/suxibuzone and flunixin meglumine. The prevalence of usage was 18.2% (95% CIs: 18.0-18.4%) for phenylbutazone and 6.1% (95% CIs: 6.0-6.2%) for suxibuzone. For flunixin meglumine estimated prevalence of use was 8.3% (95%CIs: 8.1-8.4%). Other NSAIDs that were less frequently used included metamizole (1.82%, 95% CIs: 1.75-1.89%) and meloxicam (1.05%, 95% CIs: 1.00-1.10%). Prevalence of use for ketoprofen, meclofenamic acid, eltenac, carprofen, aspirin, firocoxib and vedaprofen was less than 1% for each of these drugs and altogether accounted for 1.1% of all NSAIDs administered. Precise details on prevalence of use are reported in Table 4.1.

Table 4.1: Prevalence of NSAIDs usage in an equine population between 1987 and 2013 in the United Kingdom. Column "animals" refers to number of equids receiving each drug in the population of 141,543 equids.

Drug	Animals	Prevalence	95%CIs
Phenylbutazone	25626	18.17	17.97-18.37
Flunixin Meglumine	11649	8.26	8.12-8.40
Suxibuzone	8578	6.08	5.96-6.21
Metamizole	2565	1.82	1.75-1.89
Meloxicam	1484	1.05	1.00-1.11
Ketoprofen	796	0.56	0.53-0.60
Meclofenamic Acid	432	0.31	0.28-0.34
Eltenac	108	0.08	0.06-0.09
Carprofen	102	0.07	0.06-0.09
Aspirin	72	0.05	0.04-0.06
Firocoxib	40	0.03	0.02-0.04
Vedaprofen	31	0.02	0.02-0.03
TOTAL	**40350**	**28.6**	**28.37-28.84**

TOTAL: horses receiving any NSAID in the dataset.

Records between 2008 and 2012, the period to determine yearly prevalence of NSAID use, were available for a total of 80,083 animals of which NSAID usage was recorded in 23,328 patients (prevalence: 29.1%; 95% CIs: 28.8-29.4%). The number of horses receiving each drug in each year is summarised in Table 4.2.

The prevalence of usage for each year (Table 4.3) highlights that phenylbutazone/suxibuzone and flunixin meglumine remain the NSAIDs most commonly used. The prevalence of use has not changed substantially over this 5-year period despite firocoxib being added to the market in 2008. Meloxicam was also introduced to the British veterinary market in 2001 and between 2008 and 2010 its use slowly increased, before the annual prevalence seems to have plateaued at around 1.4%.

Table 4.2: Number of animals receiving NSAIDs between 2008 and 2012 in the United Kingdom (please note that a horse could have received more than one NSAID in the same year and its records could span over multiple years)

Drugs	2008	2009	2010	2011	2012	2008-2012
Phenylbutazone	3741	3361	3375	3403	3353	13859
Flunixin Meglumine	1850	2023	2090	1977	1748	8417
Suxibuzone	1692	1821	1969	2083	1784	7552
Metamizole	518	492	496	546	504	2271
Meloxicam	105	217	353	392	351	1330
Ketoprofen	116	102	78	79	64	419
Meclofenamic Acid	0	0	0	0	0	0
Eltenac	0	0	0	0	0	0
Carprofen	23	9	3	5	3	42
Aspirin	3	12	15	22	10	60
Firocoxib	1	6	5	12	12	36
Vedaprofen	1	0	4	1	0	6
NSAIDs	6063	6012	6314	6385	5867	23328
TOTAL	27266	25946	27195	27245	24805	80083

NSAIDs: total number of animals receiving at least one NSAID in each year; TOTAL: total number of animals in each year.

Table 4.3: Prevalence (%) of usage for each NSAID in each year between 2008 and 2012 in the United Kingdom.

Drug	2008	2009	2010	2011	2012	2008-2012
Phenylbutazone	13.72	12.95	12.41	12.49	13.52	17.31
Flunixin Meglumine	6.79	7.8	7.69	7.26	7.05	10.51
Suxibuzone	6.21	7.02	7.24	7.65	7.19	9.43
Metamizole	1.95	1.90	1.82	2.00	2.03	2.84
Meloxicam	0.39	0.84	1.3	1.44	1.42	1.66
Ketoprofen	0.43	0.39	0.29	0.29	0.26	0.52
Meclofenamic Acid	0	0	0	0	0	0
Eltenac	0	0	0	0	0	0
Carprofen	0.08	0.03	0.01	0.02	0.01	0.05
Aspirin	0.01	0.05	0.06	0.08	0.04	0.07
Firocoxib	0	0.02	0.02	0.04	0.05	0.04
Vedaprofen	0	0	0.01	0	0	0.01
NSAIDs	22.24	23.17	23.22	23.44	23.65	29.13

NSAIDs: prevalence of any NSAID in each year.

4.2.3.2 Prevalence of non-steroidal anti-inflammatory drug use with colic and orthopaedic disease

The database included a total of 38,720 days on which colic was mentioned in a clinical record (colic days) from 15,747 animals and 260,989 days on which orthopaedic was mentioned in a clinical record (orthopaedic days) from 55,728 animals (Table 4.4). The most frequently used NSAID for colic cases was flunixin meglumine (3830 days) followed by phenylbutazone (2578 days) and metamizole (2552 days). For orthopaedic disease the most commonly used NSAID was phenylbutazone (20079 days), but a large proportion of cases received also suxibuzone (7586 days) and flunixin meglumine (5290 days). Meloxicam was administered for 2552 colic days and 295 orthopaedic days, while ketoprofen was administered on 310 colic days and on 210 orthopaedic days. Firocoxib was administered only in 2 colic days and 19 times for orthopaedic days. These results are summarised in Table 4.4.

The prevalence of use of other NSAIDs licensed for use in veterinary patients was low (1.1% of the total usage of NSAIDs) and little inferences can be made on use of these drugs.

Table 4.4: NSAIDs usage in colic and orthopaedic cases; Prevalence values are reported as % of the affected population

Drug	*Colic* Episodes	n=38,720 Prevalence (95%CIs)	*Orthopaedic* Episodes	n=260,989 Prevalence (95%CIs)
Phenylbutazone	2578	6.66 (6.41-6.91)	20079	7.69 (7.59-7.80)
Suxibuzone	490	1.27 (1.16-1.38)	7586	2.91 (2.84-2.97)
Flunixin meglumine	3830	9.89 (9.6-10.19)	5290	2.03 (1.97-2.08)
Firocoxib	2	0.01 (0.00-0.02)	19	0.01 (0.00-0.01)
Ketoprofen	310	0.80 (0.72-0.89)	210	0.08 (0.07-0.09)
Metamizole	2552	6.59 (6.35-6.84)	295	0.11 (0.10-0.13)
Meloxicam	379	0.98 (0.89-1.08)	816	0.31 (0.29-0.33)

Colic: number of total days on which colic is mentioned; *Orthopaedic*: number of total days orthopaedic disease is mentioned; Episodes: number of days on which each drug was mentioned in the clinical record at the same time as colic and orthopaedic disease respectively; 95% CIs: 95% Confidence Intervals.

4.2.4 Discussion

The results of this study demonstrate that phenylbutazone and suxibuzone, followed by flunixin meglumine were the most commonly used NSAIDs in the United Kingdom equine veterinary practices used in the current study between 2008 and 2013. The use of different types of NSAIDs remained relatively constant. Only a slight increase in the use of meloxicam between 2008 and 2010 was identified, which although it was a 3-fold increase, accounted for only 246 administrations.

Suxibuzone, although more palatable, is similar to but generally slightly more expensive than phenylbutazone. Both drugs are licensed in the United Kingdom for treatment of inflammation and pain associated with musculoskeletal disease. Suxibuzone is converted to phenylbutazone and oxyphenbutazone once absorbed and has been found to be bioequivalent to phenylbutazone (Jaraiz et al., 1999). The prevalence of use of phenylbutazone and suxibuzone together accounted for 24.3% of all NSAIDs in this study. These drugs were commonly used to treat pain and inflammation associated with orthopaedic disease. However, a significant proportion of cases with visceral pain also received phenylbutazone. This finding was somewhat unexpected as the use of phenylbutazone in colic cases is off-label. Nevertheless, it appears that many equine practitioners

prefer to use phenylbutazone despite the fact that several other NSAIDs are licensed for the management of colic in the horse. These include metamizole, flunixin meglumine, meloxicam and ketoprofen. The reason for choosing phenylbutazone might be due to a blander, but effective inhibition, of clinical signs compared to flunixin meglumine, and therefore use of phenylbutazone would be less likely to delay referral of colic cases requiring surgical intervention. While peer-reviewed evidence to support this belief is lacking, practice of off-label administration of phenylbutazone to colic cases appears well established in the United Kingdom. Perceived lack of efficacy, availability and cost of meloxicam and ketoprofen might explain their reduced use.

Flunixin meglumine was the second most common NSAID used in the United Kingdom equine population that was studied and was a common choice for treatment of visceral pain but also to a lesser extent to treat musculoskeletal pain and inflammation. While phenylbutazone is advocated for treatment of orthopaedic pain, flunixin meglumine is generally preferred for the management of abdominal discomfort (Dowling, 2010). In the United Kingdom Veterinary Surgeons are obliged to follow the cascade by which the off-label use of phenylbutazone for the management of colic could be justified only in the scenario where other licenced products would not be available in an emergency situation, such as with some colic cases.

“Buscopan Compositum” is the only preparation containing metamizole available for use in horses in the United Kingdom, which is a combination with the spasmolytic butylscopolamine. Since a few preparations under the brand name of “Buscopan” have been available in the United Kingdom, precise estimation of metamizole usage was troublesome. In colloquial terms veterinary surgeons might refer to the use of “buscopan” regardless of whether this was the formulation including metamizole (“Buscopan Compositum”) or just butylscopolamine (“Buscopan 20”,

"Buscopan Ampoules"). These preparations differ in the volume to be administered so that for a typical 500kg horse the volume of "Buscopan Compositum" should be 25ml and for "Buscopan 20" only 7.5ml. "Buscopan Ampoules" have now been removed from the United Kingdom market. For the construction of the inclusion dictionary for metamizole, only the formulation referring to "Buscopan Compositum" was considered. This might have resulted in the underestimation of the use of metamizole in our population. In fact, in several instances where the volume of drug administered was recorded, this appeared too large to be just the single butylscopolamine preparation (i.e. more than 20ml) in cases where the veterinary surgeon simply referred to the preparation as "Buscopan". However, in first opinion ambulatory settings, and indeed in many of the clinical records available for this study, information is often not available regarding the precise weight of the animal and it would be difficult to use the dose as an indicator to which "Buscopan" preparation the veterinary surgeon was referring. The lack of clarity between preparations recorded as being used in the EMRs suggests that data on the prevalence of use of metamizole is to be interpreted with caution.

Meloxicam was introduced to the equine veterinary market in 2001 and the data presented here appear to suggest that its use remains limited in the United Kingdom. Meloxicam is licensed for treatment of pain from both musculoskeletal and visceral in origin as an oral and intravenous preparation. The oral preparation licensed for use in horses is licensed only for treatment of orthopaedic conditions. The licenced dose is 0.6mg/kg every 24 hours while pharmacokinetic studies suggest that a twice daily administration would be more appropriate due to a faster plasma clearance in the horse than in other species (Toutain et al., 2004). The recommendation of using a sub-optimal dose interval might have resulted in a reduced efficacy and loss of faith in this drug by veterinary surgeons, which might explain the overall low prevalence of use. The fact that meloxicam has been used more frequently with orthopaedic cases

than with colic cases might also reflect the perception of the drug being less effective than other NSAIDs and therefore less suitable for an emergency situation, when controlling pain and inflammation might be more critical. Alternatively this might also reflect the use of the oral preparation, which is licensed only for orthopaedic disease. Differentiating oral and intravenous preparations from the dataset would have been troublesome because of the lack of a persistent definition of either product in the data. Therefore only the total use of meloxicam was included.

Ketoprofen is licensed for treatment of pain associated with musculoskeletal disorders and colic in the horse in the United Kingdom. This drug was also used in a smaller proportion of cases, only slightly more frequently for the treatment of visceral pain than orthopaedic disease.

Since a firocoxib preparation licenced for use in the horse was introduced in March 2008 use of firocoxib has seen a limited increase in the United Kingdom. This drug is licensed for treatment of pain and inflammation associated with osteoarthritis in horses and the course of administration should not exceed 14 days. In our dataset the use of firocoxib was limited to a total of 40 animals and few conclusions can be drawn from the data, other than it does not yet seem to have been well accepted in equine practice in this country, at least in the practices that contributed to the data investigated during the current study.

Aspirin is used mostly to control hypercoagulative states, particularly in association with thrombophlebitis or for the treatment of laminitis. The use of aspirin appeared to be very minor in this first opinion population from the United Kingdom and mostly focused in the data from two practices (84% of all aspirin used), which likely reflects practice protocol rather than an overall use. This might suggest that in the United Kingdom aspirin might be perceived as ineffective by veterinary surgeons in first

opinion equine practice or that conditions that might require treatment with aspirin are referred to referral centres.

Preparations of Carprofen and Vedaprofen are also available for use in horses in Britain but their use also remains minimal. Other drugs such as meclofenamic acid, eltenac are not available for use in the horse in the United Kingdom anymore and any appearance in the data used in the current study referred to older records so their use is of no relevance today.

Since the analysis aimed at detecting presence of colic or orthopaedic disease in the records on the same day that each NSAID was administered one might argue that a relationship between drug administration and development of clinical signs exists. It was not possible to confirm that these drugs were administered exclusively after (i.e. as a treatment) the clinical signs in all cases. However, there was no record that veterinary surgeons were concerned that these drugs had induced abdominal discomfort or orthopaedic disease. If this was the case one might expect that a patient would be examined more than once on a given day. Since the date was used to differentiate appointments it was not possible to tell if horses received more than one visit on the same day. In the vast majority of these cases the record referred to the drug being dispensed after the episode of abdominal discomfort or to address an underlying concurrent orthopaedic condition. However it was not possible to identify cases when the horse received an NSAID in the morning to manage a certain condition and then developed colic or other clinical signs and had to be re-examined. With this scenario both examinations would have had the same date in the system and could have not been differentiated by the analysis. Due to the retrospective nature of the data it was not possible to make any inference about the temporal relationship between drug administration and recording of colic of orthopaedic conditions. It is most likely that data about drug administration and clinical signs were

added to the PMSS at the same point in time, most often after the veterinary surgeon had returned to their practice a few hours later.

The overall dataset spanned a period of 26 years between 1987 and 2013. However, inter-year comparison of drug usage was performed only between 2008 and 2012, inclusive. This period included the largest amount of data (61.2% of the data with a valid date; 1,623,409 rows of data) and data was available from all practices so that inter-year comparison was less likely to be biased by the absence of one or more practices. Including only the later years reduced the bias that could have derived from behaviour changes present over a longer period, but might still reflect some individual practice behaviour, particularly that of the largest practices.

This study did not aim to describe the prevalence of colic or orthopaedic disease in the United Kingdom horse population. The data on colic and orthopaedic disease refers to the number of times these conditions were mentioned on different days in the records. Therefore a horse could have been examined multiple times for a chronic lameness and this would have appeared as multiple lameness episodes rather than recurrence of an on-going condition. The aim of this study was to describe NSAID usage and not the actual length or prevalence of a colic episode or of orthopaedic disease.

In this study it was not possible to determine with a reasonable certainty the duration of NSAID treatment. The dataset contains details of when the veterinary surgeon inserted information in the PMSS about drug usage. For example, in the case where a 5-days course is prescribed, drugs may be dispensed to be administered by the owner; in such a scenario only the date the drugs are dispensed will be included in the record. Dates available referred to the date the drugs were dispensed to the owner but did not necessarily reflect the actual date of drug administration to the

animal. More precise information on length of treatment could have been mined from the free-text records, where available. This would have required the lengthy process of defining inclusion, exclusion and re-inclusion dictionaries and would have required more time than was available for the study. Further, dictionaries definition is likely to be very time consuming as multiple drugs might be dispensed at the same time, with differing lengths of treatment, which could be recorded in the same row of data. This would translate in the need for the dictionary to include the wide range of combination of words necessary to minimize mixing drugs and course of treatment that are written in the same sentence. The same is true also for the amounts of drug being dispensed. In conclusions, time available for analysis limited the ability of the analysis to describe length of treatment or dosages used in practice, but this should be the aim of future studies.

4.3 Non-steroidal anti-inflammatory drug usage in North America

4.3.1 Introduction

The prevalence of usage of NSAIDs in North America has not been described before in a large population of horses. Knowing the prevalence of usage of each NSAID could assist clinicians and researchers to interpret research data appropriately. For example, knowing how many horses receive NSAIDs, or combinations of NSAIDs, and approximately for how long NSAIDs are prescribed could help understand the significance of the prevalence of side effects in treated patients. Further, as a considerable effort, both economical and professionally, has been committed to develop new, safer drugs over the past decade evaluation of how drug usage has changed could provide some indication as to how veterinary practice has been modified to include their use.

The FDA[14] (Food and Drug Administration) is the body responsible for the regulation of veterinary drug usage in the United States. In Canada this role is played by Health Canada[15]. The information on each drug and indication for their use in veterinary patients is easily obtainable from their respective websites. In these countries NSAIDs are prescription drugs, meaning that they can be only purchased under veterinary guidance. Nevertheless, they are often administered to horses without veterinary supervision directly by owners and trainers.

The dataset described in Chapter 3 includes the EMRs from 225,777 animals that have a correct date of data entry and provides a valuable insight of how NSAIDs have been used in equine practice in North America.

4.3.2 Materials and methods

The dataset of EMRs from 312,610 equids from North America between 1994 and 2013, which include the 225,777 horses with a reliable date of data entry to the PMSS system and the 86,833 with a date that was not reliable, was analysed with the methods described in detail in Chapter 3. Briefly, the list of the words included in the dataset was used to identify terms that might identify NSAID usage. This included commercial brand names of NSAIDs as well as pharmaceutical names, colloquial names, financial codes and abbreviations. All NSAIDs were identified using the methodology described in Chapter 3. The inclusion dictionary was created using terms included in the dataset that identify NSAIDs in general and also in particular the following drugs: flunixin meglumine, phenylbutazone, suxibuzone, meloxicam, firocoxib, ketoprofen, metamizole, aspirin, diclofenac, meclofenamic acid, carprofen, deracoxib and vedaprofen (Appendix 4).

---

[14] http://www.accessdata.fda.gov/scripts/animaldrugsatfda/
[15] http://www.hc-sc.gc.ca/index-eng.php

Prevalence and 95% CIs were then calculated as previously described (Wilson, 1927) for the equine population in the whole dataset. The yearly prevalence of drug usage was calculated between 2008 and 2012, inclusive as this time period included the largest proportion of data (75.3%; 8,807,287 rows of data).

Finally NSAIDs usage was investigated concurrently with colic and orthopaedic disease, to describe which NSAIDs are most commonly used with these conditions. A colic day was defined as such when colic was reported in a given day for a given animal. When colic was reported in two consecutive days in the same animal, these were counted as two colic days as these could have required a repeated administration of NSAIDs and this study aimed to describe the NSAID used and not the actual length of colic. Administration of each drug was classified as related to the disease episode if given on the same day.

4.3.3 Results

The prevalence of horses receiving NSAIDs at least once in the records was calculated. A total of 115,446 (36.9%; 95% CIs: 36.8-37.1%) and 91,799 (40.7%; 95% CIs: 40.5-40.9%) animals received NSAIDs at least once out of the total of 312,610 and 225,777 with a reliable date respectively.

The NSAIDs most frequently used were phenylbutazone and flunixin meglumine. In the group of 225,777 the prevalence of usage was 28.5% (95% CIs: 28.4-28.7%) for phenylbutazone and 23.2% (95% CIs: 23.0-23.3%) for flunixin meglumine. Other NSAIDs that were seldom used included firocoxib, ketoprofen and metamizole. The prevalence for firocoxib and ketoprofen administration was 3.7% (95% CIs: 3.6-3.8%) and 3.7%(95% CIs: 3.6-3.8%) respectively, while Diclofenac was used in 1.92% (95% CIs: 1.8-2.0%) of horses, but this was only with a commercial topical preparation.

The prevalence of use for aspirin, metamizole, meclofenamic acid, vedaprofen, carprofen, meloxicam, deracoxib and suxibuzone was less than 1% for each of these drugs. These results are summarised in Table 4.5.

Table 4.5: Prevalence of NSAIDs usage in an equine population in North America. Column "Total" refers to number of horses receiving each drug in the population of 312,610 equids, while the "Valid Date" refers to the population of 225,777 equids with EMRs with a valid date of data entry in the management system. Prevalence and 95% CIs data is calculated on the population with a valid date.

Drug	Total	Valid Date	Prevalence	95%CIs
Phenylbutazone	82002	64450	28.55	28.36-28.73
Flunixin Meglumine	63430	52287	23.16	22.99-23.33
Firocoxib	8573	8375	3.71	3.63-3.79
Ketoprofen	9437	8303	3.68	3.60-3.76
Diclofenac	5045	4331	1.92	1.86-1.98
Aspirin	2022	1631	0.72	0.69-0.76
Metamizole	1767	1495	0.66	0.63-0.70
Meclofenamic Acid	722	622	0.28	0.25-0.30
Vedaprofen	532	443	0.20	0.18-0.22
Carprofen	97	86	0.04	0.03-0.05
Meloxicam	51	41	0.02	0.01-0.02
Deracoxib	7	5	0	0.00-0.01
Suxibuzone	0	0	0	0.00-0.00
TOTAL	**115,446**	**91,799**	**40.66**	**40.46-40.86**

TOTAL: horses receiving any NSAID in the dataset.

Records between 2008 and 2012 were available for a total of 171,191 animals of which NSAID usage was recorded in 68,339 patients (prevalence: 39.9%; 95% CIs: 39.7-40.1%). The number of horses receiving each drug in each year is summarised in Table 4.6.

Table 4.7 summarises the prevalence of usage of each individual NSAID and of all NSAIDs altogether in each individual year and overall between 2008 and 2012, inclusive.

Table 4.6: Number of animals receiving NSAIDs between 2008 and 2012 in North America (please note that a horse could have received more than one NSAID in the same year and its records could span over multiple years and the sum of horses in each individual years will be greater than the number of horses receiving the drugs in the 5 year period).

Drugs	2008	2009	2010	2011	2012	2008-2012
Phenylbutazone	8322	13432	14271	14024	14195	48899
Flunixin Meglumine	7167	9880	9980	10138	10148	37067
Firocoxib	795	1046	1541	2219	2865	6572
Ketoprofen	1010	1143	1348	1565	1765	5571
Diclofenac	465	641	787	797	819	3264
Aspirin	238	359	359	306	218	1217
Metamizole	134	235	274	265	267	1112
Suxibuzone	63	90	123	112	107	468
Meclofenamic Acid	39	69	78	80	91	336
Vedaprofen	7	15	16	10	18	61
Carprofen	1	8	16	8	4	30
Meloxicam	0	1	2	1	0	3
Deracoxib	0	0	0	0	0	0
NSAIDs	13150	19470	20157	20290	20967	68568
TOTAL	**42862**	**59221**	**58109**	**56844**	**57073**	**171191**

NSAIDs: total number of horses receiving at least one NSAID in each year; TOTAL: total number of horses in each year.

Table 4.7: Prevalence of usage for each NSAID in each year between 2008 and 2012 in North America.

Drug	2008	2009	2010	2011	2012	2008-2012
Phenylbutazone	19.42	22.68	24.56	24.67	24.87	28.56
Flunixin Meglumine	16.72	16.68	17.17	17.83	17.78	21.65
Firocoxib	1.85	1.77	2.65	3.9	5.02	3.84
Ketoprofen	2.36	1.93	2.32	2.75	3.09	3.25
Diclofenac	1.08	1.08	1.35	1.4	1.44	1.91
Aspirin	0.56	0.61	0.62	0.54	0.38	0.71
Metamizole	0.31	0.4	0.47	0.47	0.47	0.65
Meclofenamic Acid	0.15	0.15	0.21	0.2	0.19	0.27
Vedaprofen	0.09	0.12	0.13	0.14	0.16	0.2
Carprofen	0.02	0.03	0.03	0.02	0.03	0.04
Meloxicam	0	0.01	0.03	0.01	0.01	0.02
Deracoxib	0	0	0	0	0	0
Suxibuzone	0	0	0	0	0	0
NSAIDs	30.68	32.88	34.69	35.69	36.74	40.05

NSAIDs: prevalence of any NSAID in each year.

The database of records from the 225,777 equids, with a valid date in the dataset, included a total of 96,614 single days on which colic was recorded affecting 21,682 animals and 346,165 days on which an orthopaedic condition was recorded affecting 94,840 animals. The most used NSAID for colic cases was flunixin meglumine, followed by phenylbutazone (Table 4.8). For orthopaedic disease the most commonly used NSAID was phenylbutazone, but a large proportion of cases received flunixin meglumine. Firocoxib and ketoprofen were administered

predominantly on the same day of an orthopaedic episode and only to a lesser extent on the same day of a colic episode.

The prevalence of use of other NSAIDs approved for use in equine patients was minor (0.3% of the total usage of NSAIDs).

Table 4.8: NSAIDs usage on the same day of colic and orthopaedic cases in North America; Prevalence values are reported as % of the affected population. Not all episodes of colic or orthopaedic disease received a NSAID

	Colic	96614	*Orthopaedic*	346165
Drug	**Episodes**	**Prevalence (95%CIs)**	**Episodes**	**Prevalence (95%CIs)**
Phenylbutazone	3422	3.54 (3.43-3.66)	65550	18.94 (18.81-19.07)
Flunixin meglumine	20723	21.45 (21.19-21.71)	39542	11.42 (11.32-11.53)
Firocoxib	796	0.82 (0.77-0.88)	8878	2.56 (2.51-2.62)
Ketoprofen	259	0.27 (0.24-0.30)	8745	2.53 (2.47-2.58)

Colic: number of total colic episodes; Orthopaedic: number of total orthopaedic disease episodes; Episodes: each number of episode treated with a specific NSAID; 95% CIs: 95% Confidence Intervals.

4.3.4 Discussion

The findings of this study indicate that phenylbutazone and flunixin meglumine are the most commonly used drugs in equine veterinary practice in North America.

Between 2008 and 2012, the use of phenylbutazone had increased by 5%. This might be consistent with the reduced cost associated with this drug, which may have influenced choice of NSAID during the concurrent difficult economic conditions. Compounded phenylbutazone can be very cheap (~10¢ per 1 gram/dose or even less). During a period of economic recession a greater proportion of owners might prefer to manage non-life threatening conditions with phenylbutazone rather than spending money on more expensive investigations and treatments. Alternatively other unidentified factors might have contributed to the increase in use of phenylbutazone in North America in this time period. A relatively large proportion (13.6%) of horses received phenylbutazone on the same day of colic and this could be the result of some veterinarians preferring

phenylbutazone in colic cases as well as cases that had by chance received phenylbutazone on the same day to treat another condition (eg. orthopaedic disease) but before the start of the colic signs.

The use of flunixin meglumine remained otherwise unchanged despite the higher cost of this drug in comparison to phenylbutazone. This might be because, despite the increased cost of this drug, its perceived efficacy makes it the best choice in situations where pain or inflammation is severe. Flunixin meglumine was by far the most common drug selected for treatment of abdominal discomfort of those horses receiving NSAID on the same day as colic over 80% received the drug. This may be because flunixin meglumine is the only drug licensed for the management of colic in the United States of America and Canada.

Since the introduction of firocoxib as a licensed preparation for equine use in 2006 its use has seen a steady increase. This drug is licensed (Equioxx, Merial Limited, USA) in the United States for treatment of pain and inflammation associated with osteoarthritis in horses and the course of administration should not exceed 14 days. In Canada, no product approved for use in the horse is available, but veterinarians often resorted to a product (Previcox®, Merial Limited, Canada) licensed for use in dogs. Of the horses that suffered colic, some had also received firocoxib on the same day and this summed up to 8.2% of the total of those that had received this drug. Firocoxib is licensed for treatment of pain associated with osteoarthritis in the horse in North America. This drug was also used in 0.9%of cases, on the same day as a colic episode. Following manual evaluation of those records revealed that the drug had been dispensed for treatment of a concurrent underlying orthopaedic problem on the day colic was then diagnosed.

Ketoprofen is licensed for treatment of pain associated with musculoskeletal disorders in the horse in North America. This drug was

also used in a smaller proportion of cases, particularly in orthopaedic cases, but in a number (2% of those receiving ketoprofen) of cases it had also been administered on the same day as an episode of colic. Following manual evaluation of those records it was apparent that almost invariably these records refer to instances where these drugs had been dispensed for treatment of a concurrent underlying orthopaedic problem at the time the mild/transient episode of colic was diagnosed.

Aspirin is used mostly to control hypercoagulative states, particularly in association with thrombophlebitis or for the treatment of laminitis. In our dataset further common use of aspirin appeared to be treatment of inflammation and pain associated with ophthalmic disease. The use of aspirin appeared mostly focused in the data from two practices (70% of all aspirin used), which likely reflects practice protocol rather than an overall use. Aspirin acts by irreversibly preventing arachidonic acid-induced platelet aggregation and therefore prevents intravascular thrombus formation (Cambridge et al., 1991). Since aspirin has been shown to prevent platelet aggregation in horses (Heath et al., 1994) a theoretical beneficial effect of aspirin in cases of laminitis and thrombophlebitis has been proposed (Sellon and Wise, 2010). Nevertheless, evidence that aspirin improves outcome in cases of laminitis or thrombophlebitis in horses remains scarce. This might, in part, explain the variable use of aspirin between practices.

The prevalence of use of metamizole is also lower in this part of the world compared with Europe. In the past metamizole was used as an antipyretic (eg. dipyrone), but in the United Kingdom it is still available in combination with a spasmolytic, butylscopolamine bromide (Buscopan® Compositum, Boehringer Ingelheim, United Kingdom), licensed for the treatment of abdominal pain in horses. This combined preparation is not licensed in North America and since 1995 metamizole has been withdrawn from the US market. In Canada, metamizole is licensed for use in the

horse as an antispasmodic, analgesic, antipyretic and anti-inflammatory (Dipyrone 50%, Vétoquinol, Canada). Nevertheless, Canadian practices accounted only for 5.5% of the overall amount of metamizole recorded in the North American database. Therefore, it appears that the supposition that old stockpiles of the drug occasionally appear in US practices may be true (Payne et al., 1999); alternatively veterinary practices might import this drug from other neighbouring countries, such as Canada. In fact, products containing metamizole were referred to in data from all practices, although the vast majority (86.7%) of the metamizole used was from 3 practices, which were all in the United States.

The prevalence of use of other NSAIDs for the veterinary market was low (<1%). The preparation of diclofenac is a topical preparation (Surpass®), which is used for treatment of musculoskeletal disorders and it is used mostly in the scenario of superficial inflammatory conditions, such as muscle or tendinous injuries.

Treatment with suxibuzone saw a peak (81% of total usage) in administration in 2009. The use of this drug was recorded almost entirely at one practice (96% of all recorded uses). Suxibuzone is not licensed in North America and the use of this drug might just reflect that one practice had imported some suxibuzone from abroad.

4.4 Discussion

These two studies report the prevalence of use of NSAIDs in two different continents, which include countries with socio-economic as well as legislative differences. Socio-economic differences might affect the preference of certain drugs over others, as well as acceptance of innovation such as new drugs being licensed with different safety profiles. Cost and tradition might also have played a role in the comparative differences in use of NSAIDs in the United Kingdom and North America.

The prevalence of usage for each year (Tables 4.3 and 4.7) highlights that phenylbutazone and flunixin meglumine remain the most commonly used NSAIDs in both the United Kingdom and North America. In relative terms phenylbutazone is used more commonly in North America than it is in the United Kingdom. The explanation for this finding may lay in the availability of Suxibuzone only in the United Kingdom. Suxibuzone is very similar to phenylbutazone and prevalence of use of phenylbutazone in North America should be compared to the combined prevalence of the use of phenylbutazone and suxibuzone in the United Kingdom. Further social factors might contribute to the different prevalence of use of phenylbutazone between countries. Compared to North America, owners or veterinary surgeons in the United Kingdom might be more inclined to determine the cause of disease, and treat conditions appropriately, or equally opt for non-conventional treatments for palliative care to use in place of phenylbutazone. These theories remain unconfirmed and further studies would be required to explain this finding. Also the finding of the off-label use of phenylbutazone to manage visceral pain in horses is in line with the author's personal experience of first opinion practice in the United Kingdom and United States. The finding of this study could prompt legislative bodies of each country to re-evaluate the labelled use of phenylbutazone if appropriate. If the use of phenylbutazone were to be considered contra-indicated for the management of abdominal discomfort in horses, then a campaign aimed at educating veterinary surgeons and limit this practice would be warranted.

Firocoxib was used comparatively more in North America than in the United Kingdom where its use appears vey limited despite being available for use in horses since 2008. Firocoxib was introduced to the North American veterinary market in 2006 and has seen a 2.7-fold increase between 2008 and 2012. However it still remains far less commonly used than phenylbutazone or flunixin meglumine.

Ketoprofen is also used more commonly in North America than in the United Kingdom. The reason for this finding is difficult to explain particularly as ketoprofen is only licensed for musculoskeletal disease in North America while it is licensed for both musculoskeletal disease and colic in the United Kingdom.

Diclofenac is also commonly used in North America, and this is in the form of a topical preparation, which is not licensed in the United Kingdom where no diclofenac is available.

Legislative differences certainly influenced which drugs were available for veterinary surgeons in each country. For example, metamizole was removed from the market in the United States, while in Canada the preparation is still available as an antipyretic and anti-inflammatory. In the United Kingdom it is available only in combination with butylscopolamine.

The overall scarce usage of meloxicam in both countries might result from a perceived lack of efficacy of this drug explained by current evidence suggesting that the labelled once-a-day administration results in sub-therapeutic serum concentrations of the drug (Lees et al., 1991; Toutain et al., 2004) and that more frequent administration might be more efficacious while maintaining safety (Naylor et al., 2014). The evidence of a reduced of usage of this drug with a safer profile *in vitro* compared to phenylbutazone and flunixin meglumine should prompt legislative bodies to reconsider and up-date the instructions for its use. Updating the dosing interval would improve drug efficacy and could help reduce the prevalence of toxicity (Naylor et al., 2014). This process would be expensive (at least in the United Kingdom) and currently other drugs licensed for once a day administration (many formulations of trimethoprim sulphonamide) are generally administered twice daily to respect the

pharmacokinetic of this antimicrobial (Peck et al., 2002), despite the lack of up-to-date legislation.

While the use of carprofen is negligible in the United Kingdom, in North America the relative usage of carprofen has doubled between 2008 and 2012. This is surprising as there is no carprofen preparation licensed for the use in horses in either Canada or in the United States. While this increase might seem conspicuous, it is explained overall by the very small number of horses treated with this drug (<1%) and has little clinical relevance.

Aspirin also was relatively more used in North America than it was in the United Kingdom. Aspirin is generally limited to treat or prevent diseases resulting from increased platelet aggregation such as thrombosis. Common pathological conditions resulting from thrombus formation include jugular thrombophlebitis and laminitis (Sellon and Wise, 2010). This limited spectrum of conditions combined with the fact that no licensed aspirin preparation is present in any of these countries might explain the low prevalence of use of this drug.

Comparison between North America and United Kingdom suggests that NSAIDs usage in general is more common in United States and Canada than in the United Kingdom. While over 40% of horses in the North American dataset appear to have received NSAIDs at some point, in the United Kingdom this figure reached 28%. The reason for increased NSAID usage in North America might be a reflection of a different attitude of equine veterinary practice between continents. Similar to the difference in phenylbutazone usage between United Kingdom and North America, it is possible that the greater NSAID usage in North America is explained by a different attitude of clients towards disease. It is possible that in the United Kingdom clients request veterinary assistance only when required or British owners and veterinary surgeons might tend to investigate the

cause of disease for a more targeted treatment rather than opt for palliative care with NSAIDs. Alternatively in North America clients may be more proactive in seeking veterinary advice and treatment or veterinary surgeons might simply have the tendency to prescribe NSAIDs even when the animal does not have overt signs of discomfort. The real cause for this difference remains however undetermined.

The data also allows comparison of the attitude of the veterinary market towards new medications. The study offers a good example of how veterinary surgeons adapt their clinical practice to the introduction of new pharmaceuticals such as firocoxib and how this might vary between countries. In North America, the use of firocoxib has increased rapidly and was still on the increase in 2012, while in the United Kingdom this drug is still rarely used. Reasons for this difference are likely to be multiple and include social differences, as veterinary surgeons in the United Kingdom might be more conservative and less prone to embrace new products. Further, different marketing strategies from the producer of firocoxib also might have played a role, if product promotion in North America had been more pressing. Costs also might play a role, as firocoxib is more expensive than some of the alternative products (including phenylbutazone and suxibuzone), the concurrent difficult economic climate might have limited the willingness to try new, but more expensive drugs in the United Kingdom.

This study provides data that was then used to ascertain the relationship between usage of these drugs and the risk of developing side effects. This was the aim for the following Chapter 5.

CHAPTER 5 - Use Of Big Data To Describe Relevance Of Non-Steroidal Anti-Inflammatory Drug Toxicity In Equine Practice

5.1 Introduction

5.1.1 Non-steroidal anti-inflammatory drugs toxicity

Current evidence is consistent with an increased risk of potentially life-threatening side effects with NSAIDs administration in horses, involving mainly the gastrointestinal tract and the urinary system. (Hough et al., 1999; Jones et al., 2003; Lees and Michell, 1979; Read, 1983; Snow et al., 1981, Snow et al., 1979). Inhibition of physiologic prostaglandin production that regulates tissue blood flow is believed to be the main pathophysiologic mechanism (Lees and Higgins, 1985).

Little information is available on the prevalence of side effects and whether there is any significant difference between NSAIDs with different COX selectivity. Even though these drugs are generally well tolerated, toxicity frequently occurs at the labelled dose (Andrews and McConnico, 2009; Cohen et al., 1995), therefore current recommendations are to closely monitor all equids receiving NSAIDs for early signs of toxicity (Andrews and McConnico, 2009; Jones et al., 2003). Life-threatening complications related to NSAID toxicity include right dorsal colitis and renal medullary crest necrosis (Snow et al., 1979), but these are relatively uncommon considering how frequently these drugs are used (Cohen et al., 1995; Gunson and Soma, 1983; Jones et al., 2003; Read, 1983). A *post-mortem* survey between 1979 and 1981 at the College of Veterinary Medicine of Texas A&M University identified 35 cases of renal medullary necrosis. All of the horses in this study had received phenylbutazone for periods ranging from 6 months to 12 years. Of these 20 were subjected to euthanasia for unreported reasons, whilst the remainder died from natural

causes. The overall prevalence of this condition cannot be estimated from that study as no information is available on the total number of equine *post-mortem* examinations performed in that period (Read, 1983). Another study from the same institution was aimed at describing the outcome with medical management of right dorsal colitis within the referral population of the local teaching hospital (Cohen et al., 1995). In that study only 5 cases were included in a 9-year period. The study by Jones and colleagues (2003), aimed at describing the use of ultrasonography to detect cases of right dorsal colitis, includes 5 cases in a 2 year period at the teaching hospital of the college of veterinary medicine of North Carolina State University. Although a figure for the overall populations of these two hospitals during the period of these studies is not provided, it appears reasonable to assume that the prevalence of this condition is generally low.

5.1.2 Detection of non-steroidal anti-inflammatory drugs toxicity from Electronic Medical Records

The widespread use of EMRs in veterinary practice provides an opportunity to describe drug usage, as detailed in the previous chapter, but also to estimate the prevalence of side effects.

Informatics is “the application of information and computer science technology to public health practice, research and learning” (Friede et al., 1995) and has been applied in human as well as veterinary medicine to extract information from unstructured or semi-structured free-text clinical records (Anholt et al., 2014b; Chapman et al., 2005; Chen et al., 2008; Friedlin et al., 2008; Heinze et al., 2001; Lam et al., 2007c; Roque et al., 2011; Sager et al., 1994).

To detect cases with a specific diagnosis from free text EMR, one must search for all combinations of words that might identify that diagnosis.

This approach relies on a correct diagnosis being made by the veterinarian entering the data into the EMR and therefore is likely to underestimate the real prevalence in first opinion settings, where reaching a definitive diagnosis via appropriate testing is often not feasible. However, while for this same reason it is possible that the prevalence of toxicity in some cases may be overestimated, because of the difficulty to confirm the diagnosis, some also suffer toxicity without overt clinical signs (Read, 1983). A correct diagnosis might not be reached due to either a failure to recognise toxicity or if clinical data is not recorded in the EMR. On the other hand, clinical signs identified during the clinical examination can be relatively non-specific and if used as a marker for this disease could result in an overestimation of the prevalence. This is a problem particularly in conditions for which a diagnosis cannot be easily confirmed *ante-mortem* such as renal medullary crest necrosis (Gunson and Soma, 1983), particularly when necropsy and histopathology is often not performed for cases subjected to euthanasia in the field. Right dorsal colitis is diagnosed *ante-mortem* in cases with characteristic clinical signs, clinical-pathological picture, evidence of thickened right dorsal colon with abdominal ultrasonography, history of NSAIDs administration and after the exclusion of other major causes of diarrhoea, such as bacterial colitis and antimicrobial induced diarrhoea (McGorum and Pirie, 2009; Sanchez, 2010).

Right dorsal colitis may present with one or more of several non-specific clinical signs including anorexia, weight loss, lethargy, intermittent or sporadic episodes of acute abdominal pain, pyrexia, ventral oedema and diarrhoea (Cohen et al., 1995). Even though clinical signs such as anorexia, lethargy and weight loss are most predominant (Sanchez, 2010) these are more easily recorded in hospitalised patients compared with ambulatory first opinion settings, unless they are very severe or specifically noted and reported by the owner. Episodes of abdominal pain and pyrexia are often sporadic, while ventral oedema is a feature of a

more advanced stage of the disease (Cohen et al., 1995). Abnormal faecal consistency is noticeable at the time the bedding is cleaned and is likely to be reported by owners. Therefore, diarrhoea might arguably be a more sensitive marker for this condition in first opinion ambulatory settings. Therefore, it might be a reasonable approach to identify episodes of diarrhoea that owners considered severe enough to require a veterinary examination in order to detect cases of NSAID toxicity. Finally diarrhoea may be a more suitable outcome variable to detect NSAID toxicity as it is relatively more sensitive than confirmed right dorsal colitis in first opinion settings.

It is important to note that diarrhoea is a non-specific clinical sign and many conditions are included in a differential diagnosis list for diarrhoea such as most gastrointestinal diseases, infectious colitis, parasitic disease, excessive ingestion of sand, inflammatory or infiltrative disorders of the intestine, sudden dietary change, carbohydrate overload, anaphylaxis and ingestion of other toxic substances (arsenic, cantharidin, etc.). Further, several non-gastrointestinal conditions can result in loose faecal consistency and include any condition reducing blood oncotic pressure, either from protein and albumin loss within a body cavity, such as peritonitis, pleuropneumonia or pericarditis, or outwith the body such as protein loosing nephropathy or severe dermatological conditions or burns, or from increased vascular hydrostatic pressure, in case of uncompensated congestive heart failure, intrinsic renal failure or over-zealous intravenous fluid therapy. Finally administration of other drugs, within or outwith labelled doses might also induce diarrhoea. Antimicrobials, very frequently administered to equids under veterinary care, are also a well-documented contributing factor to the development of diarrhoea (Sanchez, 2010). Therefore using diarrhoea to identify cases of right dorsal colitis would result in obtaining several false positive cases, as many cases with diarrhoea would not have right dorsal colitis. Diarrhoea also yields low specificity to identify NSAID toxicity as it presents with a wide range

of conditions resulting in a high false positive rate. On the other hand while right dorsal colitis would be very specific, therefore with a low false positive rate, many cases with mild right dorsal colitis could remain unidentified with a higher false negative rate. This highlights the importance of accounting for confounding factors, such as other conditions leading to diarrhoea, via multiple logistic regression analysis. Confounding factors should include administration of other drugs such as antimicrobials as these drugs are often administered concurrently with NSAIDs and are also a relatively common cause of diarrhoea in the horse (McGorum and Pirie, 2009). Corticosteroids share a common pathway with NSAIDs to inhibit prostaglandin production (Vane et al., 1998) and represent a confounding factor for the development of diarrhoea. However, corticosteroids are often used as a treatment for conditions inducing diarrhoea in horses as they modulate the inflammatory component that leads to diarrhoea (Barr, 2006; Mair, 1993) and corticosteroid administration should therefore be accounted for in the logistic regression analysis. Administration of laxatives and intravenous fluids should also be included as these are often administered to soften faeces (Sanchez, 2010). Some anthelmintic drugs, particularly those from the avermectin family and benzimidazole derivatives, have also been associated with colic and diarrhoea after administration (Barrett et al., 2005). As abdominal pain is a rather common clinical sign of these gastrointestinal diseases in horses, colic should also be included in the analysis to account for underlying conditions that could predispose a horse to develop diarrhoea. Further, diseases leading to colic might result in an altered barrier function of the intestinal mucosa and ultimately result in diarrhoea (Sanchez, 2010).

Renal medullary crest necrosis is often present sub-clinically and only severe cases develop severe clinical renal failure (Gunson and Soma, 1983). Further, the diagnosis is generally only confirmed during *post-mortem* examination, suggesting that the condition is likely not to be commonly recognised in the vast majority of cases (Gunson and Soma,

1983), particularly in first opinion practice. Semantically, renal NSAID toxicity might be described simply as renal failure in many cases and therefore detection of this condition by text-mining EMRs would be potentially insensitive. Several conditions can induce renal failure in horses. Renal failure is generally classified as (i) pre-renal, mostly due to decreased renal perfusion secondary to hypovolaemia and reduced cardiac output; (ii) intrinsic renal disease, including NSAID toxicity, but also nephritis, nephropathies, nephrolithiasis, kidney neoplasia; (iii) post-renal causes, generally including cystitis, urolithiasis, rupture or trauma to the bladder or urethra. While the mere presence of renal disease is generally easy to detect by the increase in creatinine and BUN concentration in blood biochemistry, confirming the origin (pre-renal, intrinsic or post-renal) is more difficult particularly in ambulatory first opinion settings, where appropriate tools to further investigate the condition may not be readily available. These include cystoscopy, a laboratory to perform urinalysis and good quality ultrasonography. Furthermore, with cases of renal medullary necrosis, it would be very difficult to reach this definitive diagnosis, as *post-mortem* and histopathology of the renal medullary crest are not readily available. As with diarrhoea, searching for renal failure might also have a higher sensitivity but a lower specificity and accounting for confounding comorbidities is also appropriate.

The studies in this chapter might also highlight how differences in drug usage between countries might also affect the results. For example, as highlighted in Chapter 4, some NSAIDs are available only in certain countries and their availability might influence the usage of other drugs as well as the condition for which these drugs are used. This might be the case for NSAIDs such as metamizole, which is available in Canada but not in the US, but in the United Kingdom it is available only in combination with a spasmolytic as a treatment of visceral pain. As equids with colic might be predisposed to develop gastrointestinal signs such as diarrhoea, it might be speculated that diarrhoea would be more common with a drug

licensed solely for gastrointestinal disease. In other words, this could be a reflection of the intrinsic increased risk for diarrhoea in this population, as well as an increased prevalence of the drug's side effects. The true contribution of the drug to the development of diarrhoea would be very difficult to estimate in these circumstances.

Further, regional differences towards usage of certain drugs might also affect the prevalence of side effects and highlight how confounding factors intrinsic to a certain geographical region influence the results of studies on a drugs' safety profile. For example, legislative differences between countries that control drug licensing might highlight differences in the safety profile of these drugs in these different countries. Ultimately this highlights the importance of interpreting research findings with caution when data is obtained from a population from a different country, with different drug licensing regulations.

The aim of this chapter was to describe the prevalence of side effects, such as diarrhoea, that might be attributed to NSAIDs administration, accounting for the administration of several other pharmaceuticals, from data obtained from two different geographical regions, the United Kingdom and North America (U.S.A. and Canada).

5.2 Non-steroidal anti-inflammatory drug toxicity in the United Kingdom

5.2.1 Introduction

The prevalence of toxicity related to NSAID administration is unknown in the United Kingdom. Two clinical studies that describe NSAID toxicity are from North America (Cohen et al., 1995; Jones et al., 2003). The only study from the British Isles is from Ireland, reporting three cases of right dorsal colitis over an unspecified period of time (Galvin et al., 2004).

Considering the frequent use of NSAIDs in equine practice, as described in Chapter 3, and the scarcity of reports of toxicity, the prevalence of side effects is expected to be very low. This might be for a number of reasons beyond a true low prevalence including difficulty in confirming the diagnosis *in vivo*, particularly in less severe cases. This is particularly true in a first opinion population, as mild cases might not be reported by the owners or because of lack of the means necessary to reach a diagnosis in a field setting.

The aim of this study was to report the prevalence of suspected NSAID toxicity. Since the prevalence of diagnosis of NSAID toxicity was expected to be very low (<1% of equids receiving NSAIDs) the study focused not only on determining the prevalence of toxicity but also on documenting if a relationship exists between a common clinical sign that could be consistent with toxicity, such as diarrhoea, and the administration of these drugs. Diarrhoea was ideal for the purpose of the study as it is easily detected by owners and is generally a cause of concern that results in a veterinary surgeon's opinion being sought.

5.2.2 Materials and methods

5.2.2.1 Prevalence of reported non-steroidal anti-inflammatory drug toxicity

The United Kingdom first opinion dataset described in Chapter 3, which included 141,543 animals (between 1987 and 2013; 2,653,695 rows of data) was analysed with the methods described in detail in Chapter 3. Briefly, the list of the words included in the dataset was used to identify terms that might identify NSAID toxicity; including cases where toxicity was suspected but not necessarily confirmed.

5.2.2.2 Diarrhoea and control population

Diarrhoea was defined as a case where decreased faecal consistency was such a concern to warrant being mentioned in the EMR. Different cut-offs in number of days from a previous episode were evaluated at 90 and 180 days to define a case of diarrhoea as a "new case".

Text mining was performed on the dataset from the United Kingdom and all cases for which at least one episode of diarrhoea was recorded in the EMR were extracted from the dataset using the methodology described in Chapter 3. A comprehensive inclusion characterisation dictionary was created to detect all cases referring to faeces from soft to watery. A comprehensive exclusion dictionary was also used, to exclude for example, the frequent recommendation included in clinical records, to 'monitor a patient for diarrhoea' or 'discontinue a treatment in case of the development of diarrhoea'.

A control population was selected at random from those horses matching each diarrhoea episode by date and practice for which diarrhoea was never mentioned in the EMR. Control equids were selected from the same practice and if they had received an examination on the same date of the episode of diarrhoea; where no equid was examined on the same date of the diarrhoea episode, a case with the closest date available was chosen. For each episode of diarrhoea, even if from an equid that had multiple episodes, a new control subject was selected so that the number of control equids matched exactly the number of episodes of diarrhoea.

The dataset, including all cases with diarrhoea and their matched controls, was used for the following analysis.

5.2.2.3 Diarrhoea prevalence determination

The results of data mining were imported in R to calculate the difference in days between episodes, for equids in which more than one episode of diarrhoea was recorded. A dataset was created including all episodes of diarrhoea, date and practice. The prevalence estimate for diarrhoea and confidence intervals were calculated using the formula:

$$p = \frac{p_0 + t/2}{1+t} \pm \frac{\sqrt{p_0 q_0\, t + t^2/4}}{1+t}$$

In this equation p is the probable prevalence, p_0 is the observed prevalence, t is the square of the distribution divided by the sample size and q_0 is the coefficient used to calculate the standard deviation in $(p_0 q_0 / n)^{1/2}$ where n is the sample size (Wilson, 1927). This formula is appropriate for the determination of prevalence estimates as the confidence intervals are bound to be greater than or equal to zero.

5.2.2.4 Explanatory variable: non-steroidal anti-inflammatory drugs

All cases included in the dataset described in the previous section receiving NSAIDs were identified in R. The date of recorded usage of an NSAID was used to determine if drugs were administered in the two weeks preceding the development of diarrhoea.

The data was obtained for all NSAIDs together and was subsequently looked at more specifically for the five NSAIDs (phenylbutazone, suxibuzone, flunixin meglumine, metamizole and meloxicam), which were more commonly used in this country, as highlighted in Chapter 4. In addition to these, ketoprofen and firocoxib were also evaluated.

Ultimately 16 binary variables (“true” or “false” depending on whether use of the drug was recorded) were created to describe if usage of NSAIDs

as a whole, or of specific drugs, was recorded within 14 or seven days prior to the development of diarrhoea.

For the control animals these 16 variables were “true” or “false” depending on whether use of each drug was recorded within 14 or seven days prior to the date the matched horse in the diarrhoea group had developed diarrhoea.

5.2.2.5 Explanatory variable: Antimicrobials

Antimicrobial administration was defined as all antimicrobial agents for systemic administration. All cases included in the dataset described in the previous section receiving antimicrobials were identified in R. The date of recorded usage of an antimicrobial was used to determine when drugs were administered in the two weeks preceding the development of diarrhoea. Contemporary drug administration in the matched control animals was also identified. Ultimately two binary variables were created depending on whether use of antimicrobials was recorded within 14 or seven days prior to the development of diarrhoea.

5.2.2.6 Explanatory variable: Corticosteroids

Corticosteroids administration was also evaluated and included corticosteroids for systemic administration such as dexamethasone, prednisolone and prednisone. All cases included in the dataset described in the previous section receiving corticosteroids were identified in R. The date of recorded usage of a corticosteroid was used to determine when drugs were administered in the two weeks preceding the development of diarrhoea. Contemporary drug administration in the matched control animals was also identified. Ultimately four binary variables were created depending on whether use of any corticosteroid, or just systemic

corticosteroid, was recorded within 14 or seven days prior to the development of diarrhoea.

5.2.2.7 Explanatory variable: Laxatives

Laxatives were defined as drugs administered to reduce faecal consistency. Typically these included magnesium sulphate (Epsom salts) or liquid paraffin. All cases included in the dataset described in the previous section receiving laxatives were identified in R. The date of recorded usage of a laxative was used to determine if the treatment was administered in the two days preceding the development of diarrhoea. Contemporary laxative administration in the matched control animals was also identified. Ultimately one binary variable was created for when administration of a laxative treatment was recorded within the 48 hours prior to the development of diarrhoea.

5.2.2.8 Explanatory variable: Intravenous fluids

Intravenous fluid therapy was defined as the parenteral administration of fluids. It did not account for quantity administered, administration rate or treatment length. All cases included in the dataset described in the previous section undergoing intravenous fluid therapy were identified in R. The date of recorded parenteral administration of fluids was used to determine if the treatment occurred in the two days preceding the development of diarrhoea. Contemporary fluid administration in the matched control animals was also identified. Ultimately one binary variable was created when intravenous fluid administration was recorded within the 48 hours prior to the development of diarrhoea.

5.2.2.9 Explanatory variable: Anthelmintic drugs

All cases included in the dataset described in the previous section receiving anthelmintic drugs were identified in R. The date of recorded usage of an anthelmintic was used to determine if drugs were administered in the two weeks preceding the development of diarrhoea. Contemporary drug administration in the matched control animals was also identified. Ultimately two binary variables were created depending on whether use of anthelmintic drugs was recorded within 14 or seven days prior to the development of diarrhoea.

5.2.2.10 Statistical analysis

Multivariable conditional logistic regression modelling was selected for the analysis to evaluate the effect of multiple, potentially inter-related explanatory variables. The outcome variable was binomial in nature, e.g. presence or absence of diarrhoea.

Logistic regression models the natural logarithm (ln) of the odds of an outcome for a given value of explanatory variable(s). The odds are defined as the probability of having an outcome (diarrhoea) divided by the probability of not having the outcome (no diarrhoea).

The general formula depicting a logistic regression model is as follows:

$$y = ln\left[\frac{p}{1-p}\right] = \alpha + \beta_1 x_1 + \beta_2 x_2 + \cdots + \beta_i x_i$$

where the first term of the equation is the log transformation of the odds of the outcome, with *p* being the probability of that outcome actually happening, α is the intercept term, β_1 to β_i are the regression coefficients which represent the change of y for a unit change in the outcome x.

Logistic regression also allows determination of interactions between explanatory variables. Interactions should be evaluated to ascertain that the effect of two variables on the outcome is not simply additive, as the two variables might enhance or moderate each other's effect (Dohoo et al., 2010). For example, NSAIDs and antimicrobials are well recognised causes of diarrhoea in the horse (Sanchez, 2010). Physiologically this is explainable as flunixin meglumine weakens the intestinal lining by reducing mucosal blood flow. Antimicrobials increase the number of toxins released with bacterial death within the intestinal lumen, which also often results in diarrhoea. However, the effect of each drug combined might be expected to be greater as the effect of the increased quantity of bacterial toxins produced as a consequence of antimicrobial administration could have a greater effect on a mucosa weakened by flunixin meglumine. This example highlights the possible pathogenic mechanism through which the combination of both drugs might cause a greater effect than the additive effect of each individual drug. Potential interaction terms were chosen based on those that might make sense pharmacologically. Another reason for testing certain interaction terms was that they reflect common clinical practice as drugs are often administered in combination. This is the case for example with antimicrobials and NSAIDs, different NSAIDs together (flunixin meglumine and phenylbutazone), or antimicrobials and corticosteroids as these scenarios reflect common clinical practice. Conditional logistic regression is used for binary data with one or more predictors, where observations are not independent but are matched or grouped in some way.

Odds ratio (OR) calculation is also affected when including interaction terms in the regression model. The OR for each single first term is obtained by multiplying the OR, as if there was no significant interaction, with the OR of the interaction. The OR of the interaction is obtained by the multiplication of the OR of each single term, as if there was no

significant interaction, with the OR of the interaction alone (Dohoo et al., 2010). The OR for interaction terms was then calculated with the following formula:

$$e^{(\beta_1+\beta_2+\beta_{1*2})\pm 1.96\sqrt{{\sigma_1}^2+{\sigma_2}^2+{\sigma_{1*2}}^2}}$$

where β_1 and β_2 are the coefficient of each single variables and β_{1*2} is the coefficient of the interaction term and σ is the standard error.

For example, in the output of the logistic regression model the coefficient calculated for flunixin meglumine was 1.04, for phenylbutazone was -0.59 and for the interaction term flunixin*phenylbutazone was -0.69. From this the resulting OR was obtained as the exponential of -0.24 obtained from the sum of the coefficients of each term (e.g. (1.04+(-0.59)+(-0.69)=-0.24).

Univariable logistic regression was used initially to evaluate the significance, and the size of the effect, of each explanatory variable on its own and to evaluate whether each variable should be considered for inclusion in the multivariable analysis.

The Akaike Information Criterion (AIC) was used to select the regression model best fitting the data. The AIC provides an index to compare the quality of each model, by estimating the amount of information lost by each model (Akaike, 1998, 1974). The AIC value is calculated by the following formula:

$$AIC = 2K - 2\,ln(L)$$

where K is the number of parameters in the model and L is the likelihood function of the model. From this formula it is clear that the AIC can be used only to compare models that use the same number of observations,

e.g. K, so that the AIC varies only along with the log likelihood. In this study the AIC was used as every explanatory variable had a complete set of data points. The dataset used had no missing data. In fact all explanatory variables were binary and if a drug was used then it was classified as "true", otherwise as "false".

Conditional logistic regression analysis was performed in R using the function "clogit" of the package "survival" and the best fitting model was obtained by adding and removing different variables using a forward and backwards approach. The term defining the matching was added as "+strata(set)" where "set" was a variable assigning each diarrhoea positive case to its own control.

AIC was used to evaluate whether the addition of an interaction term improved significantly the fit of the model to the data. The suitability of each model was evaluated using relative likelihood of the model calculated by the formula:

$$e^{\frac{\mathrm{AIC_{min}}-\mathrm{AIC_i}}{2}}$$

where AIC_{min} is the smallest AIC, AIC_i is the one being tested. A commonly accepted cut-off AIC difference is four which means that the model with the higher AIC of the two is 0.135 times as likely to minimise information loss by addition of an explanatory variable to the model (Steyerberg, 2009)

5.2.2.11 Post-hoc analysis to account for comorbidities

Other comorbidities were also included *post-hoc* to further explain the findings of the initial analyses and account for the possible confounding effect of comorbidities that could also have triggered diarrhoea and that might have required treatment with some of the drugs included as

explanatory variables in the initial model. The EMR referring to colic and orthopaedic disease, referred to in Chapter 4, were included in the model. These were used to identify patients suspected, investigated or treated for abdominal discomfort or an orthopaedic condition in zero to seven days or seven to 14 days, or at any other point in the EMRs, preceding the development of diarrhoea. These variables were mutually exclusive. Conditional logistic regression was then used to evaluate whether inclusion of one or both comorbidities altered the statistical significance and odd ratios of variables included in the model.

Inclusion, exclusion and re-inclusion dictionaries used for the analysis are available in Appendix 5.

The study was approved by Ethics and Welfare Committee of the School of Veterinary Medicine at the University of Glasgow.

5.2.3 Results

5.2.3.1 Prevalence of reported non-steroidal anti-inflammatory drug toxicity

Of the total of 2,653,695 rows of data from 141,543 equids from the United Kingdom, NSAID toxicity was suspected or diagnosed in only eight animals. The demographic details of the population in this dataset are described in Chapter 3. The overall prevalence of NSAID toxicity in this population from the United Kingdom was therefore 0.01% (95% CIs: 0.00-0.01%).

5.2.3.2 Diarrhoea and Control population

A 90 day cut-off was arbitrarily chosen based on the author's expert opinion. Longer cut-offs (up to 180 days) were also evaluated and would have changed the number of diarrhoea episodes by a maximum of 0.8%.

Of the 141,543 equine patients in the dataset, a total 2,427 equids suffered at least one episode of diarrhoea with a total of 2,589 episodes of diarrhoea, of which 432 were recorded as horses whereas the rest were recorded as equine.

Of all equids, 2,327 had a single episode of diarrhoea recorded, 70 had two episodes, 14 had three episodes, seven had four episodes, five had five episodes, two had six episodes and the last two equids had seven and eight episodes each. The mean age of the population that suffered diarrhoea was 13.5 years (median 12 years, range: 0-40 years), while a plausible age was not available for 1,310 equids of those with diarrhoea. Diarrhoea was reported in 687 females and 613 males, of which 531 were geldings, 82 entire males. Gender had not been recorded in 1,127 equids with diarrhoea. The control population included a total of 2,589 animals, of which 434 were recorded as being horses, and the remaining simply as equines. No control animal appeared more than once in the dataset. The mean age of the control population was 12.6 years (median 11.3 years, range: 0-40 years), while age was not available for 1,315 control animals. Control animals for which gender had been recorded included 656 females and 652 males, of which 562 were geldings and 90 entire males.

The dataset including all cases with diarrhoea and their matched control animals is from now on referred to as the "diarrhoea dataset" and included a total of 5016 equids, 2427 that suffered at least one episode of diarrhoea and 2589 equids that never had diarrhoea reported in the record.

5.2.3.3 Diarrhoea prevalence determination

A total of 2427 equids from the population of 141,543 animals suffered at least one episode of diarrhoea severe enough to be mentioned in the EMR (diarrhoea prevalence: 1.71%; 95% CIs: 1.65-1.78%).

5.2.3.4 Explanatory variable: non-steroidal anti-inflammatory drugs

From the total of 141,543 animals, administration of NSAIDs was recorded at least once in the EMR of 39,816 patients, with a calculated prevalence for NSAIDs usage of 28.13% (95% CIs: 27.9-28.36%).

A total of 794 animals, out of the total of 5016 equids in the diarrhoea dataset, received NSAIDs in the preceding 14 days. Of these, 583 were from the diarrhoea group and 211 were from the control group. The overall prevalence of NSAID usage in the 14 days preceding the development of diarrhoea was 22.52% (CIs: 21.0-24.20%), while in the control group this was 8.15% (95% CIs: 7.16-9.27%).

The prevalence of phenylbutazone usage within 14 days of the 'case date' was 11.32% (293 animals; 95% CIs: 10.15-12.6%) in those with diarrhoea (n= 2427) and 4.87% (126 animals; 95% CIs: 4.10-5.76%) in control animals (n= 2589). The prevalence of suxibuzone usage with 14 days of the 'case date' was 4.83% (125 animals; 95% CIs: 4.07-5.72%) in those with diarrhoea and 1.74% (45 animals; 95% CIs: 1.30-2.32%) in control animals. The prevalence of flunixin meglumine usage within 14 days of the 'case date' was 5.95% (154 animals; 95% CIs: 5.10-6.93%) in those with diarrhoea and 2.16% (56 animals; 95% CIs: 1.67-2.80%) in control animals. The prevalence of ketoprofen usage within 14 days was 0.19% (5 animals; 95% CIs: 0.08-0.45%) in those with diarrhoea and no animal received ketoprofen in the control group (0.00%; 95%CIs: 0.00-0.15%). Prevalence of meloxicam usage within 14 days of the 'case date' was 1.04% (27 animals; 95% CIs: 0.72-

1.51%) in equids with diarrhoea and 0.35% (9 animals; 95% CIs: 0.18-0.66%) in the control group. Prevalence of metamizole usage within 14 days of the ‘case date’ was 2.05% (53 animals; 95% CIs: 1.57-2.67%) in equids with diarrhoea and 0.30% (8 animals; 95% CIs: 0.16-0.61%) in the control group. Finally firocoxib was administered to only one horse, which suffered diarrhoea within 14 days (prevalence of use 0.04%; 95% CIs: 0.01-0.22%).

When including only NSAIDs administered seven days from the ‘case date’, the prevalence of NSAIDs usage was 19.16% (496 animals; 95% CIs: 17.69-20.72%) in equids with diarrhoea and 6.1% (158 animals; 95% CIs: 5.24-7.09%) in the control group. The prevalence of phenylbutazone usage within 7 days from the ‘case date’ was 9.39% (243 animals; 95% CIs: 8.32-10.57%) in equids with diarrhoea and 3.44% (89 animals; 95% CIs: 2.80-4.21%) in the control group. The prevalence of suxibuzone usage within 7 days from the ‘case date’ was 3.75% (97 animals; 95% CIs: 3.08-4.55%) in equids with diarrhoea and 1.35% (35 animals; 95% CIs: 0.97-1.87%) in the control group. The prevalence of flunixin meglumine usage within 7 days from the ‘case date’ was 5.25% (136 animals; 95% CIs: 4.46-6.18%) in equids with diarrhoea and 1.66% (43 animals; 95% CIs: 1.24-2.23%) in the control group. The prevalence of ketoprofen usage within 7 days from the ‘case date’ was 0.19% (5 animals; 95% CIs: 0.08-0.45%) in equids with diarrhoea and no animal in the control group had received ketoprofen (prevalence 0.00%; 95% CIs: 0.00-0.15%). Prevalence of meloxicam use within 7 days from the ‘case date’ was 0.81% (21 animals; 95% CIs: 0.53-1.24%) in equids with diarrhoea and 0.35% (9 animals; 95% CIs: 0.18-0.66%) in the control group. The prevalence of metamizole use within 7 days from the ‘case date’ was 1.89% (49 animals; 95% CIs: 1.43-2.49%) in equids with diarrhoea and 0.19% (5 animals; 95% CIs: 0.08-0.45%) in the control group. Finally firocoxib was administered only to one horse, which suffered diarrhoea within 7 days (prevalence of use 0.04%; 95% CIs: 0.01-0.22%) and no equid in the control group received firocoxib (prevalence 0.00%; 95% CIs: 0.00-0.15%).

These results are summarised in Table 5.1.

Table 5.1: Prevalence of NSAIDs usage 14 and 7 days before a 'case date' from 2427 equids that suffered 2589 episodes of diarrhoea and 2589 control animals that never had diarrhoea in the United Kingdom.

Drug	Days	Total	D	Prevalence (95% CIs)	C	Prevalence (95% CIs)
NSAIDs	14	794	583	22.52 (20.95-24.17)	211	8.15 (7.16-9.27)
	7	654	496	19.16 (17.69-20.72)	158	6.1 (5.24-7.09)
Phenylbutazone	14	419	293	11.32 (10.15-12.60)	126	4.87 (4.10-5.76)
	7	332	243	9.39 (8.32-10.57)	89	3.44 (2.80-4.21)
Suxibuzone	14	170	125	4.83 (4.07-5.72)	45	1.74 (1.30-2.32)
	7	132	97	3.75 (3.08-4.55)	35	1.35 (0.97-1.87)
Flunixin meglumine	14	210	154	5.95 (5.10-6.93)	56	2.16 (1.67-2.80)
	7	179	136	5.25 (4.46-6.18)	43	1.66 (1.24-2.23)
Ketoprofen	14	5	5	0.19 (0.08-0.45)	0	0.00 (0.00-0.15)
	7	5	5	0.19 (0.08-0.45)	0	0.00 (0.00-0.15)
Meloxicam	14	36	27	1.04 (0.72-1.51)	9	0.35 (0.18-0.66)
	7	30	21	0.81 (0.53—1.24)	9	0.35 (0.18-0.66)
Metamizole	14	61	53	2.05 (1.57-2.67)	8	0.30 (0.16-0.61)
	7	54	49	1.89 (1.43-2.49)	5	0.19 (0.08-0.45)
Firocoxib	14	1	1	0.04 (0.01-0.22)	0	0.00 (0.00-0.15)
	7	1	1	0.04 (0.01-0.22)	0	0.00 (0.00-0.15)

Days: maximum number of days between drug administration and development of diarrhoea; Total: total number of equids receiving that drug; D: diarrhoea group; C control group.

5.2.3.5 Explanatory variable: Antimicrobials

From the total of 141,543 animals, administration of antimicrobials was recorded at least once in the EMR of 34,102 patients with a prevalence for antimicrobial usage of 24.09% (95% CIs: 23.87-24.32%).

The prevalence of antimicrobial usage within 14 days from the 'case date' was 16.26% (421 animals; 95% CIs: 14.89-17.73%) in those with diarrhoea and 7.34% (190 animals; 95% CIs: 6.40-8.41%) in control animals.

In the 7 days before the 'case date', the prevalence of antimicrobial usage was 13.56% (351 animals; 95% CIs: 12.29-14.93%) in the equids with

diarrhoea and 5.68% (147 animals; 95% CIs: 4.85-6.64%) in the control group.

5.2.3.6 Explanatory variable: Corticosteroids

Of the total of 141,543 animals, administration of corticosteroids was recorded at least once in the EMR of 10,518 patients with a prevalence for corticosteroids usage of 7.43% (95% CIs: 7.30-7.57%).

The prevalence of corticosteroid administration in the 14 days preceding the 'case date' was 4.36% (113 animals; 95% CIs: 3.64-5.22%) in the equids with diarrhoea and 1.74% (45 animals; 95% CIs: 1.3-2.32%) in the control group.

The prevalence of corticosteroid administration in the 7 days preceding the 'case date' was 3.51% (122 animals; 95% CIs: 2.87-4.30%) in the equids with diarrhoea and 1.20% (31 animals; 95% CIs: 0.84-1.69%) in the control group.

When looking specifically at usage of systemic corticosteroid (dexamethasone, prednisolone, prednisone), the prevalence over the total population of 141,543 equids was 4.26% (6007 animals; 95% CIs: 4.15-4.37%), which accounted for 57.11% (95%CIs: 56.16-58.05%) of all corticosteroids.

The prevalence of systemic corticosteroid administration in the 14 days preceding the 'case date' was 2.16% (56 animals; 95% CIs: 1.67-2.80%) in the equids with diarrhoea and 0.70% (18 animals; 95% CIs: 0.44-1.10%) in the control group.

The prevalence of systemic corticosteroid administration in the 7 days preceding the 'case date' was 1.82% (47 animals; 95% CIs: 1.37-2.41%) in

the equids with diarrhoea and 0.58% (15 animals; 95% CIs: 0.35-0.95%) in the control group.

5.2.3.7 Explanatory variable: Laxatives

Of the total of 141,543 animals, administration of laxatives was recorded at least once in the EMR of 5850 patients with a prevalence for laxative usage of 4.15% (95% CIs: 4.04-4.25%).

The prevalence of laxative usage in the 2 days preceding a 'case date' was 1.04% (31 animals; 95% CIs: 0.72-1.51%) in the equids with diarrhoea and 0.15% (4 animals; 95% CIs: 0.06-0.40%) in the control group.

5.2.3.8 Explanatory variable: Intravenous fluids

Of the total of 141,543 animals, administration of intravenous fluids was recorded at least once in the EMR of 2122 patients with a prevalence of fluid administration intravenously of 1.50% (95% CIs: 1.44-1.57%).

The prevalence of fluid therapy usage in the 2 days preceding a 'case date' was 3.51% (91 animals; 95% CIs: 2.87-4.30%) in equids with diarrhoea and 0.58% (15 animals; 95% CIs: 0.35-0.95%) in the control group.

5.2.3.9 Explanatory variable: Anthelmintic drugs

Of the total of 141.061 animals, administration of anthelmintic drugs was recorded at least once in the EMR of 9835 patients with a prevalence for anthelmintic drug usage of 6.97% (95% CIs: 6.84-7.11%).

The prevalence of anthelmintic drug usage in the 14 days preceding the 'case date' was 3.94% (102 animals; 95% CIs: 3.26-4.76%) in equids with

diarrhoea and 1.31% (34 animals; 95% CIs: 0.94-1.83%) in the control group.

The prevalence of anthelmintic drug usage in the 7 days preceding the 'case date' was 3.05% (105 animals; 95% CIs: 2.46-3.79%) in equids with diarrhoea and 1.00% (26 animals; 95% CIs: 0.69-1.47%) in the control group.

Prevalence data on drug usage is summarised in Table 5.2. Prevalence data of equids in the diarrhoea dataset is summarised in Table 5.3.

Table 5.2: Prevalence of drug usage in 141,543 equids from the United Kingdom. The data reflects usage of each drug at some point in the animal EMR.

Drug	Equids	Prevalence (%)	95% CIs (%)
Antimicrobials	34,102	24.09	23.87-24.32
Corticosteroids	10,518	7.43	7.30-7.57
Systemic corticosteroids	6007	4.26	4.15-4.37
Laxatives	5850	4.15	4.04-4.25
Intravenous fluids	2122	1.50	1.44-1.57
Anthelmintic drugs	9835	6.97	6.84-7.11

Table 5.3: Prevalence of drug usage 14 and 7 days before a 'case date' from 2427 equids that suffered 2589 episodes of diarrhoea and 2589 control animals that never had diarrhoea in the United Kingdom.

Drug	Days	Total	D	Prevalence (95% CIs)	C	Prevalence (95% CIs)
Antimicrobials	14	611	421	16.26 (14.89-17.73)	190	7.34 (6.40-8.41)
	7	498	351	13.56 (12.29-14.93)	147	5.68 (4.85-6.64)
Corticosteroids	14	158	113	4.36 (3.64-5.22)	45	1.74 (1.30-2.32)
	7	153	122	3.51(2.87-4.30)	31	1.20 (0.84-1.69)
Systemic	14	74	56	2.16 (1.67-2.80)	18	0.70 (0.44-1.10)
corticosteroids	7	62	47	1.82 (1.37-2.41)	15	0.58 (0.35-0.95)
Laxatives	2	35	31	1.04 (0.72-1.51)	4	0.15 90.06-0.40)
Intravenous fluids	2	106	91	3.51 (2.87-4.30)	15	0.58 (0.35-0.95)
Anthelmintic	14	136	102	3.94 (3.26-4.76)	34	1.31 (0.94-0.83)
drugs	7	184	105	3.05 (2.46-3.79)	26	1.00 (0.69-1.47)

Days: maximum number of days between drug administration and development of diarrhoea; Total: total number of equids receiving that drug; D: diarrhoea group; C control group.

5.2.3.10 Statistical analysis

Univariable logistic regression

When looking at drug administration in the 14 days preceding the development of diarrhoea, NSAIDs usage was 3.72 (95% CIs: 3.06-4.51; p<0.001) times more likely in equids that had diarrhoea. Equids with diarrhoea were 2.74 times more likely to have received phenylbutazone (95%CIs: 2.16-3.48; p<0.001) and 3.16 (95%CIs: 2.18-4.59; p<0.001) times more likely to have received suxibuzone. Equids with diarrhoea were 3.13 (95% CIs: 2.24-4.39; p<0.001) times more likely to have received flunixin meglumine. Equids with diarrhoea were 7.76 (95% CIs: 3.56-16.89; p<0.001) times more likely to have received metamizole, while equids with diarrhoea were 3.23 (95% CIs: 1.44-7.26; p=0.004) times more likely to have received meloxicam. The presence of diarrhoea was not significantly associated with the administration of ketoprofen (p=0.931) or firocoxib (p=0.953).

Equids with diarrhoea were 2.67 (95% CIs: 2.18-3.26; p<0.001) times more likely to have received antimicrobials and 2.84 (95% CIs: 1.95-4.16; p<0.001) times more likely to have received corticosteroids and 3.5 (95 CIs: 1.97-6.19; p<0.001) times more likely to have received systemic corticosteroids and 3.39 (95% CIs: 2.22-5.16; p<0.001) times more likely to have received anthelmintic drugs in the 2 weeks before the development of diarrhoea.

Equids with diarrhoea were 4.18 (95% CIs: 3.37-5.17; p<0.001) times more likely to have received some NSAIDs in the seven days prior to the development of diarrhoea administration. Equids with diarrhoea were 3.25 (95% CIs: 2.47-4.27; p<0.001) times more likely to have received phenylbutazone, 3.14 (95% CIs: 2.06-4.79; p<0.001) times more likely to have received suxibuzone, 3.64 (95% CIs: 2.50-5.30; p<0.001) times more

likely to have received flunixin meglumine, 11.59 (95% CIs: 4.48-29.98; p<0.001) times more likely to have received metamizole and 2.52 (95% CIs: 1.08-5.86; p=0.032) times more likely to have received meloxicam. There was no significant relationship between the administration of either ketoprofen (p=0.31) or firocoxib (p=0.953) in the week before development of diarrhoea and diarrhoea.

Equids with diarrhoea were 2.85 (95% CIs: 2.29-3.56; p<0.001) times more likely to have received antimicrobials, 3.36 (95 CIs: 2.16-5.23; p<0.001) times more likely to have received corticosteroids, 3.54 (95 CIs: 1.90-6.62; p<0.001) times more likely to have received systemic corticosteroids and 3.45 (95% CIs: 2.14-5.58; p<0.001) times more likely to have received anthelmintic drugs in the 2 weeks before the development of diarrhoea. Equids with diarrhoea were also 7.81 (95% CIs: 2.61-23.31 p<0.001) times more likely to have received a laxative and 7.26 (95% CIs: 4.08-12.93; p<0.001) times more likely to have received intravenous fluid therapy in the 2 days preceding the development of diarrhoea. These results are summarised in Table 5.4.

Multivariable conditional logistic regression

The best fitting model without inclusion of interaction terms included the following parameters (Table 5.5):

i. Drugs dispensed two days before diarrhoea: intravenous fluids
ii. Drugs dispensed seven days before diarrhoea: phenylbutazone, metamizole, corticosteroids
iii. Drugs dispensed 14 days before diarrhoea: suxibuzone, antimicrobials, anthelmintic drugs.

An interaction term between phenylbutazone and antimicrobials also improved model fit significantly and was therefore included.

Table 5.4: Univariable logistic regression of variables considered for the multivariable conditional logistic regression. The number of diarrhoea episodes and matched controls is 2589 each in the dataset from the United Kingdom.

Treatment	Days	Total	D	C	P-value	OR	95% CIs
NSAIDs	7	654	496	158	<0.001	3.80	3.10-4.63
	14	794	583	211	<0.001	4.18	3.37-5.17
Phenylbutazone	7	332	243	89	<0.001	2.90	2.25-3.73
	14	419	293	126	<0.001	3.25	2.47-4.27
Suxibuzone	7	133	97	35	<0.001	2.88	1.94-4.28
	14	170	125	45	<0.001	3.14	2.06-4.79
Flunixin meglumine	7	179	136	43	<0.001	3.45	2.40-4.95
	14	210	154	56	<0.001	3.64	2.50-5.30
Metamizole	7	54	49	5	<0.001	15.67	4.88-50.33
	14	61	53	8	0.032	11.59	4.48-29.98
Meloxicam	7	30	21	9	0.033	2.33	1.07-5.09
	14	36	27	9	<0.001	2.52	1.08-5.86
Ketoprofen	7	5	5	0	0.991	$1.08x10^7$	0-Infinite
	14	5	5	0	0.310	$2.87x10^5$	0-Infinite
Firocoxib	7	1	1	0	0.991	$1.47x10^6$	0-Infinite
	14	1	1	0	0.953	$1.05x10^5$	0-Infinite
Antimicrobials	7	498	351	147	<0.001	2.77	2.24-3.43
	14	611	421	190	<0.001	2.85	2.29-3.56
Corticosteroids	7	122	91	31	<0.001	3.07	2.01-4.67
	14	158	113	45	<0.001	3.36	2.16-5.23
Systemic corticosteroids	7	62	47	15	<0.001	3.29	1.81-5.98
	14	74	56	18	<0.001	3.54	1.90-6.62
Laxatives	2	35	31	4	<0.001	7.81	2.61-23.31
Intravenous fluids	2	106	91	15	<0.001	7.26	4.08-12.93
Anthelmintic drugs	7	105	79	26	<0.001	3.00	2.03-4.42
	14	136	102	34	<0.001	3.45	2.14-5.58

Days: maximum number of days between drug administration and development of diarrhoea; Total: total number of equids receiving that drug; D: diarrhoea group; C control group; OR: odds ratio.

Equids with diarrhoea were 2.75 (95% CIs: 1.86-4.07; p<0.001) times more likely to have received phenylbutazone in the preceding 7 days, 1.83 (95% CIs: 1.23-2.73; p=0.003) times more likely to have received suxibuzone in the preceding 14 days, 6.84 (95% CIs: 2.57-18.21; p<0.001) times more likely to have received metamizole in the preceding 7 days, 1.89 (95% CIs: 1.47-2.39; p<0.001) times more likely to have received antimicrobials in the preceding 14 days, 1.89 (95% CIs: 1.18-3.02; p=0.008) times more

likely to have received corticosteroids in the preceding 7 days, 4.44 (95% CIs: 2.43-8.09; p<0.001) times more likely to have received intravenous fluids in the preceding 2 days and 2.20 (95% CIs: 1.41-3.41; p=0.001) times more likely to have received anthelmintic drugs in the preceding 14 days. Further the only significant interaction term tested was that between phenylbutazone and antimicrobials, as equids with diarrhoea were 2.44 (95%CIs: 1.06-5.63; p=0.006) times more likely to have received both phenylbutazone and antimicrobials in the 7 and 14 days preceding diarrhoea respectively.

Table 5.5: Multivariable conditional logistic regression model showing variables significantly associated with the risk of developing diarrhoea in the United Kingdom. The number of diarrhoea episodes and matched controls is 2589 each.

Treatment	Days	Total	D	C	P-value	OR	95% CIs
Phenylbutazone	7	332	243	89	<0.001	2.75	1.86-4.07
Suxibuzone	14	170	125	45	0.003	1.83	1.23-2.73
Metamizole	7	54	49	5	<0.001	6.84	2.57-18.21
Antimicrobials	14	611	421	190	<0.001	1.87	1.47-2.39
Corticosteroids	7	122	91	31	0.008	1.89	1.18-3.02
Intravenous fluids	2	106	91	15	<0.001	4.44	2.43-8.09
Anthelmintic drugs	14	136	102	34	0.001	2.20	1.41-3.41
Phenylbutazone*Antimicrobials	7*14	169	124	45	0.006	2.44	1.06-5.63

Days: maximum number of days between drug administration and development of diarrhoea; Total: total number of equids receiving that drug; D: diarrhoea group; C control group; OR: odds ratio

Other variables, such as flunixin meglumine (p=0.19), ketoprofen (p=0.93), meloxicam (p=0.58), firocoxib (p=0.96) and laxatives (p=0.054) were not significant at any time before diarrhoea and were not included in the final model. An interaction term between suxibuzone and antimicrobials was tested but found not to be significant (p=0.07). The interaction between metamizole and antimicrobials was significant (p=0.041) but improved the AIC by less than two units and therefore was not retained in the final model. An interaction term between phenylbutazone and suxibuzone was also tested and shown not to be significant (p=0.45).

5.2.3.11 Post-hoc analysis to account for co-morbidities

The dataset of 141,543 animals included a total of 15,747 equids suffering at least one episode of abdominal discomfort at some point in the EMRs dataset with a total prevalence of 11.1% (95% CIs: 11.0-11.3%). In the diarrhoea dataset the number of equids suffering at least one episode of colic was 1,872 of which 1,151 (1236 episodes) were in the diarrhoea group and 721 (721 episodes) in the control group.

The dataset of 141,543 animals included a total of 55,728 equids in the orthopaedic group with a total prevalence of 39.37% (95% CIs: 39.12-39.63%). In the diarrhoea dataset the number of equids suffering at least one episode of orthopaedic disease was 3,470 of which 1,638 (1763 episodes) were in the diarrhoea group and 1,832 (1832 episodes) in the control group.

The best fitting multivariable conditional logistic regression model with co-morbidities included the following parameters (Table 5.6):

i. Drugs dispensed two days before diarrhoea: intravenous fluids
ii. Drugs dispensed seven days before diarrhoea: phenylbutazone
iii. Drugs dispensed 14 days before diarrhoea: suxibuzone, antimicrobials, anthelmintic medications
iv. Colic seven days before diarrhoea
v. Orthopaedic disease seven days before diarrhoea
vi. Orthopaedic disease at any point before 14 days before diarrhoea

With the addition of colic and orthopaedic disease to the logistic regression analysis the effects of corticosteroids (p=0.06) and metamizole (0.1) became not significant in the *post-hoc* analysis. Further, in equids with diarrhoea the odds of having received phenylbutazone and intravenous fluids had decreased remarkably (>30%). Addition of colic at seven to 14 days before diarrhoea or any other time during the lifetime before that or orthopaedic disease at 14 days before diarrhoea did not

improve the AIC of the model significantly. Finally for equids with diarrhoea the odds of having received both phenylbutazone and antimicrobials had also dropped remarkably. The interaction term between antimicrobials and phenylbutazone did not improve the AIC significantly once colic and orthopaedic disease variables were added to the model and was therefore excluded from the final model.

Table 5.6: *Post-hoc* multivariable conditional logistic regression model showing variables significantly associated with the risk of developing diarrhoea in the United Kingdom. The number of diarrhoea episodes and matched controls is 2589 each.

Treatment	Days	Total	D	C	P-value	OR	95% CIs
Phenylbutazone	7	332	243	89	0.001	1.64	1.21-2.24
Suxibuzone	14	170	125	45	0.018	1.67	1.09-2.54
Antimicrobials	14	611	421	190	<0.001	1.68	1.33-2.13
Intravenous fluids	2	106	91	15	0.030	2.10	1.09-4.07
Anthelmintic drugs	14	136	102	34	0.003	1.97	1.26-3.07
Colic	7	855	714	141	<0.001	6.53	5.21-8.18
Orthopaedic disease	7	1426	834	592	0.003	1.26	1.08-1.46
Orthopaedic disease	life	1981	871	1110	<0.001	0.62	0.54-0.71

Days: maximum number of days between drug administration and development of diarrhoea; Total: total number of equids receiving that drug; D: diarrhoea group; C control group; OR: odds ratio; life: orthopaedic disease at any point in life other than 14 days before an episode of diarrhoea.

5.2.4 Discussion

This study presents several other interesting findings such as prevalence data, which were not within the main objective of the study. Prevalence of diarrhoea has not been reported before on such a large population in first opinion equine veterinary practice in the United Kingdom. Prevalence data is extracted from the EMRs stored in the PMSS and therefore is strictly influenced by veterinary surgeons commitment to record clinical data complete with details. The prevalence of diarrhoea reported in this study needs to be interpreted in light of the first opinion nature of the data. In the authors knowledge little data is available to compare disease prevalence between first opinion and referral equine practice. First opinion data is likely to include proportionally more cases of mild diarrhoea that require little clinical intervention, whilst referred cases are more likely to be a selection of more severe cases requiring more intensive management and treatment. Also the prevalence reported in this

study refers only to the prevalence of reported diarrhoea, so the actual prevalence of diarrhoea is likely to be higher. Reasons for not reporting diarrhoea might generally include mild, self-limiting episodes or cases with extreme financial limitations for which the owner would perceive that seeking veterinary advice would be too costly. Finally cases that suffered per-acute death could also have been missed, in which case a veterinary surgeon would have not been consulted. However, from a veterinary point of view the figure is interesting as it reflects the real prevalence of diarrhoea in equids under veterinary care. Another factor that might have contributed to underestimating diarrhoea prevalence is the definition of "diarrhoea episode" adopted for the study. It is possible that some equids could have had two episodes of diarrhoea from two different causes within 90 days. It is also important to point out that, while some animals with diarrhoea had multiple episodes recorded in the EMR the cut-off of 90 diarrhoea-free days between episodes was arbitrarily chosen to ensure that if a new episode was recorded this was unlikely to be a continuation of the previous episode. A shorter cut-off would have led to an overestimation of diarrhoea prevalence due to cases being examined more than once for the same disease process. While a patient might have chronic intermitted diarrhoea for several months, in such cases examinations would be expected to be more frequent than every three months.

Other conditions, which might be consistent with NSAID toxicity, include renal failure and gastric ulceration. However, since these are not clinical signs *per se*, but rely on further testing to be identified (haematology and urinalysis and gastroscopy), the risk for under-diagnosis is high, particularly for mild cases and their detection would certainly be an underestimation of the true prevalence. Therefore these conditions were deemed not suitable for the purpose of this study.

The present study did not evaluate whether practice played an effect on the prevalence of diarrhoea and drug usage. Diarrhoea and control cases were matched by date and practice in order to minimise the effect of a local outbreak of certain conditions leading to diarrhoea that could have affected the results. Therefore no difference in practice of origin would have been present between diarrhoea and control patients. This could have accounted for the different ability of veterinary surgeons to identify and report certain conditions such as diarrhoea, colic and orthopaedic disease and also drug usage patterns. However, identification for veterinary surgeons was available only in two of the datasets from the United Kingdom.

For a discussion on the prevalence of NSAID usage, colic and orthopaedic disease please refer to Chapter 4.

This study also reports the prevalence of the use of several variables in the general population, as well as in equids with diarrhoea. The use of antimicrobials in about a quarter of the general equine population highlights that they are frequently used in equine practice and this figure is potentially alarming in light of the recent threat to public health posed by antimicrobial resistance (Weese et al., 2015). The present study did not evaluate the use of each antimicrobial category or the use of protected antimicrobials as this was beyond the scope of the study. Differentiation in each antimicrobial category would be complicated and time consuming. Although this could provide a useful insight into the relationship between different antimicrobial classes and diarrhoea, the current study was focused on NSAIDs and further differentiation of antimicrobials by class was not performed.

The variable corticosteroids included all corticosteroids, which in equids mainly include those for intra-articular, inhaled or systemic administration. As intra-articular corticosteroids include a large proportion

of corticosteroids used in equine first opinion practice and their relationship with diarrhoea is currently unknown, a further variable including only systemic corticosteroids was created. From this data it appears that the systemic corticosteroids account for 57.1% of all corticosteroids used in our population.

Data on the prevalence of use of laxatives is in line with the overall prevalence of colic as these drugs are likely to be used in a proportion of colic cases suffering from impactions. The prevalence of intravenous fluid administration includes all cases receiving fluids for the management of hypovolaemia or dehydration. However the prevalence of hypovolaemia or dehydration was not investigated in the study so its prevalence cannot be compared to that of intravenous fluid usage.

Anthelmintic drugs are often administered to equids for treatment, as well as prevention of intestinal parasitic infections, which could result in diarrhoea. This study reported a relatively low prevalence of anthelmintic usage in the overall equine population (6.97%). It is important to highlight that in the United Kingdom anthelmintic drugs are POM-VPS drugs. This means that anthelmintic drugs can be purchased not only from veterinary surgeons but also from SPQs ("suitably qualified persons") as defined in the Veterinary Medicine Regulations. This means that owners can purchase anthelmintic drugs from people other than a veterinary surgeon, which could result in a serious underestimation of their use. Undoubtedly, explanatory variables consisting of drugs that are not under the exclusive control of veterinary professional, pose a serious challenge to epidemiologic research, as there would be no reliable way to assess their actual usage.

The results of this study support existing evidence that a significant relationship exists between administration of NSAIDs and development of diarrhoea in equine species. Conditional logistic regression analysis

confirms there was a statistically significant association between phenylbutazone, suxibuzone and metamizole administration and the development of diarrhoea. In the multivariable conditional logistic regression analysis, previous administration of flunixin meglumine, ketoprofen, meloxicam or firocoxib was not significantly associated with diarrhoea.

The lack of relationship between diarrhoea and administration of flunixin meglumine in the multivariable logistic regression analysis was somewhat surprising. Previous studies, including the findings of Chapter 2 of this manuscript have suggested that flunixin meglumine induced profound inhibition of COX activity in horses and can affect gastrointestinal mucosal blood-flow and healing (Cook et al., 2009a; Duz et al., 2015). In theory, this might result in diarrhoea, particularly in cases where underlying gastrointestinal disease could contribute to diarrhoea development. The only NSAIDs licenced in the United Kingdom for treatment of abdominal discomfort include flunixin meglumine, ketoprofen, meloxicam and metamizole (in combination with the spasmolytic butylscopolamine). The findings of Chapter 4 showed that flunixin meglumine and metamizole, along with phenylbutazone, were the most commonly used for treatment of abdominal discomfort. As cases with colic are likely to suffer abdominal disease that may lead to diarrhoea, it was expected that at least some of these cases receiving flunixin meglumine might have then developed diarrhoea. The results of univariable logistic regression show that equids with diarrhoea are 3.64 times more likely to have received flunixin meglumine compared to control animals. Nevertheless multivariable conditional logistic regression analysis failed to identify a significant relationship between administration of flunixin meglumine and diarrhoea. The reason for this finding remains undetermined, but it is possible that the concomitant use of other drugs or treatments included in the multivariable analysis could have affected this finding. Flunixin meglumine is often used, along with other medications, to manage conditions such as

colic and orthopaedic disease. Some of these, including intravenous fluids, antimicrobials and corticosteroids, might have a greater effect on the development of diarrhoea thus explaining the lack of significance of flunixin meglumine in both multivariable logistic regression models.

In the *post-hoc* analysis inclusion of colic and orthopaedic disease did not change the significance of the association of flunixin meglumine with the development of diarrhoea. These results suggest that, while in the univariable analysis equids with diarrhoea appear more likely to have received flunixin meglumine, other confounding factors, such as administration of other drugs or concurrent diseases processes including colic, are more likely determinants in cases of diarrhoea. The contribution of other factors not accounted for in this study, such as dosage of flunixin meglumine, route of administration and length of treatment also remain undetermined. However, from the results of this analysis it appears that this drug might be relatively safe, assuming it is used at label doses. More data is necessary including that of dose used and course length to actually confirm that veterinary surgeons and owners keep within the recommended dose regimens.

The odds for drug administration preceding an episode of diarrhoea were by far the highest with metamizole but the effect of this drug became non-significant once the co-morbidity of colic was included in the analysis. Similarly the inclusion of colic to the multivariable analysis affected negatively the odds ratio of phenylbutazone. However, this was to a lesser extent than metamizole, as phenylbutazone was used for a wide array of conditions and not only for colic. These results highlight the confounding effect that the variable colic had on other variables such as phenylbutazone and metamizole. This effect is the consequence of the fact that these variables are not entirely independent as these drugs are often used for the management of this condition as shown in Chapter 4.

The fact that in the multivariable conditional logistic regression analysis there was no significant relationship between diarrhoea and previous administration of ketoprofen, meloxicam and firocoxib is not surprising, as only a few animals in the dataset had received these drugs, so the statistical power to identify such an association, if it exists, would be limited in this study. Similarly, no significant relationship was present in the univariable conditional logistic regression analysis for ketoprofen and firocoxib. While the effect of meloxicam appeared significant in the univariable conditional logistic regression analysis, the effect of this small group was likely diluted in the multivariable logistic regression analysis and became then non-significant. Similar to flunixin meglumine, meloxicam is often used along with other medications, which might act as a confounding effect and have a greater effect on the development of diarrhoea.

In first opinion equine practice, antimicrobials are often administered in combination with NSAIDs, to treat bacterial infections and the associated inflammatory response. Since both classes of drugs have been well documented to induce diarrhoea in equids it seemed appropriate to include antimicrobials in the model and to investigate a possible interaction with NSAIDs. However, a significant interaction term was only identified between phenylbutazone and antimicrobials, suggesting that equids with diarrhoea were 2.4 times more likely to have been administered this drug combination. The odds ratio of the interaction term indicates that combination of phenylbutazone and antimicrobials worsens slightly the odds for development of diarrhoea than if antimicrobials were administered alone, while the odds for both drugs being administered together being associated with diarrhoea is lower than if phenylbutazone was used alone. However, this interaction term had to be dropped once colic and orthopaedic disease were accounted for in the post-hoc model.

Orthopaedic disease was also added to the model because affected patients often received NSAIDs as part of the management. Moreover, orthopaedic disease is unlikely to have any effect on the gastrointestinal tract and therefore orthopaedic patients might represent a selection of the overall population that is at a normal risk of developing diarrhoea. Interestingly orthopaedic procedures were 1.3 times more likely to have been carried out in the week preceding diarrhoea in this first opinion population. The reason for this remains undetermined, but could be related to the stress of being generally unwell or undergoing a veterinary lameness investigation. Horses with diarrhoea were 0.6 times as likely to have undergone an orthopaedic investigation at some point in life before the two weeks prior to diarrhoea.

The confounding effect on diarrhoea of underlying gastrointestinal disease, was somewhat accounted for by the colic variable. A colic episode was 6.5 times more likely in the week preceding the development of diarrhoea in a patient that then went on to develop diarrhoea than in one that did not. This demonstrated that diarrhoea is more likely in equids that have suffered gastrointestinal disease from infectious, inflammatory, ischaemic or dietary cause, manifested as abdominal discomfort, as all are known potential causes of colic in equids.

It was also somewhat surprising that no significant interaction was present between colic and NSAIDs administration since NSAIDs might reduce blood flow to the intestine, which might result in mucosal damage and diarrhoea, particularly if other concurrent gastrointestinal disease is present.

A significant relationship existed between corticosteroid administration and diarrhoea both in the single as well as multivariable conditional logistic regression analyses. Corticosteroids are often used intra-articular in equids to modulate inflammation associated with arthritis. But these

became non-significant with the addition of other co-morbidities, such as orthopaedic disease. Further, corticosteroid administration is not a common cause of diarrhoea at label doses in equids. In fact dexamethasone treatment is often warranted in cases of diarrhoea associated with cyathostomiasis as well as infiltrative bowel disease (Barr, 2006). The effect of corticosteroids became not significant once comorbidities such as orthopaedic disease were added to the analysis so the variable was dropped by the final model.

The time elapsed between the date of data entry in the EMR, corresponding to the drugs being dispensed, and the actual length of treatment was summarily accounted for by the use of the seven or 14 days period. The explanatory variables evaluated in this study include drugs administered or dispensed zero to seven and seven to 14 days prior to the development of diarrhoea. The use of either the seven or 14 days cut-off was determined on the basis of clinical relevance, AIC and significance in the model. Other variables such as laxatives and intravenous fluids were included only when administered up to two days before the development of diarrhoea. The two-day period for intravenous fluid and laxative administration was chosen because both treatments would be expected to affect faecal consistency within 48 hours of administration. Further, both treatments are usually administered directly by the veterinary surgeon on a single administration and therefore the date of data entry in records likely reflects the actual date of administration. This differs from the other explanatory variables, which include drugs often dispensed for administration by the owner over multiple days, hence evaluating generally a period of one to two weeks from the data being entered in the PMSS. For this reason no continuous variable was created to document the number of days between drug administration and development of diarrhoea. In fact, having included the number of days this would likely reflect the days between the drug being sold and the diarrhoea being noticed, with no information on treatment regimen.

It is important to point out that the findings of this study are applicable to a first opinion population and the findings do not apply to a referral population. First opinion cases vary more in the severity of the condition than referral cases, as cases are usually referred if they require more intensive management and exhaustive investigations. Therefore in a first opinion caseload, many cases might be very mild. Owner perception of disease severity might vary significantly and also plays a major role in the type of cases included.

Extraction of data regarding drug dosages and length of course of administration recommended is problematic and time consuming if at all achievable from free-text EMRs and was not performed in this study. This is a significant shortcoming of the study, as reliable information on doses as well as length of course of treatment prior to the development of diarrhoea could improve model fit as well as clinical relevance of the results substantially.

The *post-hoc* analysis was performed to further investigate some of the findings of the initial analysis. For example, while phenylbutazone is widely used to treat visceral pain in the horse in the United Kingdom, this use is off-label. Further, phenylbutazone and suxibuzone are also widely used to manage pain associated with the musculoskeletal system. Therefore, addition of colic and orthopaedic disease to the multivariable logistic regression model could have clarified further the relationship between diarrhoea and usage of these drugs.

In conclusion, it appears from the analysis of the data that while a significant relationship exists between diarrhoea and NSAID administration, the overall risk appears in fact to be very low. The data was obtained from a real population where veterinary clinicians are generally aware of the risk of side effects related to the use of this drug

and therefore are likely to judiciously observe label doses. Further work is required to assess whether label doses are observed in equine practice.

5.3 NSAID toxicity in North America

5.3.1 Introduction

The prevalence of toxicity related to NSAID administration is unknown in the North America. The clinical studies that describe NSAID toxicity that are from North America include only a total of 8 cases from 2 academic institutions collected over a period of years (Cohen et al., 1995; Jones et al., 2003). Even though the frequency of case reports in the peer reviewed literature is hardly a representative estimate of the true prevalence of disease, the rarity of these reports suggests that recognised clinically significant toxicity might be rare.

Toxicity from NSAIDs usage might manifest clinically as diarrhoea, gastric ulceration and renal disease. While diarrhoea is a clinical sign relatively easy to detect, gastric ulceration and renal disease, which are not clinical signs but a diagnosis, rely on further testing to be identified (Sanchez, 2010). Therefore the risk for under-diagnosing is high in first opinion settings and their detection would certainly be an underestimation of the true prevalence. For this reason identifying these conditions to determine the prevalence of side effects from NSAIDs usage is not ideal.

The aims of this study are the same as those in section 5.2, but applied to the data from North America described in Chapter 3. These briefly include describing the prevalence of diarrhoea and documenting whether a relationship exists between diarrhoea and the administration of drugs commonly used in equine practice.

5.3.2 Materials and methods

5.3.2.1 Prevalence of reported non-steroidal anti-inflammatory drug toxicity

The dataset of EMR from first opinion equine practice in North America, described in Chapter 3 was used. The dataset of EMRs from 312,634 animals (between 1994 and 2013; 11,699,875 rows of data) was analysed with the methods described in detail in Chapter 3. Briefly, the list of the words included in the dataset was used to identify terms that might identify NSAID toxicity, including also cases where toxicity was suspected but not necessarily confirmed.

5.3.2.2 Diarrhoea and control population

Like in section 5.2.2, diarrhoea was defined as a case where faeces had such decreased consistency as to worry an owner enough to seek veterinary advice and also to warrant it being mentioned in the EMR by the attending clinician. Different cut-offs in number of days from a previous episode were evaluated at 90 and 180 days to define a case of diarrhoea as a "new case".

Analysis was performed on the dataset from North America, after removal of cases for which the date of data input was considered unreliable as instructed by the PMSS Company. This was the case when data was imported to the current system directly from the previous incompatible PMSS. A diarrhoea dataset, including cases of diarrhoea and animals matched by practice and date from North America was created with the same methodology described in section 5.2.2.2.

A control population was selected at random from those horses matching each diarrhoea episode by date and practice for which diarrhoea was never mentioned in the EMR. Control equids were also picked from the

same practice and also if they received an examination on the same date of the episode of diarrhoea. Where no equid was examined on the same date of the diarrhoea episode, a case with the closest date available was chosen. For each episode of diarrhoea, even if from an equid that had multiple episodes, a new control subject was selected so that the number of control equids matched exactly the number of episodes of diarrhoea.

5.3.2.3 Diarrhoea prevalence determination

The prevalence estimate for diarrhoea and relative confidence intervals on the dataset from North America were calculated using the methodology described in section 5.2.2.3.

The prevalence for first opinion and referral caseloads was also assessed, using the possession of specialist qualifications as the criteria to differentiate first opinion and referral cases.

5.3.2.4 Explanatory variable: non-steroidal anti-inflammatory drugs

All cases from North America receiving NSAIDs were identified in R with the methods described in Chapter 4. Instances of drug administration contemporary to recorded clinical signs of diarrhoea were identified with the same methodology described in section 5.2.2.4. For the data from North America this was obtained for all NSAIDs together and was subsequently looked at more specifically for 3 particular drugs: flunixin meglumine, phenylbutazone and firocoxib. Ultimately 8 binary variables ("true" or "false" depending on whether use of the drug was recorded) were created to describe if usage of NSAIDs as a whole, flunixin meglumine, phenylbutazone and firocoxib was recorded within 14 or 7 days prior to the development of diarrhoea for the data from North America. Contemporary drug administration in the matched control animals was also identified.

5.3.2.5 Explanatory variable: Antimicrobials

With the same methodology described in section 5.2.2.5, 2 binary variables were created depending on whether use of antimicrobials was recorded within 14 or 7 days prior to the development of diarrhoea in the data from North America. Contemporary drug administration in the matched control animals was also identified.

5.3.2.6 Explanatory variable: Corticosteroids

Using the same methodology as in section 5.2.2.6, 4 binary variables were created depending on whether use of corticosteroids, systemic corticosteroids and corticosteroids as a whole, was recorded within 14 or 7 days prior to the development of diarrhoea in the dataset from North America. Contemporary drug administration in the matched control animals was also identified.

5.3.2.7 Explanatory variable: Laxatives

Using the same methodology as in section 5.2.2.7, one binary variable was created when administration of a laxative treatment was recorded within the 48 hours prior to the development of diarrhoea in the dataset from North America. Contemporary drug administration in the matched control animals was also identified.

5.3.2.8 Explanatory variable: Intravenous fluids

Using the same methodology as in section 5.2.2.8, one binary variable was created when intravenous fluid administration was recorded within the 48 hours prior to diarrhoea development in the dataset from North America.

Contemporary drug administration in the matched control animals was also identified.

5.3.2.9 Explanatory variable: Anthelmintic drugs

Using the same methodology as in section 5.2.2.9, 2 binary variables were created depending on whether use of anthelmintic drugs was recorded within 14 or 7 days prior to the development of diarrhoea in the dataset from North America. Contemporary drug administration in the matched control animals was also identified.

5.3.2.10 Statistical analysis

Data analysis was repeated on the data from North America following the methodology described in depth in section 5.2.2.10.

5.3.2.11 Post-hoc analysis to account for comorbidities

Similar to section 5.2.2.11, the effect of comorbidities was evaluated *post-hoc* to further explain some of the findings of the initial analysis and account for the possible confounding effect of comorbidities such as colic and orthopaedic disease. These were used to identify patients suspected, investigated or treated for abdominal discomfort or an orthopaedic condition in zero to seven days or seven to 14 days, or at any other point in the EMRs, preceding the development of diarrhoea. These variables were mutually exclusive. Conditional logistic regression was then used to evaluate whether inclusion of one or both comorbidities altered the statistical significance and odd ratios of variables included in the model.

Inclusion, exclusion and re-inclusion dictionaries used for the analysis are available in Appendix 5.

The study was approved by Ethics and Welfare Committee of the School of Veterinary Medicine at the University of Glasgow.

5.3.3 Results

5.3.3.1 Prevalence of reported non-steroidal anti-inflammatory drugs toxicity

Of the total of 11,699,875 rows of data from 312,634 equids from North America, NSAID toxicity was suspected or diagnosed in only 19 animals. The demographic details of the population in this dataset are described in Chapter 3. The overall prevalence of NSAID toxicity in this population from North America was therefore 0.007% (95% CIs: 0.00-0.01%).

5.3.3.2 Diarrhoea and Control population

The overall dataset from North America included records from 312,634 equids, which were then narrowed down to a total of 225,777 equids with a reliable date of data entry to the PMSS between 1998 and 2013 (10,483,807 rows of data).

A 90 day cut-off was arbitrarily based on the expert opinion of the author. Interestingly a longer cut-off (e.g. 6 months) would have decreased the number of diarrhoea episodes by approximately 1% and the overall diarrhoea prevalence by 0.3%.

The dataset with valid dates included a total 4741 equids that suffered at least one episode of diarrhoea for a total of 5122 episodes of diarrhoea. Of these, 4430 had reported a single episode of diarrhoea, 259 reported 2 episodes, 38 reported 3 episodes, 12 reported 4 episodes and the remaining 2 equids reported 5 and 7 episodes respectively. The mean age of the population that suffered diarrhoea was 10.4 years (median 9 years,

range: 0-40 years), while age was not available for 454 equids. All cases were equids from a total of 265 breeds or crosses. Diarrhoea was reported in 1991 females and 2671 males of which 2208 were geldings, 462 entire males and 1 was recorded simply as male. Gender had not been recorded in 460 equids with diarrhoea.

The control population included a total of 5122 equids and no control animal appeared more than once in the dataset. The mean age of the control population was 11.0 years (median 10 years, range: 0-40 years), while age was not available for 982 animals. All cases were equids from a total of 282 breeds or crosses. Control equids included 1843 females and 2346 males of which 2132 were geldings, 212 entire males and 2 were recorded simply as male. Gender had not been recorded in 982 equids.

The dataset including all cases with diarrhoea and their matched control is from now on referred to as the "diarrhoea dataset" and included a total of 9863 equids, 4741 that suffered at least one episode of diarrhoea and 5122 equids that never had diarrhoea reported in the records.

5.3.3.3 Diarrhoea prevalence determination

A total of 4741 equids from the population of 225,777 individuals with a valid date of data entry suffered at least one episode of diarrhoea severe enough to be mentioned in the EMR (diarrhoea prevalence: 2.10%; 95% CIs: 2.04-2.16%).

Using possession of a specialist diploma to differentiate first opinion from referral caseload, the dataset included 211,614 equids from the first opinion population from which 5058 episodes of diarrhoea were detected from 4682 equids, with a prevalence estimate of reported diarrhoea in first opinion practice of 2.39% (95% CIs: 2.33-2.46%). The referral population included a total of 51,111 equids with 64 episodes of diarrhoea

from only 63 equids (prevalence 0.13% - 95% CIs: 0.09-0.16%). The data to differentiate first opinion and referral caseloads was not considered reliable and not included in the following analysis.

5.3.3.4 Explanatory variable: non-steroidal anti-inflammatory drugs

Of the 225,777 that had valid dates recorded, administration of NSAIDs was recorded at least once in 90,886 equids, with a calculated prevalence of NSAIDs usage of 40.25% (95% CIs: 40.05-40.46%).

A total of 2596 animals, out of the total of 9862 equids in the diarrhoea dataset from North America, received NSAIDs in the preceding 14 days. Of these, 1608 were from the diarrhoea group and 988 were from the control group. The overall prevalence of NSAID usage in the 14 days preceding the development of diarrhoea was 33.92% (CIs: 32.58-35.28%), while in the control group this was 19.29% (95% CIs: 18.23-20.39%).

The prevalence of NSAIDs usage within 14 days of the 'case date' was 33.92% (1608 animals; 95% CIs: 32.58-35.28%) in equids with diarrhoea and 19.29% (988 animals; 95% CIs: 18.23-20.39%) in equids in the control group. The prevalence of phenylbutazone usage within 14 days of the 'case date' was 9.98% (473 animals; 95% CIs: 9.16-10.86%) in equids with diarrhoea and 11.32% (580 animals; 95% CIs: 10.48-12.22%) in equids in the control group. The prevalence of flunixin meglumine usage within 14 days of the 'case date' was 26.62% (1262 animals; 95% CIs: 25.38-27.90%) in equids with diarrhoea and 10.39% (532 animals; 95% CIs: 9.58-11.25%) in equids in the control group. The prevalence of firocoxib usage within 14 days of the 'case date' was 1.48% (74 animals; 95% CIs: 1.17-1.86%) in equids with diarrhoea and 1.44% (74 animals; 95% CIs: 1.15-1.81%) in equids in the control group.

The prevalence of NSAIDs usage within 7 days of the 'case date' was 31.51% (1494 animals; 95% CIs: 30.21-32.85%) in equids with diarrhoea and 15.13% (775 animals; 95% CIs: 14.18-16.14%) in equids in the control group. The prevalence of phenylbutazone usage within 7 days of the 'case date' was 8.46% (401 animals; 95% CIs: 7.70-9.28%) in equids with diarrhoea and 8.77% (449 animals; 95% CIs: 8.02-9.57%) in equids in the control group. The prevalence of flunixin meglumine usage within 7 days of the 'case date' was 25.06% (1188 animals; 95% CIs: 23.84-26.31%) in equids with diarrhoea and 7.91% (405 animals; 95% CIs: 7.20-8.68%) in equids in the control group. The prevalence of firocoxib usage within 7 days of the 'case date' was 1.24% (59 animals; 95% CIs: 0.97-1.60%) in equids with diarrhoea and 0.82% (42 animals; 95% CIs: 0.61-1.11%) in equids in the control group.

Results of prevalence of drug usage are summarised in Table 5.7.

Table 5.7: Prevalence of NSAIDs usage 14 and 7 days before a 'case date' from 4741 equids from North America that suffered 5122 episodes of diarrhoea and 5122 control equids that never had diarrhoea.

Drug	Days	Total	D	Prevalence (95% CIs)	C	Prevalence (95% CIs)
NSAIDs	14	2596	1608	33.92 (32.58-35.28)	988	19.29 (18.23-20.39)
	7	2269	1494	31.51 (30.21-32.85)	775	15.13 (14.18-16.14)
Phenylbutazone	14	1053	473	9.98 (9.16-10.86)	580	11.32 (10.48-12.22)
	7	850	401	8.46 (7.7-9.28)	449	8.77 (8.02-9.57)
Flunixin meglumine	14	1794	1262	26.62 (25.38-27.9)	532	10.39 (9.58-11.25)
	7	1593	1188	25.06 (23.84-26.31)	405	7.91 (7.2-8.68)
Firocoxib	14	144	70	1.48 (1.17-1.86)	74	1.44 (1.15-1.81)
	7	101	59	1.24 (0.97-1.6)	42	0.82 (0.61-1.11)

Days: maximum number of days between drug administration and development of diarrhoea; Total: total number of equids receiving that drug; D: diarrhoea group; C control group; OR: odds ratio.

5.3.3.5 Explanatory variable: Antimicrobials

Of the total of 312,634 animals administration of antimicrobials was recorded at least once in the EMR of 79,301 patients with a prevalence for antimicrobial usage of 25.37% (95% CIs: 25.22-25.52%). Of the 225,777 that

had valid dates recorded, administration of antimicrobial was recorded in 61,644 equids with a prevalence of 27.30% (95% CIs: 27.12-27.49%).

A total of 1797 animals, out of the total of 9862 equids in the diarrhoea dataset, received antimicrobials in this timeframe with an overall prevalence of 18.22% (95% CIs: 17.47-19.00%). The prevalence of antimicrobial usage within 14 days of the 'case date' was 24.80% (1176 animals; 95% CIs: 23.60-26.05%) in equids with diarrhoea and 12.12% (621 animals; 95% CIs: 11.26-13.05%) in equids in the control group.

In the 7 days before the reference date, a total of 1610 equids received antimicrobials with an overall prevalence of 16.33% (95% CIs: 15.61-17.07%) in this week period. The prevalence of antimicrobial usage within 7 days of the 'case date' was 23.14% (1097 animals; 95% CIs: 21.96-24.36%) in equids with diarrhoea and 10.02% (513 animals; 95% CIs: 9.22-10.87%) in equids in the control group.

5.3.3.6 Explanatory variable: Corticosteroids

Of the total of 312,634 animals administration of corticosteroids was recorded at least once in the EMR of 75,391 patients with a prevalence for corticosteroids usage of 24.12% (95% CIs: 23.97-24.27%). Of the 225,777 that had valid dates recorded administration was recorded in 57,521 equids with a prevalence of corticosteroid usage of 25.48% (95% CIs: 25.30-25.66%).

A total of 730 animals, out of the total of 9862 equids in the diarrhoea dataset, received corticosteroids in the 14 days preceding the 'case date' with an overall prevalence of 7.40% (95% CIs: 6.90-7.94%).

The prevalence of corticosteroids usage within 14 days of the ‘case date’ was 6.12% (290 animals; 95% CIs: 5.47-6.84%) in equids with diarrhoea and 8.59% (440 animals; 95% CIs: 7.85-9.39%) in equids in the control group.

In the 7 days before the reference date, a total of 535 equids received corticosteroid with an overall prevalence of 5.42% (95% CIs: 4.99-5.89%). The prevalence of corticosteroid usage within 7 days of the ‘case date’ was 4.64% (220 animals; 95% CIs: 4.08-5.28%) in equids with diarrhoea and 6.15% (315 animals; 95% CIs: 5.52-6.84%) in equids in the control group.

When looking specifically at usage of systemic corticosteroid (dexamethasone, prednisolone, prednisone), the prevalence over the total population of 312,634 equids was 14.65% (95% CIs: 15.52-14.77%) and in the subpopulation of 225,777 animals the prevalence was 15.40% (95% CIs: 15.25-15.55%), with a total number of treated animals of 45,792 and 34,772 respectively.

In the 14 days period preceding the reference date, a total of 475 equids received corticosteroids systemically with an overall prevalence of 4.82% (95% CIs: 4.41-5.26%). The prevalence of systemic corticosteroids usage within 14 days of the ‘case date’ was 4.35% (206 animals; 95% CIs: 3.80-4.96%) in equids with diarrhoea and 5.25% (269 animals; 95% CIs: 4.67-5.90%) in equids in the control group.

In the 7 days before the reference date, a total of 356 equids received corticosteroids systemically with an overall prevalence of 3.61% (95% CIs: 3.26-3.40%) in this timeframe. The prevalence of corticosteroids usage within 7 days of the ‘case date’ was 3.40% (161 animals; 95% CIs: 2.92-3.95%) in equids with diarrhoea and 3.81% (195 animals; 95% CIs: 3.32-4.37%) in equids in the control group.

5.3.3.7 Explanatory variable: Laxatives

Of the total of 312,634 animals administration of laxatives was recorded at least once in the EMR of 20,572 patients with a prevalence for laxative usage of 6.58% (95% CIs: 6.49-6.67%). Of the 225,777 that had valid dates recorded administration of laxatives was recorded in 16,925 equids with a prevalence of 7.50% (95% CIs: 7.39-7.61%).

In the diarrhoea dataset laxative treatment was included if administered in the 2 days preceding the reporting of diarrhoea. A total of 538 animals, out of the total of 9862 equids in the diarrhoea dataset, received at least one laxative treatment in this timeframe with an overall prevalence of 5.46% (95% CIs: 5.02-5.92%). The prevalence of laxative treatment within 2 days of the 'case date' was 10.08% (478 animals; 95% CIs: 9.26-10.97%) in equids with diarrhoea and 1.17% (60 animals; 95% CIs: 0.91-1.50%) in equids in the control group.

5.3.3.8 Explanatory variable: Intravenous fluids

Of the total of 312,634 animals administration of intravenous fluids was recorded at least once in the EMR of 26,642 patients with a prevalence of fluid administration intravenously of 8.52% (95% CIs: 8.43-8.62%). Of the 225,777 that had valid dates recorded administration of fluids parenterally was recorded in 24,090 equids with a prevalence of 10.67% (95% CIs: 10.54-10.80%).

In the diarrhoea dataset intravenous fluid administration was included if it occurred in the 2 days preceding the reporting of diarrhoea. A total of 642 animals, out of the total of 9862 equids in the diarrhoea dataset, received fluid intravenously in this timeframe with an overall prevalence of 6.51% (95% CIs: 6.04-7.01%). The prevalence of intravenous fluids administration in the 2 days preceding the 'case date' was 10.80% (512 animals; 95% CIs:

9.95-11.71%) in equids with diarrhoea and 2.54% (130 animals; 95% CIs: 2.14-3.01%) in equids in the control group.

5.3.3.9 Explanatory variable: Anthelmintic drugs

Of the total of 312,634 animals administration of anthelmintic drugs was recorded at least once in the EMR of 45,368 patients with a prevalence for anthelmintic drug usage of 14.51% (95% CIs: 14.39-14.64%). Of the 225,777 for which the date was valid, administration of anthelmintic drug was recorded in 31,998 equids with a prevalence of 14.17% (95% CIs: 14.03-14.32%).

A total of 246 animals, out of the total of 9862 equids in the diarrhoea dataset, received anthelmintics in this timeframe with an overall prevalence of 2.49% (95% CIs: 2.20-2.82%). The prevalence of anthelmintic usage within 14 days of the 'case date' was 2.19% (140 animals; 95% CIs: 1.81-2.65%) in equids with diarrhoea and 2.77% (142 animals; 95% CIs: 2.36-3.26%) in equids in the control group.

In the 7 days before the reference date, a total of 1153 equids received anthelmintic with an overall prevalence of 1.55% (95% CIs: 1.33-1.81%). The prevalence of anthelmintic usage within 7 days of the 'case date' was 1.62% (77 animals; 95% CIs: 1.30-2.03%) in equids with diarrhoea and 1.48% (76 animals; 95% CIs: 1.19-1.85%) in equids in the control group.

Table 5.8: Prevalence of drug usage in 225,777 equids from North America. The data reflects usage of each drug at some point in the animal EMR.

Drug	Equids	Prevalence (%)	95% CIs (%)
Antimicrobials	61644	27.30	27.12-27.49
Corticosteroids	57,521	25.48	25.30-25.66
Systemic corticosteroids	34,772	15.40	15.25-15.55
Laxatives	16,925	7.50	7.39-7.61
Intravenous fluids	24,090	10.67	10.54-10.80
Anthelmintic drugs	31,998	14.17	14.03-14.32

Prevalence data for drug usage is summarised in Table 5.8. Prevalence data on drug usage in the diarrhoea dataset is summarised in Table 5.9.

Table 5.9: Prevalence of drug usage 14 and 7 days before a 'case date' from 4741 equids from North America that suffered 5122 episodes of diarrhoea and 5122 control equids that never had diarrhoea.

Drug	Days	Total	D	Prevalence (95% CIs)	C	Prevalence (95% CIs)
Antimicrobials	14	1797	1176	24.8 (23.6-26.05)	621	12.12 (11.26-13.05)
	7	1610	1097	10.02 (9.22-10.87)	513	10.8 (9.95-11.71)
Corticosteroids	14	730	290	6.12 (5.47-6.84)	440	8.59 (7.85-9.39)
	7	535	220	4.64 (4.08-5.28)	315	6.15 (5.52-6.84)
Systemic corticosteroids	14	475	206	4.35 (3.8-4.96)	269	5.25 (4.67-5.9)
	7	356	161	3.4 (2.92-3.95)	195	3.81 (3.32-4.37)
Laxatives	2	538	478	10.08 (9.26-10.97)	60	1.17 (0.91-1.5)
Intravenous fluids	2	642	512	10.8 (9.95-11.71)	130	2.54 (2.14-3.01)
Anthelmintic drugs	14	246	104	2.19 (1.81-2.65)	142	2.77 (2.36-3.26)
	7	153	77	1.62 (1.3-2.03)	76	1.48 (1.19-1.85)

Days: maximum number of days between drug administration and development of diarrhoea; Total: total number of equids receiving that drug; D: diarrhoea group; C control group.

5.3.3.10 Statistical analysis

Univariable logistic regression

When looking at drug administration in the 14 days preceding the development of diarrhoea, development of diarrhoea was 2.04 (95% CIs: 1.84-2.26; $p<0.001$) times more likely with NSAIDs administration. Diarrhoea was 0.78 times as likely with phenylbutazone administration (95%CIs: 0.68-0.90; $p<0.001$). Equids with diarrhoea were 3.13 (95% CIs: 2.76-3.555; $p<0.001$) times more likely to have received flunixin meglumine. The presence of diarrhoea was not significantly associated with the administration of firocoxib ($p=0.737$). Diarrhoea was 0.52 (95%CIs: 0.35-0.75; $p<0.001$) times as likely with administration of ketoprofen.

Diarrhoea was 2.34 (95% CIs: 2.07-2.63; p<0.001) times more likely with antimicrobial administration. Diarrhoea was 0.61 (95 CIs: 0.52-0.72; p<0.001) times as likely with corticosteroids administration, 0.73 (95 CIs: 0.60-0.90; p=0.003) times as likely with systemic corticosteroids and 0.7 (95% CIs: 0.53-0.92; p=0.012) times as likely with the administration of anthelmintic drugs.

When looking at drug administration in the 7 days preceding the development of diarrhoea, development of diarrhoea was 2.51 (95% CIs: 2.25-2.80; p<0.001) times more likely with NSAIDs administration. Equids with diarrhoea were 3.98 (95% CIs: 3.46-4.56; p<0.001) times more likely to have received flunixin meglumine. There was no significant relationship between the administration of either phenylbutazone (p=0.08) or firocoxib (p=0.09) in the week before development of diarrhoea. Equids with diarrhoea were 0.58 (95% CIs: 0.38-0.87; p=0.009) times as likely to have received ketoprofen than control animals. Equids with diarrhoea were 2.68 (95% CIs: 2.36-3.05, p<0.001) times more likely to have received antimicrobials than controls in the 7 days preceding the development of diarrhoea. Diarrhoea was 0.66 (95% CIs: 0.54-0.80; p<0.001) times as likely following corticosteroid administration. There was no significant relationship between development of diarrhoea and administration of either systemic corticosteroids (p=0.07) or anthelmintic treatment (p=0.9) in this 7-day period.

Diarrhoea was significantly more likely with administration of laxatives (OR: 10.18; 95% CIs: 7.64-13.57; p<0.001) and intravenous fluid therapy (OR: 4.89; 95% CIs: 3.94-6.06; p<0.001) in the previous 2 days.

Univariable logistic regression results are summarised in Table 5.10.

Table 5.10: Univariable logistic regression of variables considered for the multivariable conditional logistic regression. The dataset includes 4741 equids from North America that suffered 5122 episodes of diarrhoea and 5122 control equids that never had diarrhoea

Treatment	Days	Total	D	C	P-value	OR	95% CIs
NSAIDs	14	2596	1608	988	<0.001	2.04	1.84-2.26
	7	2269	1494	775	<0.001	2.51	2.25-2.80
Phenylbutazone	14	1053	473	580	<0.001	0.78	0.69-0.90
	7	850	401	449	0.08	0.97	0.93-1.01
Flunixin meglumine	14	1794	1262	532	<0.001	3.13	2.76-3.55
	7	1593	1188	405	<0.001	3.98	3.46-4.56
Firocoxib	14	144	70	74	0.74	0.99	0.91-1.07
	7	101	59	42	0.09	1.09	0.99-1.20
Ketoprofen	14	122	42	80	<0.001	0.52	0.35-0.75
	7	98	36	62	0.009	0.58	0.38-0.87
Antimicrobials	14	1797	1176	621	<0.001	2.34	2.07-2.63
	7	1610	1097	513	<0.001	2.68	2.36-3.05
Corticosteroids	14	730	290	440	<0.001	0.61	0.52-0.72
	7	535	220	315	<0.001	0.66	0.54-0.80
Systemic corticosteroids	14	475	206	269	0.003	0.73	0.60-0.90
	7	356	161	195	0.07	0.95	0.90-1.01
Laxatives	2	538	478	60	<0.001	10.18	7.64-13.57
Intravenous Fluid Therapy	2	642	512	130	<0.001	4.89	3.94-6.06
Anthelmintic drugs	14	246	104	142	0.012	0.7	0.53-0.92
	7	153	77	76	0.9	1.01	0.93-1.09

Days: maximum number of days between drug administration and development of diarrhoea; Total: total number of equids receiving that drug; D: diarrhoea group; C control group; OR: odds ratio.

Multivariable conditional logistic regression

The best fitting model without inclusion of interaction terms included the following parameters (Table 5.11):

iv. Drugs dispensed 2 days before diarrhoea: laxatives, intravenous fluids

v. Drugs dispensed 7 days before diarrhoea: flunixin, antimicrobials

vi. Drugs dispensed 14 days before diarrhoea: phenylbutazone, ketoprofen, corticosteroids and anthelmintic drugs.

The interaction terms between corticosteroids and antimicrobials, flunixin and phenylbutazone and antimicrobials and phenylbutazone also improved model fit significantly and were therefore included.

Table 5.11: Multivariable conditional logistic regression model showing variables significantly associated with the risk of developing diarrhoea in North America. The dataset includes 4741 equids that suffered 5122 episodes of diarrhoea and 5122 control equids that never had diarrhoea

Treatment	Days	Total	D	C	P-value	OR	95% CIs
Flunixin meglumine	7	1593	1188	405	<0.001	2.84	2.32-3.48
Phenylbutazone	14	1053	473	580	<0.001	0.56	0.44-0.71
Ketoprofen	14	122	42	80	<0.001	0.10	0.04-0.24
Corticosteroids	14	730	290	440	<0.001	0.34	0.27-0.45
Antimicrobials	7	1610	1097	513	<0.001	1.63	1.33-1.99
Anthelmintic drugs	14	246	104	142	<0.001	0.58	0.43-0.79
Laxatives	2	538	478	60	<0.001	4.57	3.33-6.28
Intravenous fluid therapy	2	642	512	130	<0.001	2.30	1.80-2.94
Corticosteroids*Antimicrobials	14*7	303	173	130	0.022	0.89	0.58-1.38
Antimicrobials*Phenylbutazone	7*14	546	311	235	0.017	1.39	0.93-2.07
Flunixin*Phenylbutazone	7*14	342	185	157	<0.001	0.79	0.52-2.07

Days: maximum number of days between drug administration and development of diarrhoea; Total: total number of equids receiving that drug; D: diarrhoea group; C control group; OR: odds ratio; Corticosteroids*Antimicrobials: interaction term true if an equid received both corticosteroids 14 days before the 'case date' and antimicrobials 7 days before the 'case date'; Antimicrobials*Phenylbutazone: interaction term true if an equid received antimicrobials 7 days and phenylbutazone 14 days prior to the 'case date' respectively; Flunixin*Phenylbutazone: interaction term true if an equid received flunixin meglumine 7 days and phenylbutazone 14 days prior to the 'case date' respectively.

Equids with diarrhoea were 2.84 (95% CIs: 2.31-3.48; p<0.001) times more likely to have received flunixin meglumine in the 7 days preceding the development of diarrhoea, 0.56 (95% CIs: 0.44-0.71; p<0.001) times as likely to have received phenylbutazone in the preceding 14 days, 0.10 (95% CIs: 0.04-0.24) times as likely to have received ketoprofen in the preceding 14 days, 0.34 (95% CIs: 0.27-0.45; p<0.001) times as likely to have received corticosteroids in the preceding 14 days, 1.63 (95% CIs: 1.33-1.99; p<0.001) times more likely to have received antimicrobials in the preceding 7 days, 0.58 (95% CIs: 0.43-0.79; p<0.001) times as likely to have received an anthelmintic drug in the preceding 14 days, 4.57 (95% CIs: 3.33-6.28; p<0.001) times more likely to have received a laxative in the preceding 2 days and 2.30 (95% CIs: 1.80-2.94; p<0.001) times more likely to have received intravenous fluid therapy in the 2 days preceding the development of diarrhoea. Further, three interaction terms were

significant and were included in the model. Equids with diarrhoea were 0.89 (95% CIs: 0.58-1.38; p=0.022) times as likely to have received both corticosteroids in the 14 days and antimicrobials in the 7 days prior to the development of diarrhoea. Equids with diarrhoea were 1.39 (95% CIs: 0.93-2.07; p<0.017) times more likely to have received both antimicrobials in the 7 days and phenylbutazone in the 14 days prior to the development of diarrhoea. Equids with diarrhoea were 0.79 (95% CIs: 0.52-2.07; p<0.001) times as likely to have received both flunixin meglumine in the 7 days and phenylbutazone in the 14 day prior to the development of diarrhoea. For all significant interaction terms the 95% confidence intervals spanned across 1. This makes interpretation of their overall clinical meaning troublesome, but they were still included in the final model as their inclusion significantly improved the AIC.

The variable for firocoxib was tested and not significant (p=0.5). Several interaction terms between the included explanatory variables were tested and those that were not significant included flunixin meglumine and antimicrobials (p=0.4), corticosteroids and phenylbutazone (p=0.4) and corticosteroids and flunixin meglumine (p=0.7), ketoprofen and corticosteroids (p=0.9) and ketoprofen and antimicrobials (p=0.7).

5.3.3.11 Post-hoc analysis to account for comorbidities

The dataset of 312,634 animals from North America included a total of 27,166 equids suffering at least one episode of abdominal discomfort at some point in the EMRs dataset with a total prevalence of 8.7% (95% CIs: 8.6-8.8%). Of the 225,777 equids with a valid date 21,682 equids suffered at least one episode of colic; prevalence of colic was 9.6% (95% CIs: 9.5-9.7%). In the diarrhoea dataset the number of equids suffering at least one episode of colic was 4669 of which 3215 (3541 episodes) were in the diarrhoea group and 1454 (1454 episodes) in the control group.

The dataset of 312,634 animals included a total of 128,384 equids in the orthopaedic group for a total prevalence of 41.1% (95% CIs: 40.9-41.2%). Of the 225,777 equids with a valid date 94,840 suffered at least one episode of orthopaedic disease; prevalence of orthopaedic disease was 42.0% (95% CIs: 41.8-42.2%). In the diarrhoea dataset the number of equids suffering at least one episode of orthopaedic disease was 7063 of which 3375 (3717 episodes) were in the diarrhoea group and 3688 (3688 episodes) in the control group.

The best fitting model included the following parameters:

vii. Drugs dispensed 2 days before diarrhoea: intravenous fluids
viii. Drugs dispensed 7 days before diarrhoea: antimicrobials
ix. Drugs dispensed 14 days before diarrhoea: corticosteroids, phenylbutazone, anthelmintic drugs
x. Colic 7 days before diarrhoea
xi. Orthopaedic disease 7 and 14 days before diarrhoea
xii. Colic and/or orthopaedic disease at some point in the EMR database before diarrhoea from 14 days before diarrhoea developed.

All previous interaction terms became non-significant once colic and orthopaedic variables were added to the model and have therefore not been included. The only significant interaction terms between drugs included administration of corticosteroids and antimicrobials in the week prior to diarrhoea ($p<0.001$). The only significant interaction terms between drug and comorbidity terms included administration of phenylbutazone and orthopaedic disease ($p=0.004$). Inclusion of any of these interaction terms did not improve the AIC sufficiently to be maintained in the final model. Interactions between phenylbutazone and colic in the first week ($p=0.4$) or at any time before diarrhoea ($p=0.1$) were not significant. The interaction between firocoxib and colic was also not significant ($p=0.06$). Odd ratios, confidence interval and level of

significance for all the variables in the *post-hoc* model are summarised in table 5.12.

Table 5.12: *Post-hoc* multiple conditional logistic regression model showing variables significantly associated with the risk of developing diarrhoea in North America. The dataset includes 4741 equids that suffered 5122 episodes of diarrhoea and 5122 control equids that never had diarrhoea

Treatment	Days	Total	D	C	P-value	OR	95% CIs
Phenylbutazone	14	1053	473	580	<0.001	0.67	0.55-0.81
Corticosteroids	14	730	290	440	<0.001	0.53	0.42-0.67
Antimicrobials	7	1610	1097	513	<0.001	2.27	1.90-2.71
Anthelmintic drugs	14	246	104	142	0.020	0.66	0.46-0.94
Intravenous fluids	2	642	512	130	<0.001	2.09	1.52-2.89
Colic	7	2927	2630	297	<0.001	17.5	14.60-21.04
Colic	life	1735	986	749	<0.001	1.43	1.23-1.68
Ortho	7	3488	1986	1502	<0.001	1.54	1.37-1.74
Ortho	life	4458	1941	2517	<0.001	0.58	0.51-0.65

Days: maximum number of days between drug administration and development of diarrhoea; Total: total number of equids receiving that drug; D: diarrhoea group; C control group; OR: odds ratio; Colic-life: true if at least one episode of colic is recorded in the EMR at any time from before 14 days from development of diarrhoea; Colic: true if an animal suffered an episode of colic in the 7 days preceding diarrhoea; Ortho-life: true if at least one orthopaedic investigation or treatment is recorded in the EMR at any time from before 14 days from development of diarrhoea; Ortho: true if an animal was investigated or treated for an orthopaedic related condition in the previous 7 days prior to diarrhoea.

5.3.4 Discussion

This study presents several other interesting findings, which were not within the main objective of the study. Prevalence of diarrhoea, colic and orthopaedic disease has not been reported before on such a large population in equine veterinary practice in North America. The prevalence estimate for diarrhoea reported in this study was 2.1% obtained from the population of 225,177 patients with a valid date of data entry to the PMSS. The overall population of 312,634 animals is more comprehensive but dates of the older data were wrong; in addition whether all patients seen by each practice in those years were actually entered in the system is unknown. However, the prevalence estimates from both populations were similarly around 2%. This value is the prevalence of diarrhoea requiring veterinary attention/intervention; the actual prevalence of diarrhoea in the population would be impossible to determine. However, the need for inclusion of cases so mild that they do not require veterinary intervention

would be also questionable. It is important to point out that while some animals with diarrhoea had more than one episode, the cut-off of 90 diarrhoea-free days between episodes was chosen to ensure that if a new episode was recorded this was unlikely to be the continuation of the previous episode. While a horse might have chronic intermitted diarrhoea for several months, in such cases examinations would be expected to be more frequent than every 3 months.

Further, prevalence data from this dataset from North America needs to be interpreted in light of the mixed first opinion and referral nature of the data. Since referral cases are for the vast majority hospitalised, they are intrinsically at a higher risk of developing nosocomial bacterial infections, which might lead to diarrhoea (Ekiri et al., 2010). Further, hospitalised patients are monitored more closely and even a single bout of diarrhoea is likely to be detected and recorded. For these reasons, one might argue that a referral population might present a higher prevalence of diarrhoea. The data did not allow a clear differentiation between first opinion and referral cases and therefore the results include both types of populations. Without a purpose built feature of the PMSS to differentiate first opinion from referral cases an alternative would be to assume that clinicians holding a post-graduate specialist diploma (ACVIM, ACVS, ACT, ECEIM, ECVS) might be the ones involved with a referral caseload. However, it has to be acknowledged that whilst this is true in many cases, a specialist qualification is not a mandatory requirement to accept referral cases. Further, a variable proportion of the caseload of many veterinary specialists in private practice might include cases that are not referred by another veterinary surgeon. Finally, cases referred internally from a veterinarian from the first opinion service to a specialist, depending on the practice's standard operating procedures, might remain under the name of the first opinion veterinarian in the PMSS. This complicates the picture further and since no clear differentiation between first opinion and referral population was possible, this was not included in the analysis

and the population was treated as a whole. The discrepancy between first opinion and referral is large and unlikely to reflect the true prevalence of diarrhoea in a referral population.

The effect of practice was not evaluated as practice was used as a criterion to match diarrhoea cases with their control for the conditional logistic regression analysis. It therefore would have been inappropriate to include practice as an explanatory variable in the analysis. North America is a vast continent and marked differences in temperature, humidity and precipitations exist between different geographical locations on the continent. These differences play a determinant role in infectious disease prevalence and might affect diarrhoea prevalence estimates. Local/seasonal outbreaks of diarrhoea might have affected the prevalence of diarrhoea in some areas. Further, different practices might have different expertise, standard operating procedures and protocols for disease diagnosis and treatment, which might also introduce bias. The effect of practice could be investigated by assessing whether the some practice was proportionally overrepresented in the diarrhoea group, but this was beyond the scope of this study and was not performed.

The prevalence of NSAIDs usage in the 14 days prior to the development of diarrhoea was 33.9% (95% CIs: 32.6-35.3%) and 19.3% (95% CIs: 18.2-20.4%) in the control group. In fact the reported overall prevalence of NSAIDs usage was 36.9% (95% CIs: 36.8-37.1%) and 40.7% (95% CIs: 40.5-40.9%) for the overall population of 312,634 and 225,777 animals respectively. While this study seem to suggest a low prevalence of NSAIDs usage in the diarrhoea group, this actually refers to the use in a specific 14 day period prior to the development of diarrhoea and so it is not directly comparable to the overall prevalence which reflects the proportion of equids receiving the drug at some point throughout their lifetime in the EMR database. For a discussion on the overall prevalence of NSAIDs usage in the data from North America please refer to Chapter 4.

Similar to the study from the United Kingdom, this study from North America also reports the prevalence of use of several variables in the general population and in equids with diarrhoea.

Antimicrobials are often administered in combination with NSAIDs, not only to treat on-going bacterial infections and the associated inflammatory response but also preventatively in the perioperative period in elective and emergency surgery cases.

This study also reported a relatively high prevalence of corticosteroid usage in North America since at least one in four equids received corticosteroids at least once. The corticosteroid category includes all corticosteroids, from those for intra-articular administration to systemic treatments. However, treatment with both systemic and non-systemic corticosteroids is not mutually exclusive, as many equids had received either treatment, at different times during the course of their life. Differentiation between systemic corticosteroids and corticosteroids for intra-articular administration is relevant as systemic corticosteroids might affect the gastrointestinal tract to a greater extent (Sanchez, 2010).

Data on the prevalence of use of laxatives is in line with the overall prevalence of colic as these drugs are likely to be used with a proportion of colic cases suffering from impactions. The prevalence of intravenous fluid administration includes all cases receiving fluids for the management of hypovolaemia or dehydration. However the prevalence of hypovolaemia or dehydration was not investigated in the study so its prevalence cannot be compared to that of intravenous fluid use.

Anthelmintic drugs are often administered to equids, for treatment as well as prevention of intestinal parasitic infections, which could result in diarrhoea. This study also reported a relatively low prevalence of

anthelmintic usage. The prevalence of anthelmintic drug usage in the 14 days prior to the development of diarrhoea was 2.2% (95% CIs: 1.8-2.7%) and 2.8% (95% CIs: 2.4-3.3%) in the control group. The overall prevalence of anthelmintic drug usage was 14.5% (95% CIs: 14.4-14.6%) and 14.2% (95% CIs: 14.0-14.3%) for the overall population of 312,634 and 225,777 animals respectively. This much lower prevalence of anthelmintic treatment in the diarrhoea group is a consequence of the inclusion of cases where the drug was administered only 14 days prior to diarrhoea. Overall, it is important to highlight that in North America owners can purchase anthelmintic drugs over the counter without the need for a prescription, which could result in a serious underestimation of the prevalence of their use. Undoubtedly, explanatory variables consisting of drugs that are not under the exclusive control of veterinary professional, pose a serious challenge to epidemiologic research, as there would be no reliable way to assess their actual usage.

The results of this analysis supports existing evidence that development of diarrhoea is more likely following administration of NSAIDs than in equine patients not receiving these drugs, but also the OR close to 1 suggests that the risk is small. This applies as long as the currently widespread practice of observing recommended dose regimen is maintained. Data on dose used and course length is necessary to actually confirm that veterinary surgeons and owners keep within the recommended dose regimens to fully confirm that current dose recommendation are safe. Conditional logistic regression confirmed a statistically significant relationship between flunixin meglumine administration and development of diarrhoea. An unexpected finding was that development of diarrhoea had an odds ratio less than 1 for the administration of non-selective COX inhibitors such as phenylbutazone and ketoprofen. The finding that there was no significant relationship between diarrhoea and previous administration of firocoxib was in line with the findings of the study described in Chapter 2 of this thesis, where this drug has been shown to have a safer profile *in vitro* as

well as *ex vivo* than other NSAIDs such as phenylbutazone or flunixin meglumine. Administration of other drugs was also included in the model as these could act as a confounding as some are often administered in combination with NSAIDs in equine clinical practice.

In North America it is common practice to use phenylbutazone to treat pain and inflammation as a consequence of orthopaedic disease. Equids in the orthopaedic group might also have a healthier gastrointestinal tract compared to that of a population with other diseases more likely to cause systemic illness. This theory is substantiated by the fact that diarrhoea is 0.58 times as likely in patients suffering orthopaedic disease at any time in the dataset up to two weeks before the development of diarrhoea. Further, equids with diarrhoea were also 1.54 times more likely to have had an orthopaedic disease investigated or treated in the week before diarrhoea was reported. This difference might be explained as equids undergoing orthopaedic investigation or treatment often receive drugs which may not be accounted for in the analysis, such as sedatives or anaesthetics, or undergo the stress of travelling to the hospital and might then be more prone to diarrhoea. This effect might become less relevant a week later as the patient has time to recover. Finally, also other undetermined comorbidities might also be involved but were not accounted for in the *post-hoc* model, however the ones included were judged to be the most relevant clinically.

The confounding effect on diarrhoea of underlying gastrointestinal disease, was somewhat accounted for by the colic variables. Diarrhoea was 17 times more likely after a recent (<7 days) episode of colic, or 1.5 times more likely in horses that had previously suffered of colic at any time during life. This suggests that diarrhoea is more likely in equids that are more susceptible to gastrointestinal disease, either infectious, inflammatory or dietary in origin, manifested by discomfort at some point in life.

No significant interaction was present between colic and NSAIDs administration. Inclusion of colic to the model changed the significance of the effect of flunixin meglumine in the *post-hoc* analysis (p=0.2). This suggests that diarrhoea was not significantly more likely in equids that received flunixin meglumine in the previous week than in those that did not. Abdominal pain plays a confounding role and the perceived increased risk of diarrhoea by clinicians with administration of flunixin meglumine in many cases is secondary to administration of these drugs to patients that might have an unhealthy gastrointestinal tract so intrinsically predisposed to develop diarrhoea. Also equids with diarrhoea were 0.1 times as likely to have received ketoprofen. An explanation of the low odds ratio of ketoprofen administration in relation to diarrhoea may lay in the fact that in North America ketoprofen is used almost exclusively to treat equids examined for orthopaedic disease as shown in Chapter 4. In the *post-hoc* analysis the effect of ketoprofen administration became non-significant (p=0.07) when colic and orthopaedic disease were added to the conditional logistic regression model suggesting perhaps the influence that orthopaedic disease had on the effect of ketoprofen.

Since antimicrobials have been well documented to induce diarrhoea in equids, and since they are often used in combination with NSAIDs in equine practice, it seemed appropriate to include antimicrobials in the model and to investigate a possible interaction with NSAIDs. The only significant interaction term was between phenylbutazone and antimicrobials and the odds ratio for their concurrent administration was higher in equids with diarrhoea. The odds ratio for administration of both drugs in cases with diarrhoea was lower than when antimicrobials where considered on their own and this finding might result from the low odds for diarrhoea along with phenylbutazone administration.

It appears that diarrhoea is 0.3 times as likely with administration of corticosteroids. Initially it was hypothesised that orthopaedic cases receiving intra-articular corticosteroids could have explained this finding: however, inclusion of the orthopaedic variables did not change the significance of corticosteroid treatment in the model and only induced a slight increase in the odds ratio. Corticosteroid administration, both systemically or intra-articular, is not a well-recognised cause of diarrhoea at label doses in equids. In fact dexamethasone treatment is often warranted in cases of diarrhoea associated with cyathostomiasis as well as infiltrative bowel disease (Barr, 2006) to ease the inflammatory response associated with these conditions.

Other variables such as laxatives and intravenous fluids were included only when administered up to 2 days before the development of diarrhoea. The 2-day period for intravenous fluid and laxative administration was chosen because both treatments would be expected to reduce faecal consistency within 48 hours of administration. Further, both treatments are usually administered directly by the veterinary surgeon on a single administration and therefore the date of data entry in PMSS likely reflects the actual date of administration. This differs from the other explanatory variables, which include drugs often dispensed for administration by the owner over multiple days, hence evaluating a period of 1 to 2 weeks from the data being entered in the PMSS. Diarrhoea was significantly more likely with administration of these drugs, however when colic was included in the model, the use of laxative became non-significant ($p=0.4$) while the odds for diarrhoea with use of intravenous fluids were only marginally decreased.

Anthelmintic administration was 0.6 times as likely in the 14 days preceding the development of diarrhoea. This did not change significantly in the *post-hoc* analysis.

The time elapsed between the date of data entry in the PMSS, corresponding to the drugs being dispensed, and the actual length of treatment was summarily accounted for by the use of the 7 or 14 days period. The explanatory variables evaluated in this study include drugs administered or dispensed 7 to 14 days prior to the development of diarrhoea. The use of either the 7 or 14 days cut-off was determined on the basis of clinical relevance and AIC and significance in the model. Also, as a significant proportion of the data was derived from first opinion ambulatory practice, there was no guarantee that owners completed the course of administration or followed precisely the instructions regarding dose and dose interval given by their veterinarian. Information on dosage, or total quantity of drug used, was not directly available in the data and extraction of this information would have required knowledge of the weight of every animal to be available to calculate the dosage used. In first opinion equine ambulatory settings a scale to determine the precise weight of a patient is not available and therefore weight is generally obtained by rough estimation. Extraction of dosages and length of course of administration recommended is hard to achieve from the free-text EMR data and was not performed in this study. This is a major shortcoming of the study, as including dosages and length of course of treatment prior to the development of diarrhoea could substantially improve the model fit as well as the clinical relevance of the results. This could be achieved only by systematically and prospectively including this information in the EMR. These are the main reasons as to why all variables were binary (“true” or “false”) and no continuous variable was created to document the number of days between drug administration/prescription and development of diarrhoea or the total amount of drug used before the development of diarrhoea.

The *post-hoc* analysis was performed to investigate some unexpected findings produced by the initial model accounting for the effect of confounding factors often associated with administration of NSAIDs, such

as colic and orthopaedic disease. These included the low odds ratios for phenylbutazone and ketoprofen and an odds ratio for flunixin lower than expected from clinical experience. Including comorbidities in the model complicated the analytic process, but at the same time the large population of the study allowed this process without a significant drop in study power.

Logistic regression was used as it provides information on the association between binary explanatory and binary outcome variables (Thrusfield, 2007, Dohoo et al., 2010). Evaluating differences between single and multiple logistic regression analysis allows understanding of how variables influence each others' effect over the outcome. Akaike Information Criterion (AIC) has been used to select the linear regression model best fitting the data in multiple conditional logistic regression analysis. The AIC provides an index to compare the quality of each model, by estimating the amount of information lost by each model (Akaike, 1998; Akaike, 1974). The best fitting model was selected manually trying different combinations of explanatory variables and interaction terms.

In conclusion, it appears from the analysis of the data that, while flunixin meglumine and ketoprofen might increase the risk for diarrhoea in a statistically significant manner, this increase is actually non-significant once episodes of abdominal pain are accounted for. The data is obtained from a real population where veterinary clinicians are usually aware of the risk of side effects related to the use of this drug and therefore judiciously observe label doses. However, further evidence on whether veterinary surgeons truly observe recommended dose regimens is needed.

The role of phenylbutazone remains puzzling as this drug, like flunixin meglumine and ketoprofen, also affect COX-1 activity. However, from the results of this analysis it appears that phenylbutazone administration is less likely to be associated with diarrhoea. However, other not evaluated

confounding factors might be relevant and the *ex vivo* data should warrant a judicious use of these drugs. Firocoxib on the other hand appeared safer *ex vivo* as well as from the results of this study. However, this drug is licenced for treatment of pain from osteoarthritis in horses and whether diarrhoea would occur more frequently if this drug was administered to patients with underlying gastrointestinal disease remains undetermined.

5.4 Discussion

This chapter has demonstrated some of the applications and analysis possible with the semi-structured free-text datasets assembled as described in Chapter 3 and analysed with the methods illustrated in Chapter 4.

Interpretation of the results of the analysis in this chapter required deep understanding of how data was obtained and of veterinary practices in each continent. While the analysis of British and North American data provided different results, analyses of each dataset share overall similar conclusions. For both datasets the prevalence of reported NSAID toxicity is extremely low despite these drugs being commonly used in veterinary practice. This is likely a consequence of several factors including the difficulty in detecting low levels of toxicity, particularly in first opinion equine practice, as this is highly dependent on owners' ability to recognise, or willingness to report, mild clinical signs. Also the difficulty in confirming the diagnosis *ante-mortem* poses a huge challenge particularly in first opinion ambulatory settings, which might result in a considerable proportion of the cases being unrecognised.

In this study a total of 27 cases of suspected NSAID toxicity were identified, out of a total of 453,695 animals. It is important to point out that the clinical data was collected over a 25-year period and that the analysis was strictly retrospective. Also the extremely low prevalence of

toxicity likely could reflect the judicious usage of these drugs by veterinary surgeons, who are aware of the risks associated with their administration and therefore adhere to the recommended dose and length of treatment. However, the present study did not evaluate the dosage used and no evidence that clinicians adhered to labelled doses is available. Despite all these limitations related to the retrospective nature of the study, including lack of data on dosages and length of treatment used, the prevalence of toxicity appears to be extremely low. This finding suggests that NSAIDs are safe to use, provided one avoids overdosing and monitors closely for side effects in order to discontinue administration before toxicity becomes severe and life threatening.

The findings of logistic regression analysis highlight that diarrhoea is slightly more likely with NSAID administration, but as the prevalence of diarrhoea is low (1-2%) while prevalence of NSAID usage is high the slight increase in risk will have a minimal impact on the prevalence at the population level. Further, the search extracted records including a wide range of severity of this aspecific clinical sign, while the prevalence of extreme toxicity resulting in diarrhoea, such as life-threatening right dorsal colitis is extremely low.

Several differences are present between the results of the multiple logistic regression analysis of the data from the United Kingdom and North America. These results should be interpreted in light of several differences in equine veterinary practice between the two continents and confirms how one should translate with caution to his/her own country studies obtained from different geographical regions. This is to account not only differences in legislation for drug usage, but also economic and social differences, which could affect not only how and when animals receive treatment but also how these animals are managed, including availability of pastures, distances travelled for competitions, etc. Further differences in climate and day-light hours could also affect findings

between countries or at different latitudes. In conclusion, this study highlights how findings from one country should be applied with caution to other countries.

One of the main differences between countries is the availability of certain drugs. For example, metamizole is available in Canada but not in the United States, while in the United Kingdom it is available only in combination with a spasmolytic licensed for the treatment of visceral pain. This is a rather relevant difference as it may have a significant repercussion on how veterinarians manage visceral pain in these countries. As highlighted in Chapter 4, the use of the metamizole/butylscopolamine combination is very popular for the management of colic in the United Kingdom, while in North America flunixin meglumine is more widely used for this condition. While flunixin is also commonly used with colic, proportionally a large amount of colic cases also received phenylbutazone in the United Kingdom. It is possible that veterinary surgeons in the United Kingdom tend to select metamizole in cases that might be slightly more likely to then develop diarrhoea within the next 7 days (hence a markedly increased OR for previous metamizole administration in cases with diarrhoea). On the other hand in the US the vast majority of colic cases receive flunixin meglumine, which might then reflect in the increased odds ratio of previous flunixin meglumine administration in cases with diarrhoea in that country. While metamizole is available in Canada as a single drug, not many vets decided to use it for the treatment of colic, despite the drug being licensed for the treatment of this condition in this country, therefore little conclusion could be drawn from the use of metamizole on its own. Also the widespread use of phenylbutazone to manage visceral pain in the United Kingdom might have played a role in affecting the difference in flunixin meglumine and phenylbutazone's odds ratios in the multivariable analysis. If a larger proportion of cases with gastrointestinal pain, which might therefore be at increased risk for diarrhoea development received phenylbutazone, then a positive

relationship between diarrhoea and phenylbutazone might be detected. At the same time, as similar cases received flunixin meglumine in North America, the same positive relationship exists between flunixin meglumine and diarrhoea. The *post-hoc* analysis was performed to account for this "drug selection bias", and the results show a marked decrease in odds ratio for phenylbutazone usage in cases with diarrhoea once the analysis included the colic variable. The relationship between diarrhoea and flunixin meglumine became insignificant once colic was accounted for in North America and it remained non-significant in the United Kingdom. Also all instances where veterinary surgeons combined multiple NSAIDs were not significant in the analysis from both regions.

The effect of firocoxib was also similar between United Kingdom and North America, but only one case receiving firocoxib was present in the dataset for analysis from the United Kingdom, so the data from this country was underpowered to assess the relationship between firocoxib and diarrhoea. In North America there was no significant relationship between diarrhoea development and firocoxib administration.

Moreover it is also possible that either, other unforeseen conditions played a significant role, or that colic as a whole is too general and including only certain colic cases, such as those with more severe gastrointestinal disease requiring referral or emergency exploratory laparotomy, might change the significance of some variables in the analysis. Ultimately it is also possible that these drugs might truly increase the likelihood of diarrhoea and therefore the findings of this study are correct.

Other differences between United Kingdom and North America might exist on the frequency with which veterinarians prescribe NSAIDs to their patients as discussed in Chapter 4. For example, in North America the prevalence of use of NSAIDs was higher than in the United Kingdom. This data suggests that veterinarians in North America use these drugs more

frequently than veterinary surgeons in the United Kingdom. The reason for this behaviour is unclear, but this behaviour might be reflective of what these clinicians are taught during the undergraduate years and at CPD events in their respective country. For example, it could be that in the United Kingdom more emphasis is placed on the severity of side effects from NSAID usage or that more emphasis is placed in North America on ensuring that pain and inflammation are managed appropriately. Alternatively, British clients might be less willing to spend money to treat their patients with NSAIDs, or alternatively prefer to have a condition investigated and treated, rather than simply managing signs of pain. Explaining this difference was not the aim of the study, but highlighting a difference was a first step. Further studies are required to evaluate this difference to understand how research findings from one continent apply to the other.

Nevertheless this difference in prescribing habit might also have influenced the result. This might be particularly true if in the United Kingdom NSAIDs are used most commonly in cases that have more severe inflammation, so are more at risk of side effects. However, the overall prevalence of diarrhoea was very similar between datasets so this effect remains undetermined.

A further, significant difference between United Kingdom and North America data is the type of cases included. While the British dataset included only first opinion cases, data from North America included a mixed population of first opinion and referral cases. Referral work includes generally first opinion cases, which are too complicated to be dealt with in ambulatory settings or that require advanced diagnostics and intensive care treatment. A proportion of the referral cases is also a sicker population and could therefore be more susceptible to develop drug toxicity, which might result in diarrhoea.

Other differences between the United Kingdom and North America are reflected in the role of corticosteroids. In the United Kingdom equids with diarrhoea were nearly twice as likely to have received corticosteroids in the preceding 7 days than controls, while in North America equids with diarrhoea were only 0.3 times as likely to have received corticosteroids than controls in the preceding 14 days. The current study did not aim at describing the prescription habits of corticosteroids and any difference between countries in usage of these drugs remains undocumented. However, a marked difference exists in NSAID usage in orthopaedic patients between United Kingdom and North America. Table 4.8 highlights a much greater proportion of orthopaedic examinations in North America documenting usage of NSAIDs than in the United Kingdom. A NSAID was used in ~35% of orthopaedic examinations/treatments in North America, while this was documented only for ~13% of orthopaedic cases in the United Kingdom. This difference might partially reflect inclusion of referral cases in the dataset from North America as well as different NSAID prescription habits between countries. This difference in drug usage suggests a difference in management of orthopaedic cases between countries and could well suggest a difference of attitude towards the use of intra-articular corticosteroids for orthopaedic disease, such as osteoarthritis. Data from the studies in this chapter also show that in the United Kingdom the use of corticosteroids is nearly 4 times smaller than in North America. Although the specific use of corticosteroids was not one of the aims of the study and it was not investigated further, the data supports the theory of a different attitude towards corticosteroids usage between countries.

The effect of intravenous fluid therapy was similar between the two continents. It appears that equids with diarrhoea were more likely to have received intravenous fluids than control animals. This might reflect the habit of providing more fluids than the animal's actual needs in equine practice, which might result in decreased faecal consistency. On the other

hand, in cases of large colonic impactions the aim of fluid therapy is to increase colonic secretions to soften the impacted faecal material in the colon, therefore cases of large colon impaction might have influenced this result. Administration of laxatives via the oral route is the currently recommended treatment for large colon impactions (Sanchez, 2010). Laxatives also yielded different results between continents. While no significant effect of laxatives over diarrhoea was detected in the multivariable regression analysis in the United Kingdom, previous laxative treatment was significantly more likely in equids with diarrhoea in North America. The reason for this difference also remains undetermined. This finding is puzzling, particularly as laxative administration was 7 times more likely in equids with diarrhoea in the univariable analysis in the United Kingdom. The relationship between diarrhoea and large colon impaction was not investigated, as this was not within the scope of the study.

A further difference between United Kingdom and North America lies in the effect of anthelmintic drugs on the output of the analysis. The recorded use of anthelmintic drugs was two times greater in North America than in the United Kingdom. The interpretation of this finding is somewhat difficult. Possible explanations include that either in North America there is a greater tendency to blanket-treat herds while in the United Kingdom monitoring faecal egg counts to identify animals with higher counts that might require treatment may be more common. However, the American Association of Equine Practitioners (AAEP) has released and recently updated guidelines for parasite control, which suggest that deworming should be focused on certain animals (so called “high-shedders”) rather than treating the whole herd (Nielsen et al., 2013). This is because in the past 40 years the population of equine intestinal parasites has shifted from the large strongyles being the most significant to the small strongyles, the cyathostomes, as the most significant clinically. Recent evidence suggests that cyathostome

populations are developing anthelmintic resistance and the best results are obtained by trying to control and not eliminate these parasites (Love, 2003). These guidelines might have in some way affected attitude towards blanket worming in North America as well as in the United Kingdom. Analysis of frequency data shows that while the prevalence of deworming is substantially unchanged in the United Kingdom since 2008, anthelmintic usage in North America has seen a significant and steady decrease in the past 5 years. Despite this attempt to reduce the indiscriminate usage of anthelmintic drugs, that might predispose to development of resistance, the fact that dewormers are available over the counter in North America while in the United Kingdom they can be purchased only under the direction of a suitably qualified person needs to be kept in consideration. This suggests that an unknown proportion of anthelmintic drugs is administered directly by owners without any veterinary supervision. Further lay social attitudes towards resorting veterinary advice for deworming might also be different between countries. Therefore the effect of this variable is difficult, if not impossible, to interpret from the data. However, anthelmintic drug usage was still included in the analysis as it was important to account for at least a portion of the anthelmintic drugs used and unrecorded anthelmintic drug usage was expected to be uniform throughout the study population.

The modelling approach adopted for both studies from the United Kingdom and North America presented several limitations. The variables included are simple and generic and not specific so may be difficult to relate the results to the specific practical clinical scenario. Further, residual confounding caused by unmeasured or imperfectly measured confounders is inherent to this type of analysis. Alternative approaches to a case-control design could include the complete dataset but would create further problems such as dealing with large amounts of missing data.

In conclusion, the findings of these studies suggest that the prevalence of reported NSAID toxicity is very low. Also diarrhoea, used as a marker for

NSAID toxicity, is slightly more likely following administration of NSAIDs, even when the concurrent administration of other substances, well documented in causing diarrhoea, is accounted for. However the prevalence of reported diarrhoea was very low in our population and this risk is minimal; clinical evidence and intuition would suggest this provided that recommendations for NSAIDs dosage and frequency of administration are respected.

CHAPTER 6 – Discussion And Conclusions

6.1 – Novelty of this work

The two goals of this PhD were to evaluate the biochemical potential for side effects of NSAIDs with different COX selectivity using an *ex vivo* model of clinical patients and also to evaluate the prevalence and clinical relevance of these side effects in a horse population.

The first goal was achieved using methodologies already validated in the literature (Beretta et al., 2005). The novelty of this work consisted of using samples from actual clinical patients and not experimental animals. This approach is relevant as it fills a gap in knowledge since, thus far, knowledge of COX-inhibition of NSAIDs has worked under the assumption that little difference would be present between experimental animals and actual clinical patients. While this assumption is understandable, scientific rigor requires that all assumptions should be examined and supported by evidence, hence the need for this study. The findings in this study on clinical patients clearly support the findings of previous studies performed on experimental animals (Barton et al., 2014; Cook et al., 2009a; Marshall, 2010). The biochemical investigations of Chapter 2 confirm the potentially deleterious effects of non-selective COX inhibitors such as flunixin meglumine and phenylbutazone and that using COX-2 selective drugs, such as firocoxib, spares COX-1 activity in clinical patients. While these findings indicate that firocoxib has a safer profile compared to flunixin meglumine and phenylbutazone, the impact on the horse population remained undetermined. The data from any biochemical investigation merely described the potential for toxicity from the use of NSAIDs, but does not provide any information as to what proportion of animals in the population actually experiences side effects in clinical practice and does not evaluate the confounding effect of other concurrent medications or comorbidities.

The second goal of this work was therefore to evaluate the impact that the side effects from the use of NSAIDs have on a clinical population of equids. The methodology implemented to achieve this goal was novel and opens a wide range of opportunities for future research studies, which go beyond the scope of this thesis. Electronic medical records from veterinary practice offer a goldmine of data for clinical epidemiologic research. The methods in Chapter 3 describe two different techniques that can be used to obtain retrospective data from the EMRs of equine veterinary practices. The advantages and disadvantages of each technique are discussed in detail and show how each technique can adapt to different circumstances. The technique applied to the United Kingdom is more labour intensive, collects data of variable quality that might require significant effort in data cleaning, but offers the advantage of being relatively cheap. On the other hand, the technique applied in North America quickly provided good quality data through the PMSS company, but incurred a significant financial expense. Equally, both techniques provided a great amount of data suitable for the scopes of the study.

Another novel aspect of the thesis is the text mining methodology validation in Chapter 3. Although the use of text mining techniques of EMRs have been reported before in the veterinary literature (Anholt et al., 2014a; Cameron et al., 2014; Lam et al., 2007a), their sensitivity and specificity has been shown to be good but not excellent and the methodology used for their validation has been suboptimal (Anholt et al., 2014a). Chapter 3 describes how these techniques have been improved to achieve excellent agreement with manual classification to justify their wider use in the future. The methodology described highlights the importance of creating exhaustive dictionaries to achieve optimal accuracy of the mining process. The use of exclusion dictionaries and re-inclusion dictionaries was novel and allows minimisation of false positive and false negative rates. This method is mostly automated with a minor

manual component necessary for dictionary definition. Certainly the improvement in sensitivity and specificity outweighs the time and effort required for the manual component of the analysis as it increases confidence in the results significantly.

Chapter 4 and Chapter 5 are also innovative as they describe for the first time the prevalence of use of several drugs gathered from two large equine clinical populations, describing in detail the differences that occur in drug usage between the United Kingdom and North America. The prevalence of conditions such as diarrhoea, colic and orthopaedic disease was relatively similar between these regions while the pharmacological management of these conditions was different as highlighted in Chapter 4. These findings highlight the general importance of interpreting research findings in light of their geographical and legislative background. Clinical studies from one country should be interpreted with caution in relation to other countries.

Finally, this work concludes with the investigation of the relationship between clinical signs consistent with NSAID toxicity, drugs administration and comorbidities. The use of such a large population provided sufficient statistical power to include several variables in the logistic regression analysis. Determination of odds ratio for NSAID administration in cases of diarrhoea provided an indication of the extent of the impact that the use of these drugs may have on these equine populations.

6.2 Overall conclusions

The overall conclusions of this Thesis are focused on whether NSAID administration is associated with significant side effect on equids and on the extent of this effect. While the findings in Chapter 2 show that there is a valid pharmacological basis to be concerned over NSAID toxicity, the epidemiological investigation highlights that the impact of these side

effects on the population is small. This conclusion needs to be interpreted in light of the fact that equine veterinary surgeons are generally well aware of the potential side effects of NSAIDs and therefore rarely exceed recommended dosages. However, data on dosages used in the datasets was not available and this is only a supposition based on the author's personal experience. Studies including data on dosages should be highly encouraged to highlight the importance of following the legislation that regulates the use of these drugs and to promote alternative means of managing pain in equids where NSAIDs alone are not sufficient. On the other hand, from a population point of view, the impact of these NSAID-related side effects is so limited that one might conclude that NSAIDs, including non-selective COX inhibitors, are generally quite safe to use. It remains undetermined whether an association exists between NSAID dosage and course length and the odds for diarrhoea development. As previously discussed, data on dosage has not been extracted from the free-text EMR. It remains unclear to what extent NSAIDs are used at dosages exceeding recommended dosages. Similarly, under-dosing could potentially have had an effect as under-dosing would be expected to result in reduced toxicity. These findings also challenge the need to spend significant financial resources to fund research aiming at identifying NSAIDs with a safer profile.

6.3 Limitations of this work

The conclusion that can be drawn from the results presented in this Thesis should be interpreted in light of their greater limitations.

The biochemical investigation of COX activity was performed in clinical patients, undergoing procedures of variable clinical invasiveness that could have triggered an inflammatory response of variable degrees. Two main intrinsic limitations were bound to the clinical study design. The first one was that allocation of horses to either flunixin meglumine,

phenylbutazone or fircoxib group was not randomised or within the control of the investigator. Group allocation was determined by the attending clinician and could have depended on presenting clinical signs, diagnosis or degree of inflammation present. A non-randomised group allocation meant that group allocation may have not been independent from the outcome being measured. So horses with more severe inflammation might have been more likely to receive a more potent COX inhibitor such as flunixin meglumine. This might have ultimately affected the results, as other NSAIDs could have been less effective at reducing COX activity in horses suffering a more severe inflammatory response. The second limitation deriving from the study design and non-randomised group allocation was the lack of standardisation in term of surgical procedures performed. Different diagnoses also required treatment with different surgical procedures, which could have triggered inflammatory responses of variable severity. However, previous studies have shown that the inflammatory response following surgical trauma was minimal in the first 24 hours and the inflammatory response was similar between minimally invasive and very invasive surgical procedures (Jacobsen et al., 2009). A further limitation of the study in Chapter 2 included sample size, which was determined based on the predicted difference in the drugs COX selectivity. Involving more horses might allow detection of other significant relationships between drug and metabolite concentrations and enzyme activity, disease states and age or breed differences and warrants future investigation. This would have been too costly and could not be performed.

The epidemiological investigation in Chapters 4 and 5 also suffered a few limitations, which were mostly intrinsic to the retrospective nature of the data. Missing data could have affected the outcome of our analysis. Information about age and gender was not available for a significant proportion of the study population and these variables could have been used as matching criteria or added to the model as possible confounders.

Large amount of missing data could indicate that the data available is also of poor quality and may not reflect what is actually happening (Coleman et al., 2015) and future studies should aim to validate that what is in the records reflects what actually happened. Data was also missing from some veterinary practices for some of the early years in the database. Older data from the United Kingdom was available only from one practice while older data from North America included proportionally a very small amount of patients. How this could have biased the results of the study remains also unclear. Finally, data on patients migrated to another practice, or deceased, but not recorded in the system was also missing. This is reflection of the dynamic nature of veterinary practice, where equine patients might change practice as they are sold, momentarily or definitively relocated or if the owner decides to change veterinary care provider. The data used for this study has no information on animal movement and loss of information could be prevented in future prospective studies only if the vast majority of veterinary practices in one country contributing data using PMSS that permits tracking of patients between practices. Further, unreliable data, such as that with wrong dates of data entry limits the use of data migrated from older PMSS to the current and may result in loss of data.

From a clinical standpoint it would have been very useful to have data regarding dosages to be able to evaluate any effect of dose on toxicity. The current analysis is performed under the assumption that overdosing of NSAIDs is a rather rare occurrence in veterinary practice, but the extent to which NSAID are used at inappropriate dosages remains currently undetermined. In ambulatory first opinion equine practice particularly, patient weight is often estimated and not directly measured and has been shown to affect the accuracy with which dosages are calculated (Ross et al., 2015). It is possible that in some cases some animals were administered amounts of certain drugs that were outside the recommended dose ranges. Also information on administration route,

course duration and clear definition of the indication for using the drug was also lacking. In a prospective study this information may be specifically requested to increase the precision of the information collected and limit the amount of missing data.

Another limitation is the assumption that owners administered drugs as instructed by the prescribing veterinary surgeons. Monitoring owners' compliance would be near to impossible even with a prospective study design. The data available for this study did not account for the difference between date of drug dispensation and actual date of drug administration and all analyses were done under the assumption that once a drug is dispensed administration would be on the same day. While this may be generally true for some drugs (e.g. antimicrobials, intravenous fluids, laxatives), which are generally prescribed to treat acute onset conditions requiring immediate care, this may not necessarily be the case for other drug categories, which may be dispensed to treat more recurrent disease, such as NSAIDs to manage chronic lameness or corticosteroids to use during exacerbation of a recurrent disease, or to fit a scheduled protocol, such as deworming. The effect that time elapsed between drug prescription and administration may have had on the overall results is unclear. In the future, collecting data where date of drug administration is recorded will be possible, but probably only for studies on hospitalised animals, since recording actual date of administration by owners is hardly feasible.

Both dataset from United Kingdom and North America were obtained from a convenience sample of veterinary practices. This could have intrinsically introduced some bias. Whether the population included in the study was truly reflective of the overall veterinary population remains undetermined. Whether these veterinary practices worked at a higher standard than the general population or whether they had some purchasing deals with certain companies that could have biased towards

certain NSAIDs remains unclear. Ideally in future studies practices should be selected at random from the veterinary practice population.

A final significant limitation of this study results from our ability to differentiate the type of comorbidities included. For example, including only certain types of colic or orthopaedic conditions might have affected the results significantly. For example, diarrhoea is considered generally a condition generated from the hindgut, or cases with endotoxaemia might be more prone to diarrhoea development. Similarly, some orthopaedic conditions require long term management that could result in an overall greater amount of NSAIDs administered which could result in a higher risk of toxicity. Therefore including more specific variables in place of the generic colic and orthopaedic terms could have affected the results significantly. This was not performed largely due to time constraints, but would be one of the logical next steps in this particular area of research.

Performing these studies in a prospective manner would incur significant cost and time, necessary to develop a tool to allow communication between PMSS and obtain uniform data from different PMSS. Further, years would then be required so that enough data could be collected to perform some meaningful analysis. However, this process would provide several advantages, including data of better quality, minimising missing data and would hopefully include a sample of veterinary practices including a more representative sample of the equine population under veterinary care.

6.4 Future work

The work of this Thesis has laid the basis for several future studies.

The biochemical investigation in Chapter 2 could include other drugs such as meloxicam and ketoprofen. Drug selection was not under the

investigator's control and the clinicians responsible for these clinical cases did not use these two drugs to manage perioperative inflammation and pain of their patients. Further work could also aim at comparing the effect of these drugs in ischemia-reperfusion injury as done by Cook and colleagues (2009a). Flunixin meglumine has been shown to induce mucosal neutrophil infiltration after ischaemia-reperfusion insult and this might induce delayed return of normal peristalsis in some cases (Cook et al. 2009b). Other non-selective COX inhibitors might have a lesser effect on neutrophil migration and could improve overall survival in these cases (Cook et al., 2009a).

The epidemiological investigation started by creating a dataset of EMRs, which includes all the information stored in the PMSS, therefore the data can be used to evaluate the prevalence of any disease or usage of any drug. The relationship between administration of any drug and disease development could be investigated with the same methodology of Chapter 5. Further the data could also be used to evaluate time to recovery after certain procedures or with a particular disease. Moreover investigation of how disease prevalence evolves overtime or varies geographically could also be investigated. The data offers the advantage of a very large population, which could be used to calculate a reliable prevalence even for very rare conditions, impossible with smaller datasets.

A further development of this work would include prospective collection of data, from practices willing to collaborate. Data obtained prospectively for specific research purposes would offer the advantage of less missing information, for example regarding dosages used. Prospective data collection should be implemented by working with PMSS companies to improve the compatibility of their systems with the scope of epidemiological research, without altering the user interface of their software. A key component of a prospective study of this type would be developing software to support veterinary surgeons' compliance. While

complex coding systems offer the advantage to provide a clear classification of the cases in a dataset (O'Neill et al., 2012a), they may result in poorer compliance by veterinary staff in the long run. To ensure the best compliance, complete data should be obtainable directly from the PMSS systems without requiring an effort by veterinary staff that goes much beyond the general record keeping tasks of everyday practice.
Prospective data, as highlighted by studies in small animal species (Jones et al., 2014; O'Neill et al., 2012a; Radford et al., 2011), can provide useful information in disease surveillance and alert veterinary surgeons of disease outbreaks in the their area. Studies of this type could have a great impact on equine welfare.

In conclusion, much more can be done to build on the work in this Thesis, both on the topic of NSAID toxicity but also by using the data for many other varied studies.

Appendices

Appendix 1.11

Table summary of products containing NSAIDs

Summary of products containing NSAIDs licensed for use in horses in the United Kingdom, United States of America and Canada.

Drug	Brand name	Form	Route	Available conc.	MLD	Indications	Country	YFM
Phenylbutazone	Butagran Equi	Pwd	PO	200mg/1g	4.4mg/kg BID	MSK	UK	1994
	Pro-Dynam							1994
	Equipalazone							1994
		Pst		1g				1994
		Inj	IV	200mg/ml		MSK, AP		2013
	Butazolidin	B	PO	1g	4g/day	MSK	US	
		Tab						
		Grn						
		Inj	IV	200mg/ml				
	Butatron	Tab	PO	1g				
		B	PO	1g				
	Tevcodyne	B	PO	1g				
		Inj	IV	200mg/ml				
	EquiBute	Tab	PO	1g				
		Inj	IV	200mg/ml				
	Butasone 1000	Bolus	PO	1g			CA	1999
	Butasone 400	Pwd	PO	1g				1989
	Butasone Conc	Pwd	PO	1g				1989
	Butequine	Pst	PO					2014
	Buzone conc	Pwd	PO	1g				1997
	Buzone Inj	Inj	IV	200mg/ml				2012
	Phneylbutazone 20%	Inj	IV	200mg/ml				2010
	Phenylbutazone concentrate	Pwd	PO					2005
	Phenylbutazone Inj	Inj	IV	200mg/ml				1974
	Phenylbutazone powder	Pwd	PO	1g				1993
	Phenylbutazone tab	Tab	PO	1g				1974
Suxibuzone	Danilon Equidos	Grn	PO	1.5g	6.26mg/kg/day	MSK	UK	2001
Flunixin Meglumine	Allevinix	Inj	IV	50mg/ml	1mg/kg	MSK, Colic	UK	2013
	Cronixin							1996
	Flunixin							1998
	Meflosyl							1998
	Norixin							1997
	Pyroflam							2006
	Finadyne							1987
	Finadyne Paste	Pst	PO			MSK		1989
	Equinixin	Gr	PO	25mg/g				2006

	Banamine-S	Inj	IV, IM	50mg	0.5mg/lb/day	MSK, Colic	US	
	Banamine Inj							
	Flu-nix							
	Flunixin Meglumine Inj							
	Flunixin Meglumine Sol							
	Flunixin Inj							
	Banamine Granules	Grn	PO			MSK		
	Banamine Paste	Pst						
	Banamine	Inj	IV, IM	50mg/ml			CA	1979
	Cronyxin							1997
	Flunazine							2001
	Flunixin inj							1997
	Influx-50							1999
	Suppressor							2004
Metamizole	Buscopan Compositum Inj	Inj	IV		5ml/100kg	Dx, Colic, UO	UK	2001
	Dipyrone Inj		IV, IM, SC	500mg/ml			CA	1965
	Dipyrone 50		IV, IM					1997
Meloxicam	Animexolan	Inj	IV	20mg/ml	0.6mg/kg SID	MSK, Colic	UK	2012
	Contacera	Inj		15mg/ml				2014
		Inj		20mg/ml				2012
	Emdocam	Inj		20mg/ml				2011
	Inflacam	Inj		15mg/ml				2011
		Inj		20mg/ml				2011
		Grn	PO	330mg				2011
	Loxicom	Inj	IV	20mg/ml				2009
		Pst	PO	50mg/g				2009
	Melosolute	Inj	IV	40mg/ml				2013
	Melovem			20mg/ml				2009
	Meloxidolor			20mg/ml				2013
				40mg/ml				2013
	Meloxidyl			20mg/ml				2007
	Metacam	OS	PO	15mg/ml				2003
		Inj	IV	20mg/ml				2001
				40mg/ml				2015
	Novaquin	OS	PO	15mg/ml				2015
	Recocam	Inj	IV	20mg/ml				2011
	Rheumocam	OS	PO	15mg/ml				2008
	Recocam	Inj	IV	20mg/ml				2011
Ketoprofen	Dinalgen	Inj	IV	150mg/ml	2.2mg/kg SID up to 3 days	MSK, Colic, P-O	UK	2010
	Ketink			100mg/ml	2.2mg/kg SID up to 5 days	MSK, Colic, P-O		2012
	Ketodolor							2013
	Nefotek							2012
	Rifen					MSK, Colic P-O, AP		2010
	Comforion Vet					MSK, Colic		2005
	Ketofen							1992

	Kelaprofen							2012
	Ketofen				1mg/lb SID up to 5 days	MSK	US	
	Anafen		IV, IM				CA	1993
	Ketoprofen V		IV, IM					2016
Diclofenac	Surpass	Crm	Topical	10mg/1g		OA	US	
Meclofenamic acid	Arquel Granules	Grn	PO	1mg/lb	1g/1000lbs SID up to 7 days	MSK		
Firocoxib	Equioxx	Inj	IV	20mg/ml	0.09mg/kg SID up to 14 days	OA	UK	2008
		Pst	PO	8.2mg/g	0.1mg/kg SID up to 14 days			2008
		Inj	IV	20mg/ml	0.09mg/kg SID up to 5 days	OA	US	2008
		Pst	PO	8.2mg/g	0.1mg/kg SID up to 14 days, or 9 days after IV Equioxx			2008
Aspirin	Acetylsalicylic acid bolus	B	PO	15.6g			CA	1996
Vedaprofen	Quadrisol	Gel	PO	100mg/ml		MSK, P-O	UK	1997
Deracoxib	No product currently licensed for use in equids							
Eltenac	No product currently licensed for use in equids							
Carprofen	No product currently licensed for use in equids							

MLD: maximum licensed dose; YFM: Year First Marketed; Pwd: powder; Pst: Paste; Inj: injectable; B: bolus; Tab: tablet; Grn: granules; OS: oral suspension; Crm: cream; PO: orally; IV: intravenous; IM: intramuscular; SC: sub-cutaneous; BID: twice daily; SID: once daily; MSK: muscleskeletal disorders; AP: antipyretic; Dx: as a diagnostic aid; UO: Urinary obstruction; P-O: peri-operatively; OA: osteoarthritis.

Appendix 3.1 – Letter for Veterinary Practices

The clinical use and adverse effects of NSAIDs in the UK horse population

Marco Duz MedVet MVM(Res) MRCVS
Tim Parkin BSc BVSc PhD DipECVPH FHEA MRCVS
John Marshall BVMS DipACVS/ECVS PhD MRCVS

Non-steroidal anti-inflammatory drugs (NSAIDs) are among the most widely used medications in both human and veterinary medicine. Phenylbutazone and flunixin meglumine have been associated with adverse effects in horses including gastric ulceration, right dorsal colitis and renal failure. Recently, NSAIDs designed to reduce adverse effects have been introduced to the veterinary market, including meloxicam and firocoxib, but their level of use in the UK is currently unknown.

AIMS: Describe the use of NSAIDs in the UK horse population. Identify the true prevalence of NSAIDs induced toxicity and whether significant differences between NSAIDs exist.

METHODS: We need to access the records of horses under your care over the years through the practice management software provider or locally at your practice. All data collected will be handled as anonymously as possible and we will provide a signed confidentiality agreement form for you. Records will be analysed with a content analysis and text mining software to automatically extract the data of interest to be included in the statistical analysis.

WHAT ARE YOU REQUIRED TO DO? Only sign the agreement form. Collection of digitally stored data will be done on-line (through an agreement with the management software company) or alternatively at your practice. You can withdraw from the study at any time.

WHAT'S IN IT FOR YOU? You have the opportunity to participate in a large-scale epidemiological study. The results may guide your future choice of the best NSAID/dosage for your equine patient.

If you are willing to participate, or simply want more information, please contact John Marshall or Marco Duz at the Weipers Centre (0141 330 5999 – equine@vet.gla.ac.uk).

Best regards, Marco, Tim and John

WEIPERS CENTRE EQUINE HOSPITAL
Division of Companion Animal Sciences
Faculty of Veterinary Medicine
University of Glasgow
Bearsden Road, Glasgow, G61 1QH
Telephone: 0141-330 5999 *Fax:* 0141-330 6025 *Email:* equine@vet.gla.ac.uk
University of Glasgow Charity Number: SC004401

Consent for template for participating practices

(Please write this letter on your practice stationary if possible - with letterhead/ name of practice)

To:
Tim Parkin BSc, BVSc, PhD, DipECVPH, MRCVS
Boyd Orr Centre for Population and Ecosystem Health
School of Veterinary Medicine
University of Glasgow
464 Bearsden Road
Glasgow – G61 1QH

Dear Tim,

Regarding the use of clinical data for the study of the clinical use and adverse effects of NSAIDs in the UK horse population

Our usage policy, including data ownership, anonymisation, security and confidentiality for our data is as follows:

1. The data will remain property of (practice name)
2. Any costs involved with providing the data will be payable by the investigators of the study
3. Data will be supplied in an anonymised format wherever possible
4. Confidentiality will be maintained and secure physical and electronic storage ensured
5. The facilities for secure and physical electronic storage of the data may be scrutinized
6. Any papers, publication or presentations resulting from the data will be made available to (practice name).

Please sign below to show your agreement with this policy for this project.

Practice representative to sign Date: ……..

Practice representative print name *MRCVS*, Partner at (practice name)

Please sign one copy of this letter and return to me at the address above

Signed: …… Date: ……..

Name: Tim Parkin

Appendix 3.2

Microsoft Access Instructions For Data Anonymisation

MS Access - assign unique ID procedure

STEP 1

Open dataset with excel.
Add new column by RIGHT clicking on letter A of the first column (see Figure 1 – black arrow – this will highlight the whole column) and selecting *Insert* in the menu that then should open (with the RIGHT click). This will add an empty column and shift all the others to the right.

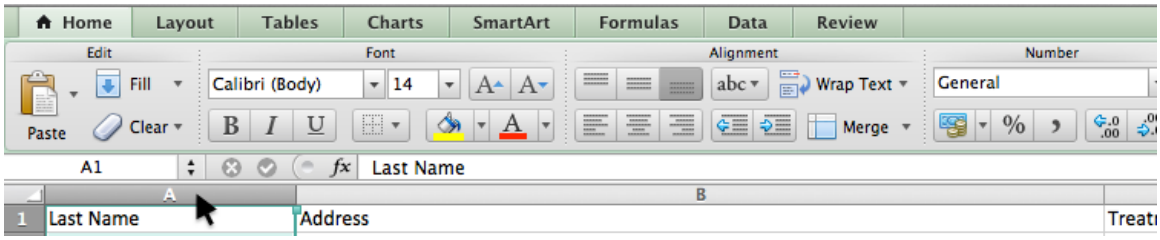

Figure 1

Then one LEFT click on cell A2 and type as follows:
=concatenate(B2,G2,H2,I2)
Press enter to confirm

Please, substitute B2, G2, H2 and I2 with the name of the cells that contain the first Last Name, Horse Name, Breed and DOB – please do not include the address column (B,G,H,I may vary but should always be followed by "2" – as you are selecting cells in row 2).

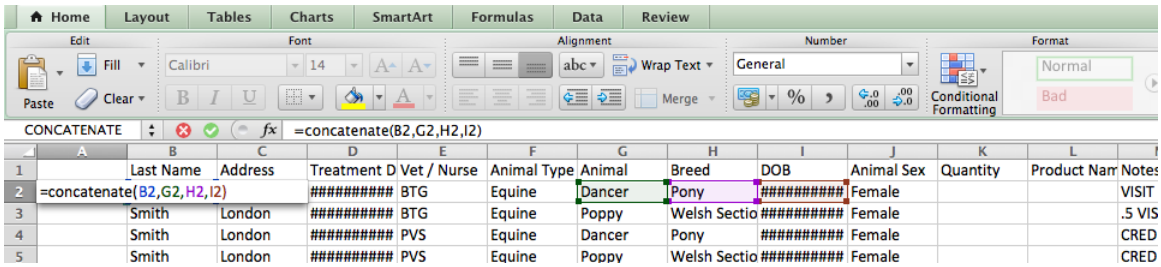

Figure 2

This will collate all the information in the cells selected into one string in one cell.

Then double click on the bottom right corner of cell A2 (the cursor should become a black cross if you are in the right spot). This will apply the concatenate function for the rest of the dataset (it may take a few moments to do so depending on the size of the dataset).

Save your file as a new file (eg. Record_with_ID.xlsx)

STEP2

In Access create a new blank database (Figure 3):

1. Click on *Blank Database*
2. Write a file name under *File Name* on the right
3. Click *Create*

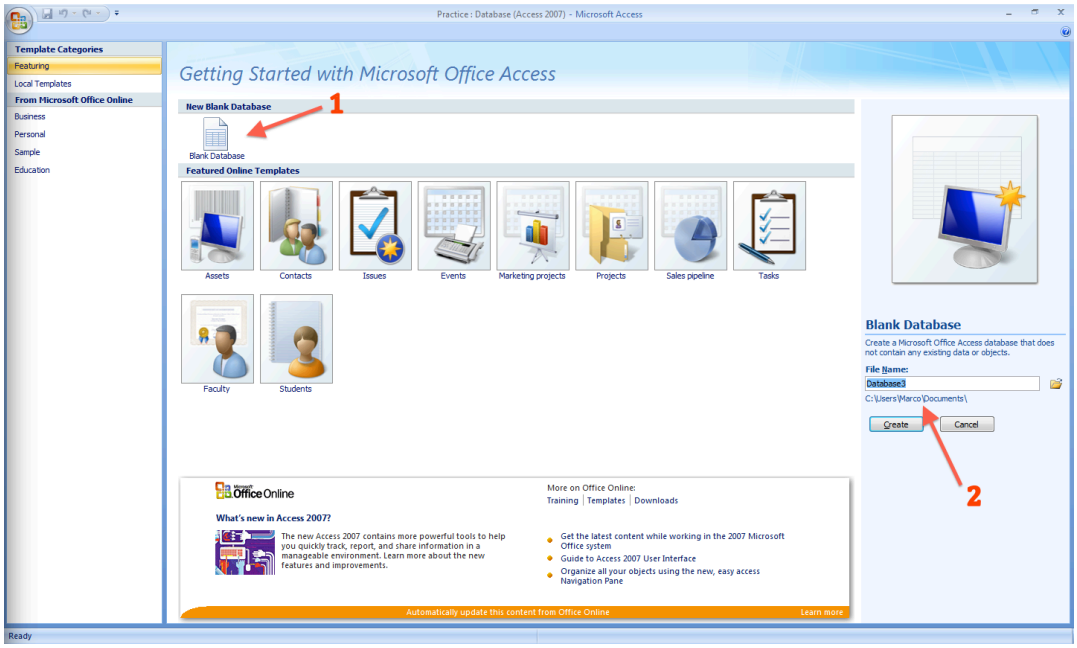

Figure 3

Import excel spreadsheet in MS Access by following these steps:

1. Click on *external data* tab –> *excel* icon under Import tab-> click on Browse and select path where the file you just created "*Record_with_ID.xlsx*" is located.
2. This will open a wizard window.
3. Select "*Import the source data into the new table in the current database*" and click OK.
4. In the first page please tick on *First Row Contains Column Headings* (if an error message opens just click OK) and click Next.
5. Under tab field name write *NameID* and click Next
6. On the next page select *No primary key* then click Next.
7. Click on Finish then Close (this will add *Sheet1:Table* on the menu on the left).

Assign a unique ID:

- Select the Tab *Create* and then *Query Design* (under Other). When a wizard entitled "*Show table*" opens add the table with the name you gave it when you imported the data (there should be only one table with data in it at this stage – we named it *Practice* in this example), then click on close in the *Show table* window.
- Then click on the design tab at the top (arrow 1) and on Σ_{Totals} (arrow 2). This will take you to the window shown in **Figure 4**. Double-click on NameID TWICE (arrow 3 - this will add NameID TWICE in the panel at the bottom. Then click where indicated by arrow 4 (in the row called *Total:* of the second column – it should say *Group by* at this stage but once you click on it a scroll down menu should open) and select *Count*. Then click on save (and OK in the save window that opens) and Run (arrows 5 and 6) – see Figure 4 as a guide.
- Once you run the query it will give your NameID columns and a Count column (each number corresponds to the times each horse is entered in the records).

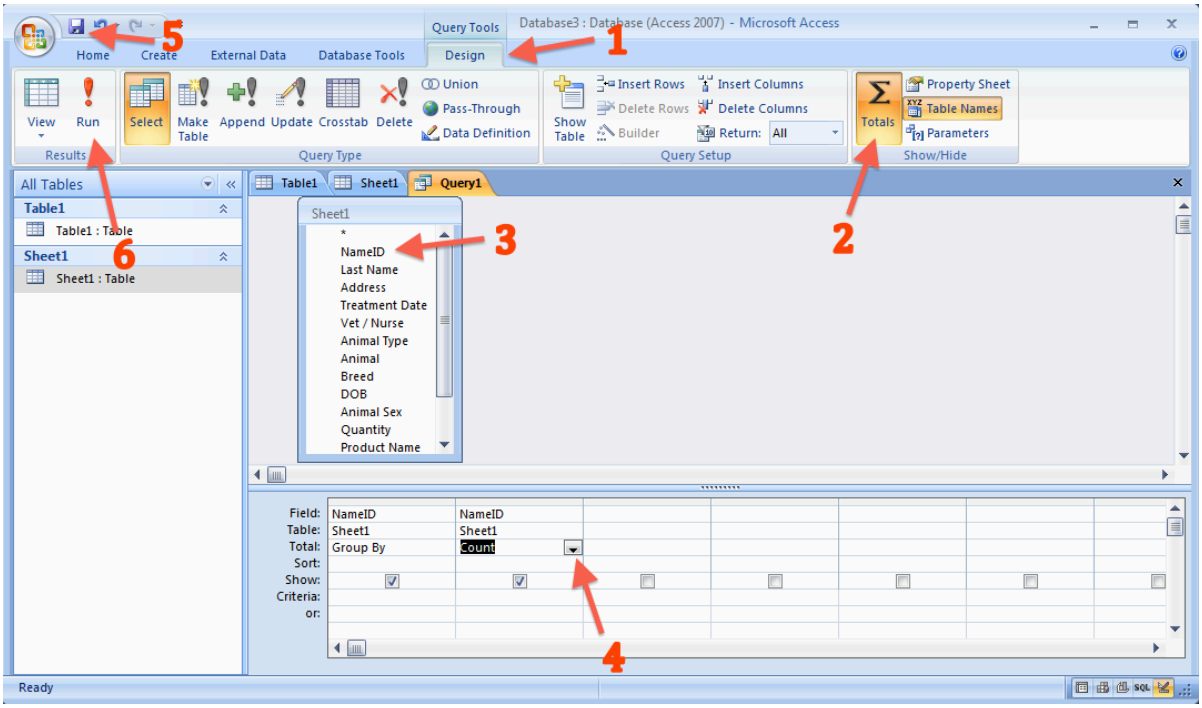

Figure 4

- Copy and paste the column *NameID* by RIGHT clicking where indicated by Figure 5 (arrow 1) and selecting copy in the menu that opens. Then click on the *Create* tab and select *Table* (steps 2 and 4 of figure 5).

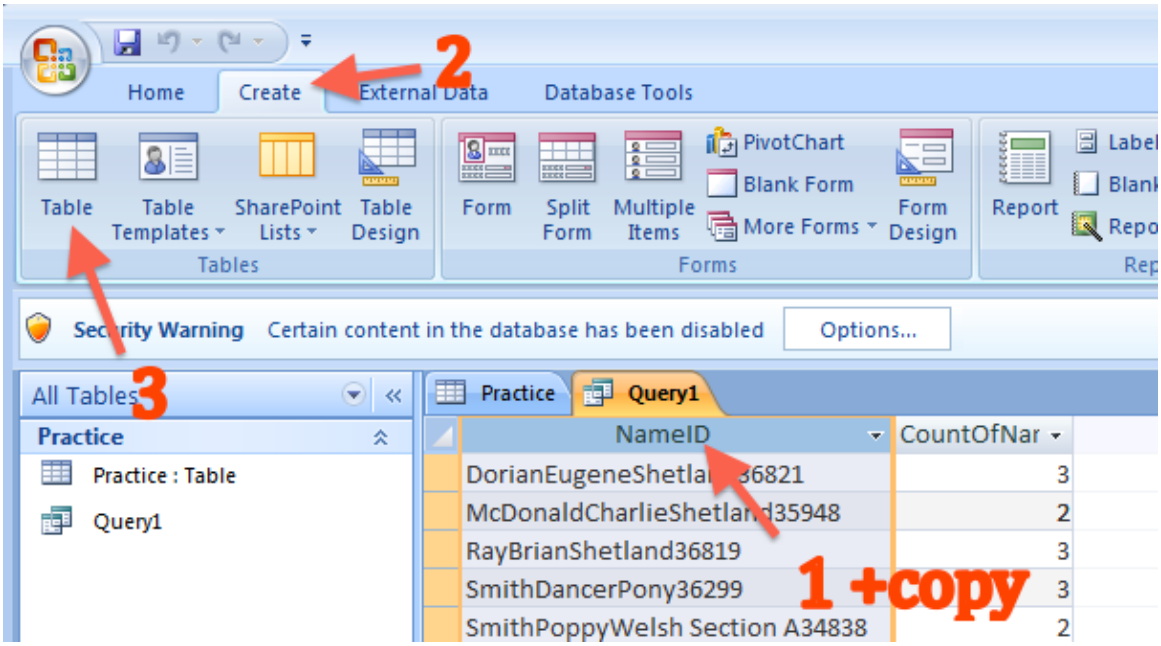

Figure 5

- On the menu on the left, LEFT click on *Table1:Table* and select *Design view*. Name the new table "*ID number*" when asked.
- On the new tab window that opens, type *ID number* in the first row of column *Field name* and leave *AutoNumber* in the *Data Type* column. In the second row write *ID name* in the first and select *text* in the second column respectively. After saving, RIGHT click on *ID number* tab and select *datasheet view* (use figure 6 as a guide).

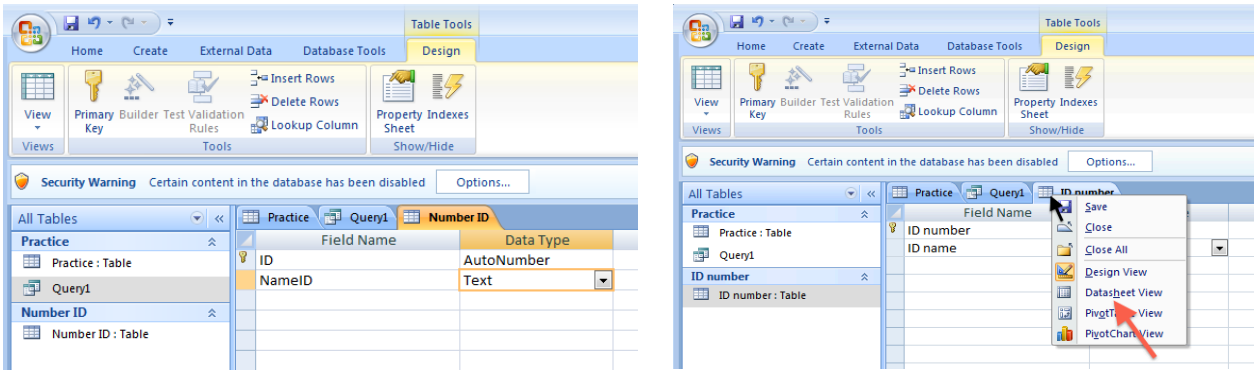

Figure 6

- Now, RIGHT click on *ID name* and select *paste*. Click on yes if Access asks if you are sure to paste the XXX record(s) (Figure 7). This will add all the ID name content in the ID name column and assign automatically an ID number in the ID number column.

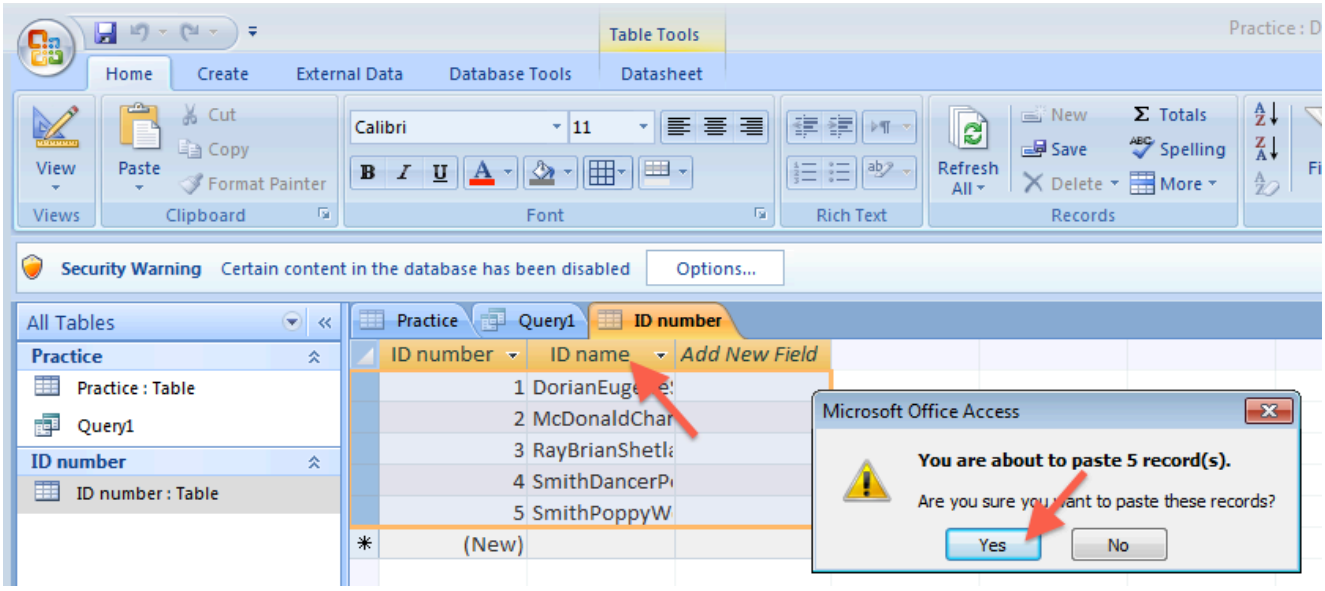

Figure 7

- Now, create a query (*Create* -> *Query Design*) and include both (double click on each) initial table with dataset ("Practice" in this example) and the newly created table ("ID number") then select close.
- Drag *ID name* from the ID number window to NameID on the left (be precisely right on NameID). Use Figures 8-9-10 as a guide.

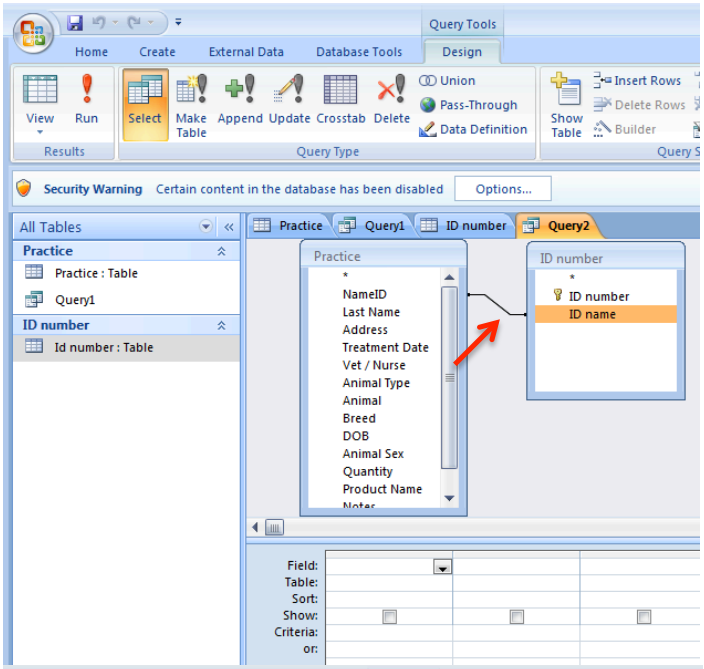

Figure 8: Drag *ID Name* from ID precisely onto *Name ID* in the Practice window. This will create a link (black line between these two).

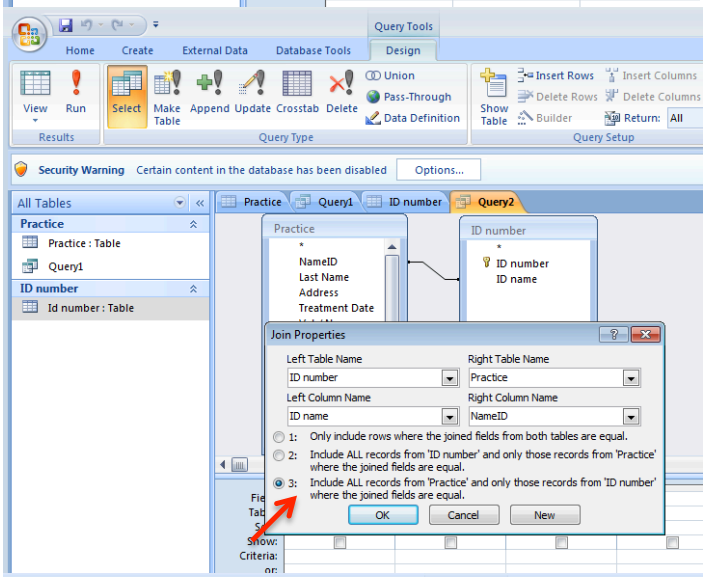

Figure 9: double click on the black link line and the *Join Properties* window will open. Here select the option that says: *Include ALL records from 'Practice' and only those from 'ID number' where the joined fields are equal* -> this is option 3 in this example. Then click OK.

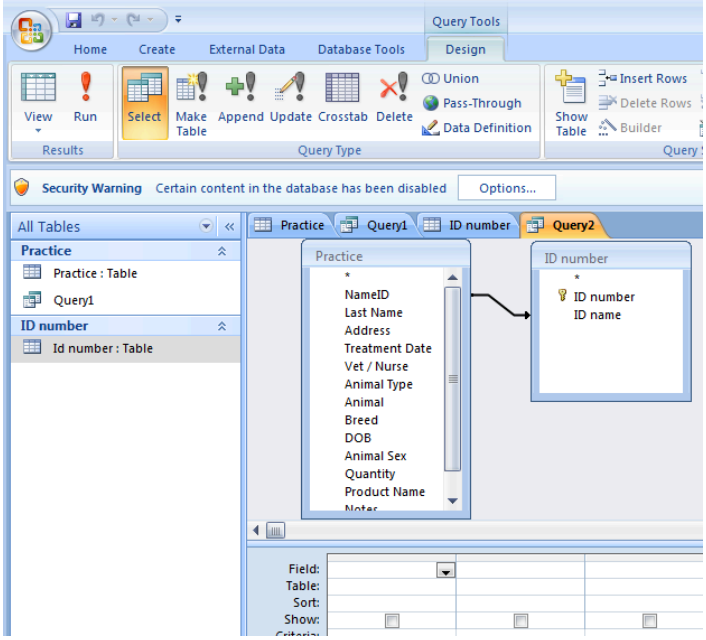

Figure 10: If these steps were done correctly the black link line will become a black arrow.

- Double click on *ID number* in ID number window, to add ID number in the first column at the bottom. Select all the content in Practice window and drag to second column at the bottom (this will add each of them in each column). See figure 11 for reference.

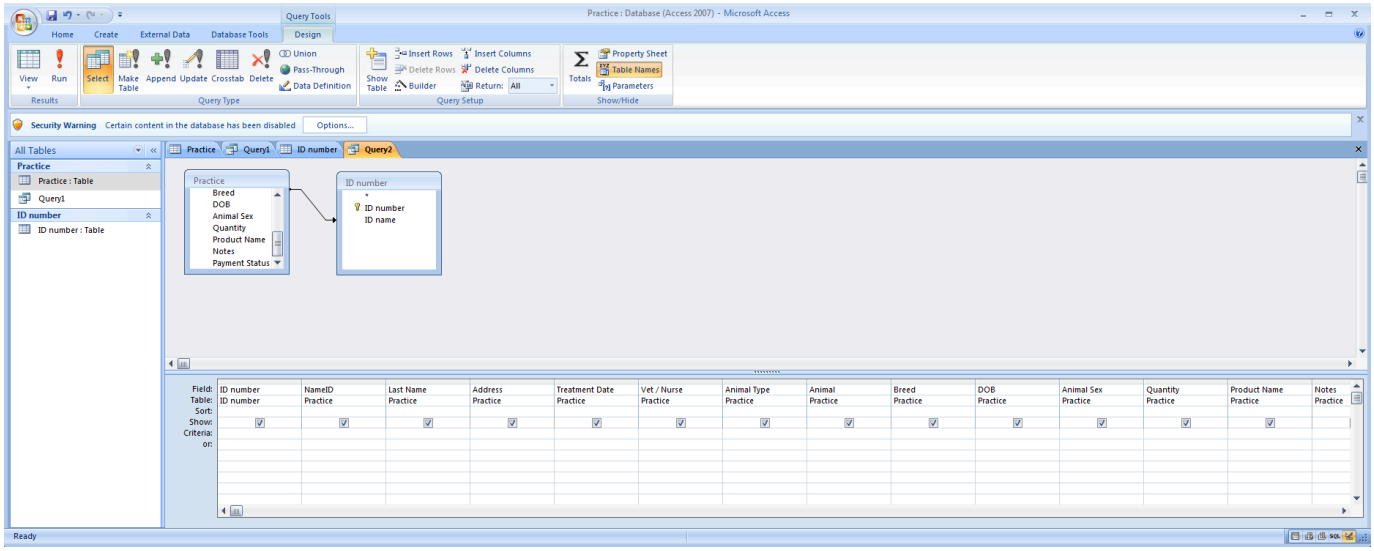

Figure 11

- Click on Run and obtain something like this in Figure 12 (The original Practice table with the first column containing unique ID numbers – same horse has same ID number when revisited).

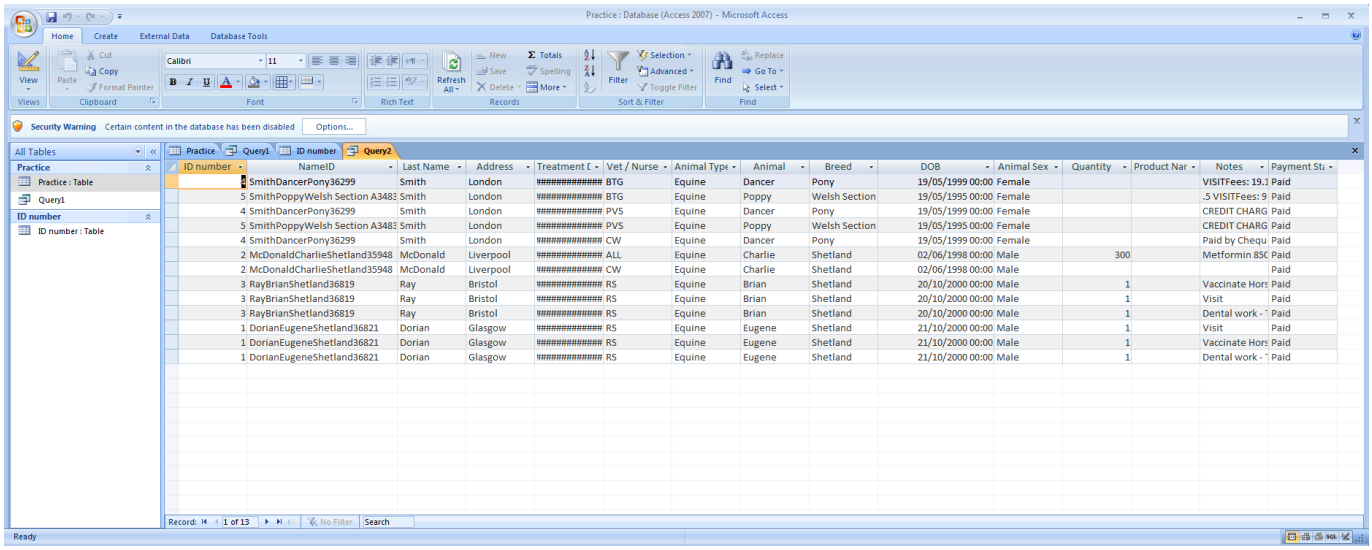

Figure 12

Now only few more clicks and we are done.

Click on External Data and Excel in the Export tab. This opens the export wizard. Select a filename (maybe the name of your veterinary practice) and location and tick the first 2 options (*Export data with formatting and layout* and *open the destination file*) and click on OK (Figure 13 as reference).

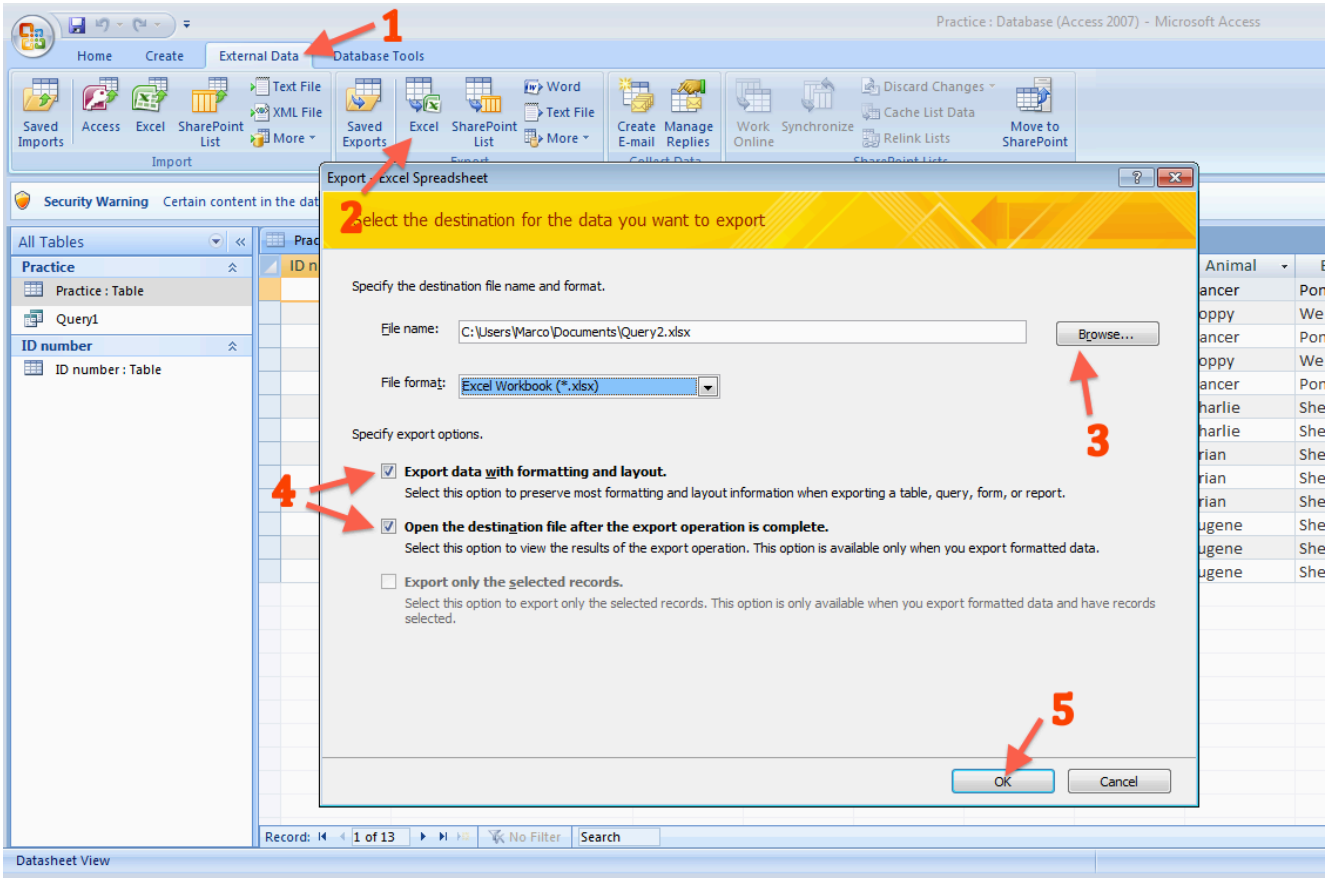

Figure 13

This will then open an excel spreadsheet containing the records with the new anonymous ID numbers in the first column. If an error message shows just click on *yes* or *ok*. If the file opens as a read only please "save as" with another name in order to be able to modify it.

STEP3

Now we have only to remove the columns containing owners' details to anonymise the dataset. To do so LEFT click on the letter at the top of the column you want to delete (Name ID and the one with owner last name, address and animal name) whilst pressing on the CTRL key on your keyboard. This will select each column in its entire length. Finally one RIGHT click on the Last column and select delete (blue arrow). This will delete all the selected columns and shift all the others to the left. Alternatively you can delete each column individually (right click and select delete at the point of each read arrow in figure 14).

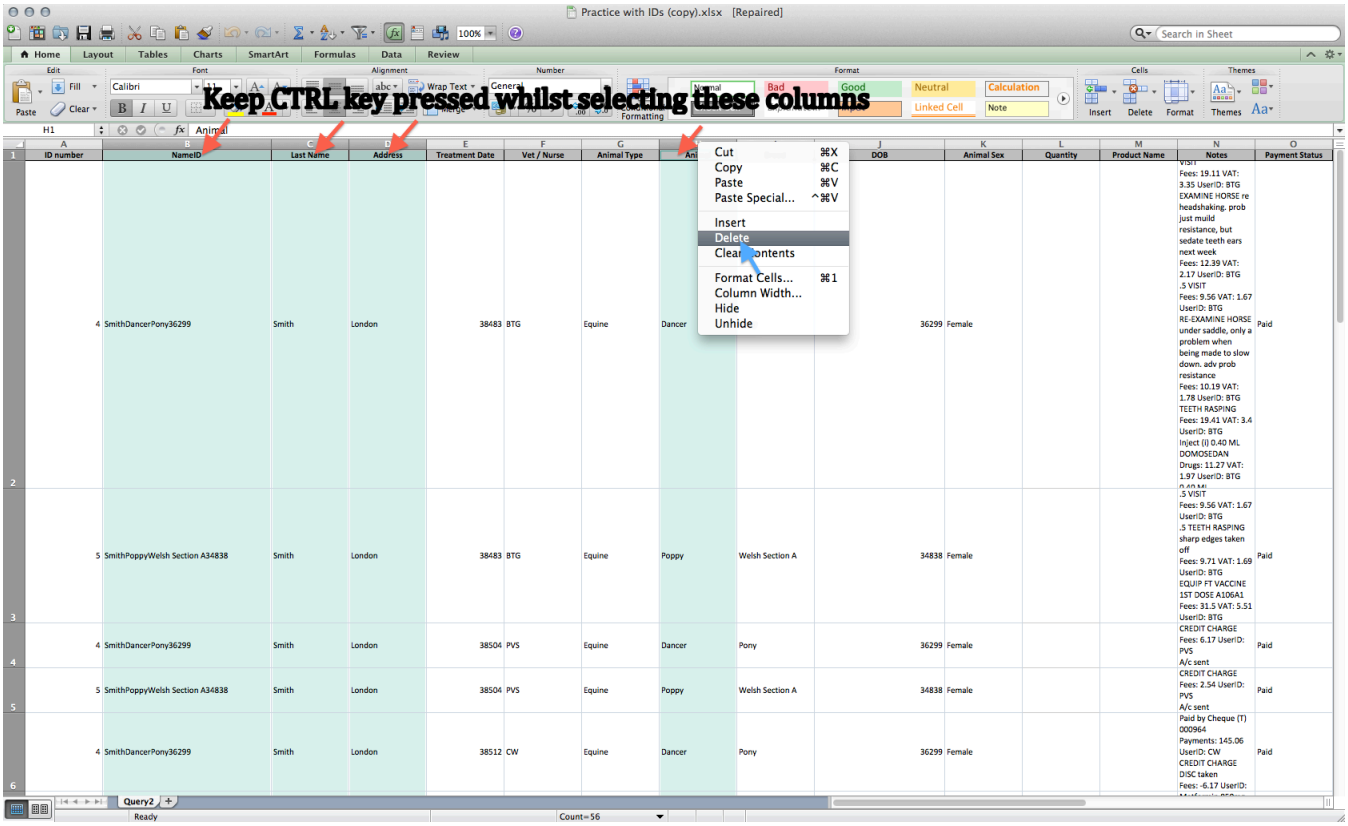

Figure 14

Finally save the file and you are done. Hopefully it will be small enough to be sent by email.

Thank you very much again for all your effort and help.
May you not succeed with these instructions (eg. your access/excel version is remarkably different from mine), call me [redacted]) or alternatively I am quite happy to come there in person to sort it out in the near future.

Kind regards,
Marco

Appendix 3.3

NSAIDS INCLUSION DICTIONARY

ABS/NSAIDS
ALSODANILON
ANTI-INFLAMMATORIES
ANTIINFLAM
ARQUEL
ASPIRIN
ASPSOL
BINIXIN
BUSCA
BUSCO
BUSCOPAN
BUSCOPAN_CO
BUSCOPAN_COMPOSITUM
BUTE
BUTEEQUIP
BUTEINJ
BUTEPASTE
COMPOSITUM
CRONY
CRONYXIN
DANIL
DANILON
DANILON-PER
DANILONAS
DYNAM
EQUIDOX
EQUIOXX
EQUIPAL
EQUIPALAZONE
FELDENE
FINAD
FINADNE
FINADYNE
FLUN
FLUNIXIN
KETOFEN
KETOPROFEN
MELOXICAM
METACAM
METCAM
NOROCARP
NSAIDS
PAINKILLERS
PBZ
PBZ?
PBZ/DANILON

PHENYLBUTAZONE
PRO-DYNAM
PRODYNAM
REST/BUTE
SACHETSBUTE
TELZENAC
TOLFINE

NSAIDS EXCLUSION_DICTIONARY
BUSC
BUSCA
BUSCOPAN
BUSCO

NSAIDS RE-INCLUSION_DICTIONARY
BUSCOPAN_CO
BUSCOPAN_COMPOSITUM
BUSC_CO
BUSC_COMPOSITUM

COLIC INCLUSION_DICTIONARY
?COLIC.HR40,PULSE
?COLICKY
ADB_DISCOMFORT
CHOKE
CLINICAL_NOTE_SEVERE_PAIN_ON_ARRIVAL.
COLIC
COLIC
COLIC,PARAMETERS
COLIC.
COLIC.
COLICING
COLICING.
COLICKING
COLICKING
COLICKY
COLICKY.
COLICY
COLICY,
COLICY.PASSED
COLITIS
COLOUR,?COLIC
CONSTIPATION
DISPLACED
DISPLACEMENT
DISPLACEMENT/IMPACTION_FELT.STOMACH
DISTENDED_LARGE_INTESTINE
DROPPINGS_PASSED._LYING_DOWN
ENTEROCOLITIS
EPISODE_OF_ROLLING

EPSOM
FLANK_WATCHING
FLANK_WATCHING.
GASSEOUS_DISTENDEN
GASTRIC
GASTROSCOPY
HAVE_BEEN_RESTLESS_AND_PAWING_BEDDING.
IMOPACTION
IMPACTION
MASSIVELY_IMPACTED
MILDLY_UNCOMFY
NEFROSPLENIC
NEPHRO_SPLENIC_ENTRAPM.
PARAFFIN
PASSSTOMACHTUBE
PELVIC_FLEXURE_IMPACTION.
PSYLLIUM
REFLUX
RETROFLEXION_OF_LARGE_COLON.
ROLLING
SAND
SANDOUT
SHOWING_SIGNS_OF_ABDOMINAL_PAIN
SIGNS_OF_ABDOMINAL_PAIN
SPASMODIC
SPASMOLYTIC
SPASMOTIC SQUAMOUS
TWIST
STARTD
UNCOMFORTABLE._GUT_SOUNDS_-VE.
UNSETTLED_IN_BOX

COLIC EXCLUSION_DICTIONARY
!TRANSPORT_STRESS/VIRAL
-03-14_LAB:_INFLAMMATORY_PROFILE
-04-20_JOURNEY
-09-28_SAND_OUT
...LAMINITIS,_V_DULL
0.5LTR_REFLUX
1999-09-28_SAND_OUT
1____EQUINE_INTRAVENOUS
?TRANSPORT_STRESS/VIRAL
ATTEMPTS_TO_STOMACH_TUBE_FAILED
BURPS._LIKELY_GASTRIC_IN_ORIGIN
CAER_RE_IMPACTION
CHECKED_TEETH
CLEAR_IMPACTED_DIASTEMATA
CLINICAL_NOTE_NO_NET_REFLUX
COFFIN
COLIC_ON_THE_29.01.10

D++_BUT_NO_COLIC_SIGNS
DENTAL_EXAM
DEPRESSED_BUT_NOT_COLIC
DEPRESSED_BUT_NOT_COLIC.
DIASTERMA
DIDN'T_NEED_TO_STOMACH_TUBE_TODAY
DISCUSSED_POSS_CAUSES_AND_POSS_GASTRIC_ULCERATION
DISPLACED
DISPLACED,_MONITOR_AS_ERUPTS
DISUCSSED_COLIC_AND_FEET
DOLX_COLITIS,
EQH_BAXC_BAXTER_DRAPE-_COLIC
EQH_COLIC_GOWN
EQH_HICU_INTENSIVE_CARE_DAILY_FEE_HICU_INCL_MULTIPLE_ST
OMACH_TUBE
EQH_HICU_INTENSIVE_CARE_DAILY_FEE_INCL._MULTIPLE_STOMAC
H_TUBE
EQH_HICU_INTENSIVE_CARE_DAILY_FEE_INCLUDING_MULTIPLE_ST
OMACH_TUBE
EQH_SEDAL_SEDALIN_GEL_GIVE_1/3_TUBE_ORALLY_TWICE_DAILY
_FOR_5_DAYS
EQO_EPS2_MAGNESIUM_SULPHATE_EPSOM_SALTS_2KG_0.25_0.25_
BOXES_0
EQO_EPSOM_EPSOM_SALTS_100G_1_1_BOX_0
EQO_HST_STOMACH_TUBE
EQO_THST_STOMACH_TUBE
EQUINE_STOMACH_TUBE
FAECAL_BALLS_PRESENT
FAECES_IN_RECTUM_-_NO_OTHER_ABNS
FIBRE
FLUSHED_ABDOMEN_AND_TUBED_HORSE
FRACTURE
GUTS_QUITE_GURGLY_BUT_NO_CSX_OF_COLIC
GUT_SOUNDS_REDUCED_ALL_ROUND,
G_VENTIPULMIN_2_1/2_SCOOPS_TWICE_DAILY_BATCH:0256358
HOCK_DISPLACED
DIFFICULTY_SWALLOWING
HOOF
HR_32,_MM_PINK,_CRT_2SECS,_GUT_SOUNDS_NORMAL_TO_LOUD,
HST_STOMACH_TUBE
IF_COLICS_THEN_REFLUX
ILIAL_WINGWHICH_WAS_DISPLACED
IMPACTION_CLEARED,
INFLAMMATORY_PROFILE_-_LAB
INTERMITTENT_D+._NO_WEIGHT_LOSS._BRIGHT_INSELF._STILL_H
AS_GRADE_III_SYSTOLIC_MUMUR_AND_ARRHYTHMIA
LAB:PARASITOLOGY_-_LAB_REF
LAB:_INFLAMMATORY_PROFILE_LAB_REF:_A167_A167_MILD
LAME
LAMENESS

LAST_DONE_FOR_LARVAL_STAGES.NAD_ON_RECTAL.HAS_PASSED
LATERALLY_DISPLACED
LATERALLY_DISPLACED_TOOTH
LHS_AFTER_30_MINUTES
LIKELY_GASTRIC_IN_ORIGIN.
LIQUID_PARAFFIN_5_LITRE
LIQUID_PARAFFIN_X_1LT_LA
LYING_DOWN_NOT_COLICING.
LYING_DOWN_NOT_COLLIDING
MAKE_COMFORTABLE_RECTAL_NAD_'RETCHING'_TYPE
NA_PAID_BY_VISA_T_PAYMENTS:_15.42_USERID:_CY_BISHOPTON
NON-MALODEROUS_PURULENT_L_SIDED_DISCHARGE
NOTE_FLUSHED_ABDOMEN_AND
NOT_SHOWING_ANY_COLIC
NOT_SHOWING_ANY_COLLIC_SIGNS
NO_ABDOMINAL_HEAVE_BUT_INC_ADVENTIOUS_SOUNDS
NO_ACTIVE_COLIC_SIGNS
NO_COLIC_SIGNS
NO_COLIC_SIGNS,
NO_COLIC_SIGNS.
NO_CSX_OF_COLIC.
NO_FBS_DETECTED_&_STOMACH_TUBE_PASSED_FINE
NO_IMPACTION_FELT_SOME_REDUCED_AMNT_FAECES_ADV_PROB_
URTI
NO_SIGNS_CHOKE
NO_SIGNS_OF_COLIC
NO_SIGNS_OF_COLIC.
NO_SIGN_OF_COLIC
OF_CONSTIPATION_AND_POOR_APPETITE.
OF_CONSTIPATION_AND_POOR_APPETITE._EYE_LOOKS_AS_BEFOR
E,
OVERNIGHT_COMFORTABLE_NO_SIGNS_OF_COLIC
OWNER_CONCERNED_HORSE_HAS_BEEN_ACTING_STRANGELY._ON
OWNER_CONCERNED_RE:_SQUAMOUS_CELL_CARCINOMA.
PASSEDNG_TUBE
PASS_STOMACH_TUBE_1_PFEE
POLE?STOMACH_TUBE_-_NO_REFLUX
POOR_INCISORS,_208_LARGE_SHARP_POINT
PRESENT,STOMACH_TUBE_REFLUX_APPROX
PREVNT_IMPACTION
PSYLLIUM_500GM
RECTAL_EXAM:_IMPACTION_CLEARED
RECTAL_EXAM:_IMPACTION_GONE
REMOVE_FOOD_PACKING,
RESTRICTED_FEED,_IF_COLICKY_OR
SAND_CRACK
SAND_ON_R_CIRCLE
SEDATED_TO_MAKE_COMFORTABLE_RECTAL_NAD_'RETCHING'_TYP
E
SHARED_VISIT

SINUS
SL_DISPLACED._ON_SCOPE_-_DUE_TO_SWELLING_ONLY_POSS_TO
SOFT_F+_PRESENT_IN_RECTUM
SOFT_F+_PRESENT_IN_RECTUM_NO_IMPACTION_PALPABLE
SOME_FORAGE_IMPACTION
SO_SIGNS_CHOKE.
SQUAMOUS_CELL_CARCINOMA_SHEATH_HAS_A_LOT_OF_SMEGMA
STOMACH_TUBE_REFLUX_APPROX_1L
STOMACH_TUBE_THIS_AM.ALSO_ONTO_B-SURE_AS_POSS_MORE
STOMACH_TUBING_MIDDAY_AND_FIRST_LITTLE_MASH
ST_LINE_SAND_ON
PROB_URTI_COS_OF_TRAVELLING
TOPAZ_HAD_RADIOGRAPHS_OF_HER_TEETH_AND_SINUSES
TO_STOMACH_TUBE_FAILED.DISCUSSED,ADV
TUBED_AGAIN_WITH_5_L_ELECTROLYTES_AND_PARAFIN
UNSTABLE_SANDCRACK
VALLEY_VETS_SAND_SUPP
VENTIPULMIN_2_1/2_SCOOPS_TWICE_DAILY_BATCH:0256358
WELL_NO_COLIC_SIGNS,
WENSUM_1_1_LIQUID_PARAFFIN_2_LITRES
WENSUM_BUTEINJ_15_MLS_INJECT_EQUIPALAZONE_INJ_WENSUM
WEN_SUM_SANDOUNT_1_TUB_SAND-OUT_908G
WORMS/STABLING/GASTRIC_ULCERATION/MOTILITY_CHANGES
WOUND_SLIGHT_ODEMA

COLIC RE-INCLUSION DICTIONARY
FLUSHED ABDOMEN AND REFLUXED HORSE AGIAN THIS PM
FLUSHED ABDOMEN AND TUBED HORSE
BUSCOPAN. POSS SPASMODIC COLIC
MILD COLIC. NORMAL DROPPINGS
COLON NOT DISTENDED SO ADVISE PROB MILDLY DISPLACED

RIGHT DORSAL COLITIS INCLUSION
RIGHT DORSAL COLITIS,
RDC EPISODE
RD_COLITIS
RD COLITIS.
NSAID_INDUCED
?NSAIDS INDUCED

RENAL_FAILURE INCLUSION DICTIONARY
KIDNEY_DISEASE
KIDNEY_FAIL
KIDNEY_INSUF
KIDNEY_INSUFFICIENCY
PAPILLARY_NECROSIS
RENAL_DISEASE
RENAL_FAIL
RENAL_FAILURE
KIDNEY_FAILURE

RENAL_INSUF
RENAL_INSUFFICIENCY
URINARY_DISEASE
URINARY_FAILURE

RENAL_FAILURE EXCLUSION DICTIONARY
ACUTE_RENAL_FAILURE_NO_URINE_RECTAL
RENAL_DISEASE_?_!_C/EXAM_NAD.
SG_LOW_1.040_DISCUSS_DILUTE_URINE
SEVERE_DEHYDRATION_AND_PRE_RENAL_FAILURE

Appendix 4

ASPIRIN INCLUSION DICTIONARY
- ACETYLSALICYLIC
- ACETYLSALICYLIC_ACID
- ASPIRIN
- ASPRIN
- ASPSOL

PHENYLBUTAZONE INCLUSION DICTIONARY
- BIZOLIDIN
- BUTATRON
- BUTAZOLIDIN
- BUTE
- BUTEEQUIP
- BUTEINJ
- BUTEPASTE
- BUTEQUINE
- COMPANAZONE
- DYNAM
- EQUIBUTE
- EQUIBUTE
- EQUIPA
- EQUIPAL
- EQUIPAL
- EQUIPALA
- EQUIPALAZ
- EQUIPALAZO
- EQUIPALAZONE
- EQUIPALAZONE
- EQUIPALAZONE
- EQUIPALIZONE
- EQUIPALOZONE
- EQUIPALZONE
- EQUIPHEN
- EQUIZONE
- MBUTAZOLIDIN
- PBZ,
- PBZ.
- PBZ/DANILON
- PBZ?
- PHEMYLBUTAZONE
- PHEN_BUTA
- PHENBUTA
- PHEN-BUTA
- PHENYBUTAZONE
- PHENYLBUATZONE
- PHENYLBUTASONE
- PHENYLBUTAZONE

PHENYLBUTE
PHENYLZONE
PRIBUTAZONE
PRO_DYNAM
PRODYNAM
PRO-DYNAM
REST/BUTE
ROBIZONE
RXBUTE
SACHETSBUTE
SUPERIORBUT
TEVCODYNE
THERAZONE

SUXIBUZONE INCLUSION DICTIONARY
ALSODANILON
DALILON
DANALON
DANALONE
DANEQ
DANEQUIB
DANI
DANIDOL
DANIL
DANILAN
DANILAON
DANILLON
DANILON
DANILONAS
DANILON-PER
DANILONE
DANILONSID
DANION
DANOLIN
DANOLONE
DNAILON
SAXIBUZON
SUXIBUZONE
SUXILON
VET-DANILON
XDANILON

SUXIBUZONE EXCLUSION DICTIONARY
ASPHALT_DANI
BY_DANI
CC_DANI
CHIP_DANI
DANI_-_BASIC
DANI_-_LAMENESS_EVAL
DANI_..._BEHAVE

DANI_APPEARS
DANI_BUHLER
DANI_CALLED
DANI_DAUGHTER
DANI_FROM
DANI_HAS
DANI_IS
DANI_LACERATION
DANI_MADILL
DANI_OSTER
DANI_RINDFLEISCH
DANI_VOLK
FOR_DANI
HAVE_DANI
OSTER_DANI
DANI_RINDFLIESCH
PALPATED_DANI
PER_DANI
TEETH_DANI
WITH_DANI
SHOES_ON_DANI

FIROCOXIB INCLUSION DICTIONARY
EQUIOOX
EQUIOX
EQUIOXX
FIBROCOX
FIBROCOXIB
FIROCOXIB
PREVACOX
PREVICOX
PREVICOXX

FLUNIXIN MEGLUMINE INCLUSION DICTIONARY
BANA
BANAMIN
BANAMINE
BANMINE
BANNAMINE
BINIXIN
CRONIXIN
CRONY
CRONYXIN
EQUINIXIN
FINAD
FINADINE
FINADNE
FINADYN
FINADYNE
FINADYNE/BUSCO

FINDYNE
FLUN
FLU_NIX
FLUNAZINE
FLUNIX
FLU-NIX
FLUNIXAMINE
FLUNIXIN
FLUNIXIN_MEGLUMINE
FLUNXIN
FYNADINE
HEXASOL
MBANAMINE
MEFLOSYL
MEGLUMINE
NOXIRIN
OXYCOMPLEX
RESFLOR
RXBANAMINE

KETOPROFEN INCLUSION DICTIONARY
ANAFEN
AXORID
COMFORION
DANIDOL
DINALGEN
KELAPROFEN
KETOCID
KETODALE
KETOFEN
KETOPHEN
KETOPROFEN
KETOPROPIG
KETOVAIL
ORUDIS
ORUVAIL
POWERGEL

MELOXICAM INCLUSION DICTIONARY
ADOCAM
ANIMELOX
FLEXICAM
LOXICOM
MELOSUS
MELOVEM
MELOXICAM
MELOXIVET
MELOXORAL
MELOXYDYL
METAC

METACAM
METACINJ
METACJ
METCAM
METCAMMETAC
MOBIC
NOVEM
RECOCAM
RHEUMOCAM

METAMIZOLE_UK INCLUSION DICTIONARY
BUSC
BUSCA
BUSCAPAN
BUSCAPAN
BUSCO
BUSCO
BUSCOPAN
BUSCOPAN
COMPOSITE
COMPOSITION
COMPOSITUM
DIPIRONE
DIPYRONE
DYPIRONE
DYPIRONE

METAMIZOLE INCLUSION DICTIONARY
_CO
_CO.
_COMP
_CO,
_COMP.

METAMIZOLE_UK EXCLUSION DICTIONARY
ADV_CO
ALL_3_COMP.
BANDAGE_-_CO
BEAN&CO._
COLAST10_CO_
COMP_ID_DRAW
CO_BACTAN
FOR_A_WEEK,_CO
INCREASE_CO
INDIVIDUALLY_SO_NO_COMP.
INSURANCE_CO,
INSURANCE_CO_
I_WILL_CO_
PRESENT._CO_
RELUCTANT_CO_X_

TJHIS_IS_CO_
TO_BE_CO_
REX_HORSE_CO_
TO_IMPROVE_CO_
TRIMMING_TO_CO
_CO_PLUS.

METAMIZOLE NORTH AMERICA INCLUSION DICTIONARY
COMPOSITUM
DIPIRONE
DIPYRONE
DYPIRONE
METAMIZOLE

ACETAMINOPHEN INCLUSION DICTIONARY
ACETAMINOPHEN
NOXIRIN
NOXPIRIN

CARPROFEN INCLUSION DICTIONARY
CARPROFEN
CARPROFENL
NOROCARP
RYMADIL
RIMADYL

DERACOXIB INCLUSION DICTIONARY
DERACOXIB
DERAMAX
DERAMAXX

DICLOFENAC INCLUSION DICTIONARY
DICLOFENAC
DICLOFINEC
SURPASS

ELTENAC INCLUSION DICTIONARY
ELTENAC
TELZENAC

MECLOFENAMIC_ACID INCLUSION DICTIONARY
ARQ
ARQUEL
ARQUELL
MECLOFENAMIC_ACID
MECLOFENAMATE

TOLFENAMIC_ACID INCLUSION DICTIONARY
TOLFENAMIC
TOLFINE

VEDAPROFEN INCLUSION DICTIONARY
QUADRASOL
QUADRISOL
VEDAPROFEN

COLIC INCLUSION DICTIONARY
ABDOCENTESIS
ABDOMINOCENTHESIS
ADBOMINOCENTESIS
CELIOTOMY
CHOKE
CHOKED
CHOKES
CHOKING
CHOLIC
COELIOTOMY
COILC
COLCI
COLCIKY
COLCKY
COLIC
COLICAGAIN
COLICALLY
COLICCS
COLICCY
COLICD
COLICE
COLICED
COLICER
COLICERY
COLICEXAM
COLICFY
COLICI
COLICIING
COLICIKY
COLICING
COLICK
COLICKE
COLICKED
COLICKED
COLICKER
COLICKEY
COLICKING
COLICKLY
COLICKS
COLICKY
COLICKYING
COLICL
COLICLY

COLICN
COLICS
COLICSTRIC
COLICTHAT
COLICY
COLICY
COLICYING
COLIIC
COLIITIS
COLIKED
COLIKING
COLIKY
COLILC
COLILCY
COLITIS
COLLIC
COLLICED
COLLICKED
COLLICKING
COLLICKY
COLLICS
COLLICT
COLLICY
COLONIC
COLONIC
CONSTIPATED
CONSTIPATION
DISPLACED
DISPLACEMENT
DISPLACEMENTS
DISPLACMENT
DISTENDED_LARGE
DISTENDED_SI
DISTENDED_SMALL_INTESTINE
DOCUSOL
ENTERITIS
ENTEROCOLITIS
ENTEROLYTH
ENTEROLYTHIASIS
ENTEROTOMY
EPIPLOIC
EPSOM
EPSOM_SALTS
ESOPHAGEAL_DIVERTICULUM
ESOPHAGEAL_FOREIGN_BODY
ESOPHAGEAL_OBSTRUCTION
ESOPHAGEAL_RUPTURE
ESOPHAGEAL_STENOSIS
ESOPHAGEAL_STRICTURE
ESOPHAGEAL_TEAR

EXPLORATORY
FECALITH
FLANK_WATCHING
GASEOUS_DISTENDEN
GASSY_COLIC
GASTRIC
GASTRIC_ULCER
GASTROSCOPY
GAS_COLIC
GLANDULAR
GUT_INFLAMMATION
IBD
IMOPACTION
IMPACTED
IMPACTION
IMPACTION
INFLAMMATORY_BOWEL
INTESTINAL_ISCHAEMIA
INTESTINAL_ISCHEMIA
INTRALUMINAL_OBSTRUCTION
ISOGEL
LAXATIVE
LDD
LIQUID_PARAFFIN
MARGO_PLICATUS
MEED
NEFROSPLENIC
NEMATODES
NEPHORSPLENIC
NEPHROPLENIC
NEPHROSLENIC
NEPHROSPENIC
NEPHROSPLENIC
NEPHRO_SPLENIC
NOT_IN_THE_RIGHT_PLACE
PARAFFIN
PASSSTOMACHTUBE
PAWINGAND
PAWINGS
PAWINIG
PSYLLIUM
PYLORIC
PYLORUS
RDD
REFLUX
REFLUXING
RESECTION
RETROFLEXION_OF_LARGE_COLON
ROLLING
SAND

SANDOUT
SANDOUT
SPAMODIC
SPASMODIC
SPASMODICS
SPASMOLYTIC
SPASMOLYTICS
SPASMOTIC
SPASMOTIC
PAWING
STANGULATION
STOMACH_TUBE
STOMACH_ULCER
STRANGULATED
STRANGULATING
TIPHLITIS
TORSION
TUBED
TWIST
TYMPANTIC
SQUAMOUS
TYPHLITIS
VOLVOLUS
VOLVO
VOLVULUS
WINDY_COLIC
WIND_COLIC
?COLIC.HR40,PULSE
?COLICKY
ADB_DISCOMFORT
CHOKE
CLINICAL_NOTE_SEVERE_PAIN_ON_ARRIVAL.
COLIC,PARAMETERS
COLICY.PASSED
COLOUR,?COLIC
CONSTIPATION
DISPLACEMENT/IMPACTION_FELT.STOMACH
DISTENDED_LARGE_INTESTINE
DROPPINGS_PASSED._LYING_DOWN
ENTEROCOLITIS
EPISODE_OF_ROLLING
EPSOM
FLANK_WATCHING
FLANK_WATCHING.
HAVE_BEEN_RESTLESS_AND_PAWING_BEDDING.
UNCOMFORTABLE._GUT_SOUNDS_-VE.
UNSETTLED_IN_BOX

COLIC EXCLUSION DICTIONARY
3RD_EYELID_REMOVAL_SQUAMOUS_CELL

ABAXIAL_DISPLACEMENT
APICAL_DISPLACEMENT
ARCH
ARCH_DISPLACEMENT
AREA_OF_DISPLACEMENT
ARYEPIGLOTTIC
ASSESSMENT_ON_ORAL_EXAM
ASSESSMENT_ROUTINE_MOUTH
ATLAS
BLEEDING_FROM_LIP
BONE
BONE-NO_DISPLACEMENT
BONES
BONE_+_DISPLACEMENT
BONE_-NO_DISPLACEMENT
BONE_LATERAL_TO_MIDLINE_NO_DISPLACEMENT
BONE_NO_DISPLACEMENT
BONE_NO_DISPLACEMENT
BONE_WITH_DISPLACEMENT
BONE_WITH_MILD_DISPLACEMENT
BOTH_HORSES_IN_GOOD_HEALTH
BSC_4/9_TEETH
BUCCAL
BUCCAL_DISPLACEMENT
BUCCAL_ULCERATION
BUCCAL_ULCERATION
C1
C1_DISPLACEMENT
C2
C3
C3_NO_DISPLACEMENT
CALCIFICATION_NO_DISPLACEMENT
CALCIFYING
CALCIUM_DEPOSITION_ON_LATERAL_VIEW._NO_DISPLACEMENT
CANINE_TEETH
CANNON_BONE_DERMATITIS_PRESENT
CAUDAL_HOOK
CHECKED_TEETH
CHECKED_TEETH_-_DUE_FOE_FLOAT
CHECK_HALINAS_TEETH
CHECK_POST_COLICE
CHECK_TEETH_CAMIEO
CHECK_TEETH_CASSIE
CHECK_TEETH_MARJI
CHECK_TEETH_±FLOAT
CHRONIC_HISTORY_OF_DIARRHEA
COFFIN
COLICKS
COLICS_WHEN_VACCS
COLIC_PREVENTION_PROGRAM

COMMINUTION
CONJUNCTIVITIS
CONSIDER_PERGOLIDE
CONTINUES_WIEGHT_LOSS
CORNEAL_INJURY
CORNEAL_ULCER
CORNER_INCISOR_DECIDUOUS_TEETH
COULD_BE_PRONE_TO_CHOKE_OR_IMPACTION_COLIC
COULD_NOT_VISUALIZE_EYE_TO_TAKE_MEASUREMENT
COXAE
CRANIAL_HOOK
CREPITUS
CREPITUS_OR_DISPLACEMENT
CUP_EPSOM_SALTS_WITH_16_OZ_H2O
DDSP
DECIDUOUS_TEETH
DEEP_INFUNDIBULUM
DENTAL/ORAL_RESULT
DENTAL_EXAM
DENTAL_HISTORY
DENTAL_NOTE
DEXAMETHAZONE_ANTIHISTAMINE_AND_PROCAINE_PENICILLIN
DEX_SUPPRESSION_TEST
DIARRHEA_THAT_COMES_AND_GOES_POSSIBLY_WITH_DIET
DIASTEMA
DISCHARGE_GENERAL
DISCHARGE_OTHER
DISCHARGE_PROCEDURE
DISPALCEMENT_OF_TEETH
DISPLACED_CAUDALLY_BEHIND_DECIDIOUS_TEETH
DISPLACED_DUE_TO_TRAUMA
DISPLACED_LABIALLY
DISPLACED_LINGUALLY
DISPLACED_TEETH
DISPLACEMENT._MILD_CARPITIS
DISPLACEMENT._SMALL_AVUSION
DISPLACEMENT._SPLINT
DISPLACEMENT_AND_BONE
DISPLACEMENT_AND_FRACTURE
DISPLACEMENT_AND_HEAT
DISPLACEMENT_AND_INCREASED_OSTEODENSITY
DISPLACEMENT_AND_THE_APPEARANCE_OF_THE_FRACTURE
DISPLACEMENT_AT_ARTICULATING
DISPLACEMENT_AT_JOINT
DISPLACEMENT_BETWEEN
DISPLACEMENT_BUT_CALCIFYING
DISPLACEMENT_DISTALLY
DISPLACEMENT_DISTAL_FEMUR
DISPLACEMENT_ELICITED_WITH_SCOPING
DISPLACEMENT_IN_HIP

DISPLACEMENT_IN_L
DISPLACEMENT_LEFT_SIDE_RIB
DISPLACEMENT_LEFT_SINUS
DISPLACEMENT_MEDIAL_TO_LATERAL
DISPLACEMENT_NOTED_OF_THE_FRACTURED
DISPLACEMENT_OF_408
DISPLACEMENT_OF_ARTICULAR
DISPLACEMENT_OF_ARYEPIGLOTTIC
DISPLACEMENT_OF_ATLAS
DISPLACEMENT_OF_BONES
DISPLACEMENT_OF_COFFIN
DISPLACEMENT_OF_DDFT
DISPLACEMENT_OF_DISTAL
DISPLACEMENT_OF_DORSAL_SACROILIAC
DISPLACEMENT_OF_DORSAL_SACROILLIAC
DISPLACEMENT_OF_DORSAL_SPINES
DISPLACEMENT_OF_ENTIRE_BODY_TO_LEFT
DISPLACEMENT_OF_EPIGLOTTIS
DISPLACEMENT_OF_FRACTURE
DISPLACEMENT_OF_G._TROCANTER
DISPLACEMENT_OF_GREATER_TROCANTER
DISPLACEMENT_OF_HAIRLINE
DISPLACEMENT_OF_HEAD
DISPLACEMENT_OF_HIP
DISPLACEMENT_OF_HIS_SOFT
DISPLACEMENT_OF_IRIS
DISPLACEMENT_OF_LH
DISPLACEMENT_OF_LH_GREATER_TROCANTER
DISPLACEMENT_OF_LUMBAR
DISPLACEMENT_OF_MANDIBLE
DISPLACEMENT_OF_MEDIAL
DISPLACEMENT_OF_MM_WITH_ROUGHING
DISPLACEMENT_OF_MT
DISPLACEMENT_OF_ONE_THORACIC
DISPLACEMENT_OF_P
DISPLACEMENT_OF_PELVIS
DISPLACEMENT_OF_PII
DISPLACEMENT_OF_PIII
DISPLACEMENT_OF_RH
DISPLACEMENT_OF_RIGHT_HIP
DISPLACEMENT_OF_RIGHT_SACRAL
DISPLACEMENT_OF_RT_HIND_HIP
DISPLACEMENT_OF_RT_HIP
DISPLACEMENT_OF_SACRAL
DISPLACEMENT_OF_SACRUM
DISPLACEMENT_OF_SCAPULA
DISPLACEMENT_OF_SCAPULAS
DISPLACEMENT_OF_SOFT_PALATE
DISPLACEMENT_OF_SOFT_PALATE
DISPLACEMENT_OF_STERNUM

DISPLACEMENT_OF_T
DISPLACEMENT_OF_T._ISCHEII
DISPLACEMENT_OF_T.COXAE
DISPLACEMENT_OF_TEETH
DISPLACEMENT_OF_THESE_VERTEBRAE
DISPLACEMENT_OF_THE_4TH
DISPLACEMENT_OF_THE_ARCH
DISPLACEMENT_OF_THE_BONE
DISPLACEMENT_OF_THE_COFFIN_BONE
DISPLACEMENT_OF_THE_CRANEAL
DISPLACEMENT_OF_THE_DISTAL
DISPLACEMENT_OF_THE_EPIGLOTTIS
DISPLACEMENT_OF_THE_EXTRAORBITAL
DISPLACEMENT_OF_THE_FRACTURE
DISPLACEMENT_OF_THE_FRAGMENT
DISPLACEMENT_OF_THE_FRAGMENTS
DISPLACEMENT_OF_THE_MAXILLA
DISPLACEMENT_OF_THE_MEDIAL
DISPLACEMENT_OF_THE_MEDIAL_SESAMOID_BONE
DISPLACEMENT_OF_THE_PALATOPHARYNGEAL
DISPLACEMENT_OF_THE_PATELLA
DISPLACEMENT_OF_THE_RIGHT_HIND
DISPLACEMENT_OF_THE_SACROILAC
DISPLACEMENT_OF_THE_SOFT_PALATE
DISPLACEMENT_OF_THE_SOFT_PALLATE
DISPLACEMENT_OF_THE_TUBER
DISPLACEMENT_OF_THORACIC
DISPLACEMENT_OF_TIP_OF_P
DISPLACEMENT_OF_TMJ
DISPLACEMENT_OF_TRACHEA
DISPLACEMENT_OF_WING
DISPLACEMENT_OF_WITHERS
DISPLACEMENT_OR_COMMINUTION
DISPLACEMENT_OR_SEPARATION
DISPLACEMENT_OR_WORSENING_OF_THE_FRACTURE
DISPLACEMENT_OVER_EPIGLOTTIS
DISPLACEMENT_REACTIVE_PASTERN
DISPLACEMENT_SEEN_ON_INITIAL_SCOPING
DISPLACEMENT_SENSITIVE_L_HORN
DISPLACEMENT_T
DISPLACEMENT_TO_GREATER_TROCANTER
DISPLACEMENT_TO_L
DISPLACEMENT_TO_PELVIS
DISPLACEMENT_TO_RH
DISPLACEMENT_TO_RIGHT
DISPLACEMENT_TO_TMJ
DISPLACEMENT_UNCHANGED._APPLIED_A_SAPA_SHOE
DIST._DISPLACEMENT
DISTAL_DISPLACEMENT
DKROUTINE_CALL

DONE_IN_FALL_07_10CC_BANAMINE_IV_-
_DUE_TO_PREVIOUS_COLIC_AFTER_VACCINES
DYSYMETRY
EDGES_OF_MAXILLARY_AND_LINGUAL_EDGES_OF_MANDIBULAR_CHEEK
_TEETH
EEE/WEE/VEE/TETANUS_VACCINATION
EIV-EHV-EWT_VACCINE_PRESTIGE
ELICITED_WITH_SCOPING
EPIGLOTTIS
EXAM_RE:_POOR_CONDITION
EXAM_TEXT_CONSULTATION_NUMBER_#113799_VISIT_#79594
EXAM_TEXT_TEETH
EXCESS_TARTAR_ON_LATERAL_ASPECT_OF_CRANIAL_MOLARS
EXOSTOSIS_NO_DISPLACEMENT
EXPECT_THAT_DENTAL_WORK_WILL_IMPROVE_HIS_ABILITY_TO_CHEW
_BUT_CANNOT_RULE_OUT
EXPLORATORY_ARTHROSCOPY
EXTRAORBITAL
F/R_BOTULISM
FECAL_SAND_SEDIMENTATION
FETLOCK_DISPLACEMENT
FIBRE
FIBRE_DISPLACEMENT
FIRE_-_LOSING_WEIGHT_-_PRIMARILY_MUSCLE_MASS
FIRST_HALF_VACCINES
FLOAT_ONLY_IF
FLOAT_TEETH
FOR_PURCHASE
FRACTURE
FRACTURE-NO_DISPLACEMENT
FRACTURED
FRACTURES
FRACTURES_OR_DISPLACEMENT
FRACTURE_AND_DISPLACEMENT
FRAGMENT_DISPLACEMENT
FRIDAY._WATCH_CLOSELY_FOR_SIGNS_OF_COLIC
FX
GASTRIC_RELIEF_FORMULA
GAVE_PROSTEN_TODAY_FOR_FIRST_BREEDING
GLUCOSE_TOLERANCE_TEST
HAIRLINE

HAS_HAD_DIARRHEA_FOR_3_DAYS_ALSO_PASSING_FORMED_MANURE
HAS_HISTORY_OF_COLIC_AFTER_VACCINATIONS
HAS_LOST_WEIGHT_FOR_THE_LAST_SEVERAL_MONTHS
HAS_NOT_APPEARED_PAINFUL_OR_COLICY
HAVE_BABYS_TEETH_FLOATED_WITHIN
HAVE_NASAL_DISCHARGE_AND_A_COUGH_THIS_A.M
HEALED_NO_DISPLACEMENT
HEALED_WITH_SOME_DISPLACEMENT

HEALING_WELL

HE_IS_NOT_COUGHING._HE_HAS_NOT_CHOKED_IN_THE_LAST_YEAR
HIGH_TEETH
HIND._DISPLACEMENT_SIGNIFICANT_TO_LEFT_OF_T.S
DISPLACEMENT_OF_THIS_VERTEBRA
HIP
HISTORICALLY_HAS_HAD_POOR_FEET
HISTORY:_HORSE_IS_TO_BE_ADOPTED
HISTORY:_INTERMITTENT_COUGH_AT_REST
HISTORY_OF_ADR
HOCK-NO_DISPLACEMENT
HOOF
HOOFTESTERS
HOOFWALL_RESECTION
HORSE_IS_DOING_FUNNY_THINGS_WITH_HIS_TONGUE
HUMEROUS_BELOW_JT._MIN._DISPLACEMENT
HYPERCEMENTOSIS
IF_NORMAL_TODAY_WATCH_CLOSELY_FOR_SIGNS_OF_COLIC
INSIDE-NO_DISPLACEMENT
INSTRUCTED_OWNER_TO_WATCH_CLOSELY_FOR_SIGNS_OF_COLIC
IN_ADDITION_TO_FLOAT_SHEATH_AND_TEETH
IN_GOOD_SHAPE._NO_SYMPTOMS_OF_COLIC
IRIS
ISCHEII
IS_STABLE_WITH_NO_COLLIC_EPISODES_THIS_WEEKEND
JOINT_SPACE_BUT_NO_DISPLACEMENT
L1
LAME
LAMENESS
LATERALLY_DISPLACED
LAVAGE_THORACIC
LEFT_SIDE_RIB
LEG_EXAM
LETHARGIC_AND_NOT_INTERESTED_IN_FEED_FOR_ONE_WEEK
LGA_NOTE:_WAVE_MOUTH
LIGAMENT
LINE_OR_DISPLACEMENT
LINGUAL
LINGUAL_DISPLACEMENT
LOOSE_STIFLE_SL._POS._SPAVIN_TEST
LOUNGE_IN_SAND
LUMBAR
MANDIBLE

MARE_OFF_FEED_AND_DEPRESSED_THIS_MORNING._NOT_COLLICY_OR_PAINF
UL
MARKED_ENAMEL_POINTS
MEDIAL_DISPLACEMENT_UNCHANGED
MENISCUS

MIDLINE_NO_DISPLACEMENT
MINIMAL_DISPLACEMENT
MODERATE_ENAMEL_POINTS
MOD_DISPLACEMENT
MONITORED_FOR_SIGNS_OF_ABDOMINAL_DISCOMFORT_OR_DRIBBLING_OF_URINE
MT4
MUCUS_REFLUX
NAVICULAR
ND_VISIT:_EIA_IN_BOX
NEGATIVE_ALL_FLEXIONS
NEVER_COLICED
NODULAR_NECROBIOSIS
NOTE_PPE
NOT_ACTING_COLCKY
NO_COLIC
NO_COLLIC_PAIN._BAG_LUNGS--NORMAL

NO_OBSERVED_COLIC_SIGNS_MARE_COMFORTABLE._NO_GRINDING_TEETH
NO_RABIES_GETS_COLLICKY_AFTER_RABIES
NO_REALLY_COLICKY_BUT_NOT_EATING_OR_DRINKING
NO_SIGNIFICANT_CHANGES_IN_DISPLACEMENT
NO_SIGNS_OF_COLIC
NO_SIGN_OF_COLIC
NO_STRANGLES_AND_CHECK_TEETH
NO_TOOTH_ABNORMALITIES_WERE_NOTED_ON_X-RAYS
OF_THE_4TH
OLD_FX_LINE_BARELY_VISIBLE_NO_DISPLACEMENT
ON_ORAL_EXAM_-_UPPER_INCISORS_SHORT
OPTHALMIC
ORAL_EXAM_WITH_SEDATION
ORBIT

OR_CONCERNS_MONITOR_FOR_SIGNS_OF_COLIC_INCREASED_LETHARGY
OSTEODENSITY
OWNER_COMPLAINS_HORSE_IS_NOT_EATING_WELL
OWNER_STOPPED_ME_IN_THE_BARN
P1
P2
P3
PALATE
PALATOPHARYNGEAL
PALLATE
PASTERN_DISPLACEMENT
PATELLA
PELVIS_DISPLACEMENT
PERIODONTAL_POCKETING
PERIODONTAL_POCKETS
PERIO_POCKETS
PFIZER_COLIC_PREVENTION

PHYSICAL_NOTE_DISPLACEMENT
PI
PII
PIII
PII_DISPLACEMENT
PLL_DISPLACEMENT
POSTERIOR_LATERAL_DISPLACEMENT
POWERFLOAT_TEETH
POWER_FLOAT
PPE_FINDINGS
PRE-LEASE_EXAMINATION
PRE-PURCHASE
PREPURCHASE
PREVENT_IMPACTION_DUE_TO_TEETH
PROCEDURE_WET_THE_GRAIN_DOWN_SLIGHTLY_BEFORE_GIVING._PA
TIENT_CHOKED_WHEN_SHE_WAS_2_YR_OLD
PUPIL_REACTIVE
QUARTER
QUARTER_WITH_DISPLACEMENT
RADIOGRAPHS_WERE_TAKEN_OF_FRONT_TEETH
RADIOGRAPH_CONFIRMED_DISPLACEMENT
REBREATHING_BAG
RECOMMEND_TEGUMET
RECTAL_TUBE_DISPLACEMENT
REFERRED_HER_FOR_MORE_EXTENSIVE_DENTAL_WORK
REMOVAL_OF_RETAINED
REPLACED_ICE_BOOTS
REPRODUCTION
RESPIRATORY_RESULT
REVOVE_STAPLES_FROM_COLIC_SURGERY
REXRAY_FOR_DISPLACEMENT
RIB_DISPLACEMENT
ROLLING_TOE
ROSTRAL_DISPLACEMENT
ROSTRAL_HOOK
ROUTINE_CALL
SACRAL
SACRALE
SACRALIS
SACRAL._DISPLACEMENT
SACRO-ILLIAC_DISPLACEMENT
SACROILIAC_DISPLACEMENT
SACROILLIAC
SACRUM
SACRUM_DISPLACEMENT
SAME_DAY_AS_THE_NAXCEL_AS_CAN_CAUSE_COLIITIS
SAND_ARENA
SAND_CANYON
SAND_SCHOOL
SCAPULA

SCAPULAS
SCC
SCIRRHOUS_CHORD_RESECTION,APPOINTMENTINFO
SCREWS
SCREWS_STILL_LOOK_GOOD_NO_DISPLACEMENT
SEEN_ON_INITIAL_SCOPING
SEQUESTRUM
SEQUESTRUM_DISPLACEMENT
SESAMOID
SEVERE_COLIC_SEPT_96
SEVERE_INJECTION_SITE_REACTIONS
SHARP_POINTS
SHE_MAY_NOT_CHEW_IT_ENOUGH_AND_COULD_BE_PRONE_TO_CHOK
E_OR_IMPACTION_COLIC
SHE_WAS_STILL_VERY_LAME._STILL_LAYING_DOWN_REGULARLY
SIDEBONE_NO_DISPLACEMENT
SINUS
SOAK_1-2X_A_DAY_IN_WARM_WATER_AND_EPSOM_SALTS
SOAK_FOOT_IN
SOAK_THE_FOOT_IN_WARM_WATER_AND_EPSOM_SALTS
SOAK_W/_EPSOM_SALTS
SOUNDNESS
SPINE
SPINES
SPLINT
SPLINTER
SPRING_VACCINES
SQUAMOUS_CELL_CARCINOMA
SQUAMOUS_CELL_CARCINOMA-LOWER
STERNAL_DISPLACEMENT
STIFLE_LATERLA_DISPLACEMENT
SUPERNUMERARY
SWAYBACK_CONFORMATION
SWELLING_UNDER_JAW_AND_IN_EYES
T.S
T._S
T15
T16
T17
T18
TEETH_-_NO_HOOKS/POINTS
TEETH_-_NO_HOOKS/POINTS
TEETH_-_WAVE_HAS_NOT_RETURNED
TEETH_ARE_DUE_TO_BE_DONE_SMALL_REAR_RAMPS
TEETH_BEEN_FLOATED_NO_COLIC_SURGERY_SCAR
TEETH_CHEEKS_&_BARS
TEETH_COULD_DO_WITH_A_FLOAT_-_SCHEDULED_WITH
TEETH_FLOAT
TEETH_HAVE_MILD_SHARP_POINTS
TEETH_HAVE_SHARP_POINTS

TEETH_OK._LOW_WEIGHT_-
_RECOMMEND_TRYING_DIFFERENT_SENIOR_FEED_OR_FAT_SUPPLEMEN
T
TEETH_VERY_SHARP
TENDENCY_TO_COLIC_EITHER_THE_EVENING_OR_MORNING_AFTER_FL
OATING_HIS_TEETH
TENDS_TO_COLLIC_AFTER_SEDATION
THERE_IS_AN_ABNORMAL_GROWTH_ON_THE_LEFT_SIDE_OF_THE_BAS
E_OF_THE_TAIL
THERE_WAS_NO_FEED_IMPACTION
THORACIC
THORACIC_DISPLACEMENT
THORACIC_U/S
THORACIC_ULTRASOUND
THROW_HIS_HEAD_UP_WHEN_RIDING
TMJ
TOMORROW._WATCH_CLOSELY_FOR_SIGNS_OF_COLIC
TOOTH_ROOTH_ABSCESS
TRACE_AMOUNTS_OF_SAND_IN_HIS_FECES._THIS_WOULD_BE_CONSIDE
RED_NORMAL
TRAINER_REPORTS_LETHARGY_UNDER_SADDLE
TROCANTER
TUBED_WITH_COLOSTRUM
TUBER
TUBER-COXAE._NO_DISPLACEMENT
VERTEBRA
VERTEBRAE
VERTEBRAE_DISPLACEMENT
VERTEBRAL
VERTEBRAL_DISPLACEMENT
TROCANTER_DISPLACEMENT
VERY_DEPRESSED_NO_SIGNS_OF_ABDOMINAL_PAIN
VERY_STIFF_AT_A_JOG_GOING_TO_THE_RIGHT
WADDING_HAY_IN_MOUTH._NO_TEMP_NO_COLIC_SYMPTOMS
WANTED_TO_EAT_NO_SIGNS_OF_COLLIC
WAS_PREVIOUSLY_TREATED_FOR_INTERNAL_PARASITES_AND_SAND
WEIGHT_IS_IMPROVED
WELLNESS
WITHERS
WITHERS_DISPLACEMENT
WING_NO_DISPLACEMENT
X-RAY_HEAD
YEARS_AGO_COLICED
WOLF_TEETH

COLIC_RE-INCLUSION DICTIONARY
FLUSHED ABDOMEN AND REFLUXED HORSE AGIAN THIS PM
FLUSHED ABDOMEN AND TUBED HORSE
BUSCOPAN. POSS SPASMODIC COLIC
MILD COLIC. NORMAL DROPPINGS

COLON NOT DISTENDED SO ADVISE PROB MILDLY DISPLACED
CLICKING TODAY
COLICKING UNTIL MORNING
UNTIL I ARRIVED

ORTHOPAEDIC INCLUSION DICTIONARY
ABAX
ABAXIAL
ABAXIAL_SESAMOID
ABAXIALLY
ABDUCTION
ABNB
ABSNB
ADEQ
ADEQM
ADEQUAN
ADEQUIM
ADEQUON
ANNULAR
ANTEBRACH
ANTEBRACHIA
ANTEBRACHIAL
ANTEBRACHIO
ANTEBRACHIOCARPAL
ANTEBRACHIUM
ANTERACHIUM
ANTIBRACHIUM
APONEUROSES
APONEUROSIS
ARTHIRITIS
ARTHRITIC
ARTHRITIS
ARTHRITUS
ARTHROCENTESIS
ARTHROPATHY
ARTICULAR
ARTICULATION
ASNB
BACK_PAIN
BAPTEN
BAREFOOT
BICIPITAL
BLOCKABLE
BLOCKED
BOW-TENDON
BRACHIAL
BRACHIALIS
BRACHIO
BURSAL
BURSITIS

BURSOSCOPY
BUTTOCK
CALCANEAL
CALCANEOUS
CALCANEUS
CANNON
CANNONS
CANON
CANONBONE
CANTER
CARPAL
CARPII
CARPIS
CARPITIS
CARPLE
CARPLUS
CARPOMETACARPAL
CARPUS
CARTILAGE
CARTOPHEN
CARTROPHEN
CELES
CHIRO
CHIROPRACTOR
CIRCUMDUCTION
COFFIN
COLLATERAL
CONDYLAR
CONDYLE
CONTRACTURE
CONTROPROTEC
CORONARY
CORONARY_BAND
CORONERY
CORONET
CORONITIS
CORONORY
COXA
COXAE
CREPITUS
CRUCIATE
CRURAL
CYST
DDFT
DEFORMITIES
DELTOID
DERMOBIAN
DERMOBION
DESMITIS
DESMITITIS

DESMITITS
DESMITS
DESMITUS
DESMONTOMY
DESMOPATHY
DESMOPLASTY
DESMOTAMY
DESMOTOMY
DFTS
DIAPHYSIS
DIGTAL
DIG_SHEATH_EFFUSION
DJD
DYNACAST
EABAXIAL
EFFUSSION
ELBOW
ENDOSTEAL
ENTESIOPHYTRE
ENTHESIOPHYTE
ENTHESOPHYTE
ENTHESYOPHYTE
ENTHRESIOPHYTES
EPAXIAL
EPIPHYSIS
EPIPHYSITIS
ESW
EXAMINELAMEHORS
EXLAMEHORSE
EXLAMEPONYADVIS
EXOSTOSES
EXOTOSIS
EXPAXIAL
EXTENSOR
FEETLOCK
FEET_BALANCE
FELOCK
FELTOCK
FEMERAL
FEMEROPATELLA
FEMEROPATELLAR
FEMEROTIBIAL
FEMORAL
FEMORIS
FEMORO
FEMOROPATELLA
FEMOROPATELLAR
FEMOROTIBIAL
FETLCOK
FETLCOKS

FETLLOCK
FETLLOCKS
FETLOCK
FETLOCKS
FETLOK
FETOCK
FIBIA
FIBULAR
FLEXING
FLEXION
FLEXIONS
FLEXON
FLEXOR
FLEXORA
FLEXORIA
FLEXORS
FLEXURAL
FOOT
FOOTSORE
FOREARM
FOREFEET
FORELIMB
FOUNDER
FOUNDERD
FOUNDERED
FOUNDEREED
FOUNDERING
FOUNDERS
FOUNDERT
FOUNDING
FOUNDNO
FOUNDRED
FOUNDS
FOUNF
FOUNR
FRACTURE
FRACTURED
FRACTURES
FRAGMENT
FRAGMENTS
FRCATURE
FROG
FROGS
FX
GASKIN
GASTROC
GASTROCNEMIUS
GLENOID
GLYCOSAMMINOGLYCAN
HAEMARTHROSIS

HAMSTRING
HANDWALK
HEARTBAR
HEARTBARS
HEART_BAR
HEART_BARS
HEEL
HEELS
HEMARTHROSIS
HIGH_PLANTAR
HINDLEG
HINDLEGS
HINDQUARTERS
HIP
HOCK
HOCKS
HOOF
HOOFTESTER
HOOFTESTERS
HOOFWALL
HYALARTIN
HYALASE
HYALOVET
HYALURONATE
HYALURONIC
HYALURONIC_ACID
HYGROMA
HYLARTIL
HYLARTIN
HYONATE
HYPEREXTENSION
HYVISC
HYVISK
ILEAC
ILEO
ILIAC
ILIAL
ILLIAC
ILLIUM
INCREASED_DP
INT/ART
INTERCARPAL
INTERPHALANGEAL
INTERTARSAL
INTRAARTICULAR
INTRASYNOVIAL
IRAP
IRAPP
ISCHIUM
IVRP

JOINT
JOINTS
J_BLOCK
KERATOMA
KNEE
KNEES
LAEMNESS
LAIMINITIS
LAMANESS
LAMANITIC
LAMANITIS
LAMANITITIS
LAMANTIS
LAME
LAMEESS
LAMEINITIS
LAMENES
LAMENESS
LAMENESSS
LAMENITIS
LAMENITISIS
LAMENITITS
LAMENSS
LAMENTSS
LAMESNESS
LAMESS
LAMIINITS
LAMIITIS
LAMINAITIS
LAMINAL
LAMINECTOMY
LAMINIIS
LAMINITIS
LAMNEESS
LAMNESS
LANMENESS
LAT_PLANATR
LEGEND
LFEXION
LFORE
LHIND
LHOCK
LIGAMENTOUS
LIGAMENTS
LONGISSIMUS
LORDOSIS
LUMBAR
LUMBARS
LUMBOSACRAL
LUXATION

MALEOLUS
MALLEOLUS
MCII
MCIII
MEADIAL
MEDIALL
MEDIOLATERAL
MEDRONE
MENISCAL
MENISCUS
MESOTENDON
METACARPAL
METACARPOPHALANGEAL
METACARPUS
METATARSAL
METATARSUS
MIDBRACHIUM
MIDCARPAL
MIDSOLE
MRI
MTIII
MTPJ
NAVIC
NAVICULAR
NAVICULARS
NAVILOX
NERVE-BLOCK
NERVEBLOCK
NERVE_BLOCK
NERVE_BLOCK
NEUROPATHY
N_BLOCK
N_BLOCK
OCD
OLECRANON
ORTHO
ORTHOPAEDIC
OSTECTIS
OSTEITIS
OSTEOARTHERITIS
OSTEOARTHRITIC
OSTEOARTHRITIS
OSTEOCHONDRAL
OSTEOCHONDROMA
OSTEOCHONDROSIS
OSTEODENSITY
OSTEOLYSIS
OSTEOMYELITIS
OSTEOPATH
OSTEOPHYTE

OSTEOPHYTES
OSTEOPHYTOSIS
OSTITIS
OVERGROWING
OVERREACHED
OVERRIDING
P3_DISPLACEMENT
PALMAR
PALMAR-DIGITAL
PALMARDIGITAL
PALMARODISTAL
PALMAROLATERAL
PALMAROMEDIAL
PALMAR_N.B
PALMAR_NB
PALMER
PARALUMBAR
PARATENDON
PARATENON
PASTERN
PASTERNS
PATELA
PATELLA
PATELLAE
PATELLAR
PATELLAS
PDNB
PEDAL
PELVIC
PELVIS
PELVIS_DISPLACEMENT
PERIARTICULAR
PERILIGAMENTOUS
PERINEURAL
PERIOSTEAL
PERIOSTEUM
PERIOSTITIS
PERITARSAL
PERITENDINOUS
PERITENDONOUS
PERONEUS
PEROREUS
PHALANGEAL
PHALANX
PHYSIOTHERAPIST
PHYSITIS
PI
PII
PIII
PLANTAR

PLANTARO
PLANTAROLATERAL
PLANTAROMEDIAL
POSTURAL
POTTERING
POULTICE
PUNCTURE
PUS_IN_THE_FOOT
QUADRICEPS
RADIAL
RADIALIS
RADIUS
REINJURED
REINJURY
RETINACULUM
RIDGE
RING-BONE
RINGBONE
RING_BONE
ROSTROVENTRAL
ROTAION
ROTATES
ROTATING
ROTATION
ROTATIONAL
ROTATIONS
RUMP
SACRAL
SACRALE
SACRALIS
SACROILEAC
SACROILIAC
SACROILIACS
SACROILIAE
SACROILLIAC
SACRUM
SAGGITAL
SAGITAL
SAGITTAL
SANDCRACK
SCAPULA
SCAPULAS
SCOUT
SDFT
SEMIMEMBRANOSUS
SEMITENDINOSUS
SEMITENDINOUS
SEQUESTRAE
SEQUESTRIUM
SEQUESTRUM

SESAMOID
SESAMOIDEAN
SESAMOIDIAN
SESAMOIDITIS
SESAMOIDS
SESMOID
SHOCK-WAVE
SHOCKWAVE
SHOCKWAVED
SHOLDER
SHOULDER
SKELETAL
SKELETON
SOLAR
SOLESUPP
SPAVIN
SPAVINS
SPINE
SPINOUS
SPLINT
SPLINTS
SPONDYLOSIS
SPRAIN
STERNEBRA
STIFEL
STIFFEN
STIFFENED
STIFFENING
STIFFENS
STIFFF
STIFFLE
STIFLE
STIFLES
STIFNESS
STILES
STILFE
STRAIN
STRINGHALT
SUB-LUXATION
SUB-SOLEAR
SUBCARPAL
SUBCHONDRAL
SUBLUX
SUBLUXATING
SUBLUXATION
SUBSOLAR
SUBSOLEAR
SUBTARSAL
SUPENSORY
SUPRASCAPULAR

SUPRASPINOUS
SUSPENSORIES
SUSPENSORY
SUSPENSORYS
SYNOVIA
SYNOVIAL
SYNOVILA
SYNOVIOCENTESIS
SYNOVIOCOELE
SYNOVITIS
SYNOVIUM
TARSAL
TARSO
TARSOCRUAL
TARSOCRURAL
TARSOCURAL
TARSOCURUAL
TARSOMETATARSAL
TECHNOVIT
TENDINITIS
TENDON
TENDONITIS
TENDONOUS
TENDONS
TENDONSHEATH
TENDON_SCAN
TENDON_SHEATH
TENOSINOVITIS
TENOSYNOITIS
TENOSYNOVITIS
TESTERS
THORACOLUMBAR
THORACOLUMBER
THORN
THOROUGHPIN
TIBIA
TIBIAL
TIBIOTARSAL
TILDREN
TILUDRONATE
TMJ
TMT
TMTINJECTION
TMTS
TRAPEZIUS
TROCHANTER
TROCHLEA
TROTED
TROTING
TROTUP

TUBER
TUBERCLE
TUBERCOXEA
ULNAR
VALGUS
VERSATRON
VERTEBRAL
VETCAST
VOLUVEN
WEIGHBEARING
WEIGHTBEAR
WEIGHTBEARING
WEIGHTSHIFTING
WHITELINE
WITHERS
_OA_
OBLIQUES

ORTHOPAEDIC EXCLUSION DICTIONARY
AURICOLOPALPEBRAL_NERVE_BLOCK
BLOCK_EYE
CANTOS_OF_THE_EYE
CAUTERY_OPTHALMIC_NERVE_BLOCK
CEVA_OPEN_-_FARRIER_WORK
DORM_GELS_FOR_DONKEYS_AND_FARRIER_WORK
EYE_EXAM
EYE_NERVE_BLOCK
EYE_TUMOR
EYE_ULCER
FARRIER_WORK_-_OUTSIDE_FARRIER
FARRIER_WORK_APPOINTMENTINFO
FARRIER_WORK_[DR_DOUGLAS_LANGER
FROG_SUPPORT_IN_PLACE
FRONTAL_NERVE_BLOCK
IF_BO_BECAME_LAME_SORE_SWOLLEN
IS_A_NICE_FREE_MOVER_IN_ALL_4_LIMBS
NORMAL_FARRIER_WORK
NOTE_FARRIER_WORK_TODAY
NO_EVIDENCE_OF_LAMENESS
OPHTHALMIC_EXAM
OPHTHALMIC_NERVE_BLOCK
PALPEBRAL_NERVE_BLOCK
PALP_TEXT_RESULT_SARCOID._LIKE_2_CM._LESION_ON_PASTERN
REDUCED_DORM/TORB_-_.4/.4CC_-_FARRIER
REDUCED_DORM/TORB_-_.5/.5CC_-_FARRIER
REDUCED_DORMOSEDAN_-_.35CC_-_FARRIER
REDUCED_DORMOSEDAN_-_.4CC_-_FARRIER
REDUCED_DORMOSEDAN_-_.5CC_-_FARRIER
REGULAR_DORMOSEDAN_.3CC_-_FARRIER
RIGHT_EYE_HAS_MILD_AMOUNT_OF_WHITE_DISCHARGE

SHOE_CYCLE
TRANQ_FOR_FARRIER
SUPRAORBITAL_NERVE_BLOCK
ABRASIONS_ON_WITHERS
BRIEF_WITHERS_WNL
AUROCOLOPALPEBRAL_NERVE_BLOCK
CUT_ON_WITHERS
DISCHARGE_OUT_OF_WITHERS
EXCORIATED_L_WITHERS
FISTULOUS_WITHERS
FRONTAL_NERVE_BLOCK
INFECTED_WITHERS
INFECTION_OF_WITHERS
LLIAN_BIOPSY
LUMP_ON_WITHERS
OCULAR_NERVE_BLOCK
OVER_WITHERS
PALPEBRAL_NERVE_BLOCK
PRESSURE_SORE_WITHERS
PRURITIC_AREA_ALL_OF_NECK
PYODERMA_AROUND_WITHERS
RUB_ON_WITHERS
SOME_ON_WITHERS
STAPLES_REMOVED_FROM_SURGICAL_WITHERS
SUPRAORBITAL_NERVE_BLOCK
WHITE_HAIR_@_WITHERS
WITHERS_-_MOIST_DERMATITIS
WITHERS_INFECTION
WITHERS_WOUND_HEALING
WOUNDS_ON_WITHERS
WOUND_DORSAL_WITHERS

Appendix 5

ANTIMICROBIALS INCLUSION DICTIONARY

A/BS
ABS
ABX
AB'S
ABS
AGRIMYCIN
AIVLOSIN
ALAMYCIN
AMIKACIN
AMIKIN
AMOXICILLIN
AMOXINSOL
AMOXLA
AMOXYCILLIN
AMPEQUINE
AMPICILLIN
AMPROLIUM
ANIMEDAZON
ANITBIOTICS
ANTBX
ANTI B
ANTIB
ANTIBACTERIAL
ANTIBIOSIS
ANTIBIOTCS
ANTIBIOTIC
ANTIBIOTICS
ANTIBOTICS
ANTIOBIOTICS
ANTIROBE
ANTI_BIOTICS
APRALAN
APRAMYCIN
AQUATET
ATBS
AUREOMYCIN
AUROFAC
AZIMYCIN
AZITHROMYCIN
BACTRIM
BANTIBIOTIC
BAYCOX
BAYRIL
BAYT
BAYTIL
BAYTRIL

BAYTRIL
BAYTRILL
BENZATHINE
BENZYLPENICILLIN
BETAMOX
BIMECTIN
BIMOTRIM
BINIXIN
BIOMYCIN
BORGAL
CEFA
CEFALAC
CEFALACK
CEFALAK
CEFA-LAK
CEFALEXIN
CEFAPIRIN
CEFAZOLIN
CEFELAK
CEFENIL
CEFGUARD
CEFITOFUR
CEFOTAXIME
CEFOVECIN
CEFPODOXIME
CEFQUINOME
CEFTAZIDIME
CEFTIFLEX
CEFTIOCYL
CEFTIOFUR
CEFTRIAXONE
CEFUROXIME
CEPHAGUARD
CEPHALAC
CEPHALAK
CEPHALEXIN
CEPHALOTIN
CEPHAPIRIN
CEPHE
CEPOREX
CEPRAVIN
CEVAXEL
CHLO
CHLO
CHLOR
CHLORAM
CHLORAMPHEN
CHLORAMPHENICAL
CHLORAMPHENICOL
CHLORBIOTIC

CHLOREPHENICAL
CHLOROMPHENICOL
CHLOROMYCETIN
CHLOROPHENICOL
CHLORSOL
CHLORTETRACYCLINE
CHLPO
CILASTATIN
CILOXAN
CIPROFLOXACIN
CLAMOXYL
CLARITHROMYCIN
CLAVAMOX
CLAVUCILL
CLAVULANIC_ACID
CLINDACYL
CLINDAMYCIN
CLORTETRACYCLINE
CLOXACILLIN
CLYNDAMYCIN
COBACTAN
COBACTIN
CRYSTAPEN
CYCLOSOL
DAILYTMPS
DELVOPRIM
DEPCILLIN
DEPO
DEPOCCILIN
DEPOCIILIN
DEPOCILIN
DEPOCILL
DEPOCILLIN
DEPOCILLING
DEPOCILLLIN
DEPOCILLON
DEPOCOLLIN
DEPOMYB
DEPOMYCIN
DEPOMYCN
DEPOMYJ
DFOXYCYCLINE
DIMYCIN
DOXICYLINE
DOXY
DOXYC
DOXYCYCLENE
DOXYCYCLINE
DOXYCYLCINE
DOXYCYLINE

DOXYSEPTIN
DRAXXIN
DTRIMPASTE
DUPGRAN
DUPHACILLIN
DUPHACYCLINE
DUPHAMOX
DUPHAPEN
DUPHAPEN
DUPHATRIM
DUPHATRIM
DUPLOCILIN
DUPLOCILLIN
ENGEMYC
ENGEMYCIN
ENGYMYCIN
ENROFLAXACIN
ENROFLAXCECIN
ENROFLAXCIN
ENROFLOXACIN
ENROX
ENROXIL
EQUIFUR
EQUITRIM
EQUITRS
ERYTHROCIN
ERYTHROMICIN
ERYTHROMYCIN
ETHICILIN
EXCENEL
EXENEL
FENOFLOX
FLOROCOL
FLO_CILLIN
FLOCILLIN
FLORFENICOL
FLUORQUINOLONES
FORCYL
FORTAZ
FRAMOMYCIN
GENOTCIN
GENT
GENTA
GENTACIN
GENTAJECT
GENTAMAX
GENTAMICIN
GENTAMYCIN
GENTICIN
GENTOCIN

GSULPHADIAZINE
HEXASOL
HYDRODOXX
IMIPENEM
KARIDOX
KLARICID
KPEN
LINCOCIN
LIQUAMYCIN
MARBIFLOX
MARBOCYL
MARBOFLOXACIN
MARBOX
METORNIDAYOLE
METRO
METRONDIAZOLE
METRONEX
METRONIDAOLE
METRONIDAZLOE
METRONIDAZOL
METRONIDAZOLE
METRONIDAZOLEL
METRONIDOZOLE
METRONITAZOLE
MICOTIL
MILIMYCIN
MINOCYCLINE
MINOCYLINE
MMETRONIDAZOLE
MOXIFLOXACIN
MOXYFLOXACIN
MTRIMETHOPRIM
NAXC
NAXCEL
NAXCELL
NAXEL
NAXIV
NEOMYACIN
NEOMYCIN
NEOPEN
NEOPEN
NISAMOX
NITROFURANTOIN
NORADINE
NORDINE
NOROCILLIN
NOROCILLINLAINJ
NOROCLAV
NORODINE
NOROG

NOROTYL
NUFLOR
OPTICLOX
ORBENIN
ORNICURE
OXACILLIN
OXYMYCIN
OXYMYCINE
OXYTET
OXYTETRA
OXYTETRACYCLIN
OXYTETRACYCLINE
OXYTETRIN
OXYTETS
OXYTOCIN
OXYTRACYCLINE
PEN&STREP
PENCILLIN
PENICILLI
PENICILLIN
PENICLLIN
PENLA
PENSTR1
PENSTREP
PETERCILLIN
PIPERACILLIN
POWERFLOX
PROCAINE
PULMODOX
READYCEF
RIFADIN
RIFAMPICIN
RIFAMPIN
RONAXAN
SMZ
SMZS
SMZT
SOLUDOX
SPECTAM
SPECTIN
SPECTINOMICIN
SPECTINOMYCIN
SPECTOGARD
SPECTRAMAST
SPECTROMAST
STOMORGYL
STREPTOMYCIN
STREPTOPEN
SULFADIAZINE
SULFADIAZONE

SULFADOXINE
SULFAMETHOXAZOLE
SULFATRIM
SULPHADIAZINE
SULPHADOXINE
SULPHAMETHOXAZOLE
SULPHANILAMIDE
SULPHATHIAZOLE
SULPHONAMIDE
SULPHONAMIDES
SYNULOX
SYNUTRIM
TAZICEF
TAZOBACTAM
TAZOBACTUM
TERRAMYCIN
TETRACYCLINE
TETRAMIN
THIOSTREPTON
TIACIL
TICARCILLIN
TIMENTIN
TMPS
TMS
TRIBISSEN
TRIBRESSEN
TRIBRESSIN
TRIBRISSAN
TRIBRISSEN
TRIBRISSON
TRICARCILLIN
TRIMEDIAZINE
TRIMETHOPRIM
TRIMETOPRIM
TRIMS
TRIVETRIN
TUCAPRIM
TUCAPRIME
TUCCOPRIM
TUCOPRIM
TULATHROMYCIN
TULOSIN
TYLAN
TYLAN
TYLUVET
UBIFLOX
UBROLEXIN
ULTRAPEN
ULTRAPEN
UNIPRIM

VANCOMYCIN
VETRIMYCIN
VIGAMOX
ZIMYCIN
ZITHROMAX
ZYMICIN

DIARRHOEA INCLUSION DICTIONARY
ABX_INDUCE
ABX_INDUCED
ABX-INDUCED
ANTIBIOTIC_INDUCED
ANTIBIOTIC-INDUCED
ANTIMICROBIAL_INDUCED
ANTIMICROBIAL-INDUCED
BIOSPONGE
CHARCOAL
COCCIDIA
CODEINE
COLITIS
COWPAT
COW_PATCH
COW_PIE
COW-PATCH
COW-PIE
CYATH
CYATHOSTOME
CYATHOSTOMES
CYATHOSTOMIASIS
D+
D++
D++.
D+++
DARRHOEA
DIAAHORREA
DIAARRHOEIC
DIAFFHOEA
DIAHOREEA
DIAHORREA
DIAOHHREA
DIAOHROEA
DIAORRHEA
DIARHAEA
DIARHEA
DIARHHOEA
DIARHOEA
DIARHOEE
DIARHORRA
DIARHORREA
DIAROEAH

DIAROHEEA
DIAROHHEA
DIAROHOEA
DIAROOHEA
DIARR
DIARRAHEA
DIARREHA
DIARREHEA
DIARRH
DIARRHAE
DIARRHAEA
DIARRHEA
DIARRHEOA
DIARRHIAEA
DIARRHOA
DIARRHOAE
DIARRHOE
DIARRHOEA
DIARRHOEAL
DIARRHOEAPAINFUL
DIARRHOEIA
DIARRHOEIC
DIARRHOERA
DIARRHOES
DIARRHORA
DIARRHOREA
DIARRHOSIN
DIARROEA
DIARROEAH
DIARROHEA
DIARROHEOA
DIARROHOEA
DIARRORHAEA
DIARRORHEA
DIOROHEA
DIORREAH
DIORRHAEA
DIORRHEOA
DIORRHOEA
ENTEROCOLITIS
FORGASTRIN
FROM_ANTIMICROBIAL_TREATMENT
FROM_NSAID_TREATMENT
KAOGEL
KOLIN
LOOSE_FAECES
LOOSE_FECES
LOOSE_MANURE
LOOSE_TOOLS
NSAID_INDUCED

NSAID-INDUCED
NSAIDS_INDUCED
NSAIDS-INDUCED
PEPTO
PEPTOBISMOL
POST_NSAID_ADMINISTRATION
REDWORM
REDWORMS
SALMONELLA
SCOUR
SCOURING
SOFT_FAECES
SOFT_FECES
SOFT_MANURE
SOFT_TOOLS
THICK_COLON
THICKENED_COLON
WATERY_FAECES
WATERY_FECES
WATERY_MANURE
WATERY_TOOLS
FROM_NSAID_TREATMENT
NSAID_INDUCED
NSAID-INDUCED
NSAIDS_INDUCED
NSAIDS-INDUCED
POST_NSAID_ADMINISTRATION
RD_COLITIS
RDC_EPISODE
RIGHT_DORSAL,COLITIS
RIGHT_DORSAL_COLITIS
RIGHT_DORSAL_COLITIS,
RIGHT_DORSAL_COLITS,

DIARRHOEA EXCLUSION DICTIONARY
ALSO_MONITOR_FOR_DIARRHEA_AS_WELL
BREAKS_WITH_DIARRHEA_HIS_PROGNOSIS_GOES_DOWN
BUT_TAPEWROM_AND_TX_LARVAL_CYATH
CALL_IF_ANY_DIARRHEA
CALL_IF_COLT_GETS_DIARRHEA
CALL_IF_DIARRHEA_OR_DECREASED_MANURE
CALL_IF_DIARRHEA_OR_OTHER_CONCERNS
CALL_IF_HE_HAS_DIARRHEA
CALL_IF_HORSE_HAS_DIARRHEA
CALL_IF_HORSES_HAVE_DIARRHEA
CALL_IF_PAIN_RETURNS_APPETITE_DROPS_FEVER_DIARRHEA
CALL_IF_SHE_HAS_DIARRHEA
CAN_COME_AFTER_THE_DIARRHEA_PHASE
CHARCOAL_DRESS
CHARCOAL_DRESSING

CHARCOAL_HAS_BEEN_PREVENTING_MORE_EXCESS_PROUD
CHARCOAL_STUCK_ON_WITH_E-BAND
CONTINUE_MONITORING_FOR_SIGNS_OF_COLIC_DIARRHEA
CSSOTP_IF_DIARRH
CSSTOP_IF_DIARRHEA
DEVELOPS_DIARRHEA
DEVELOPS_LOOSE_MANURE_AND_CALL_US
DEVELOPS_SOFT_MANURE
DIARRHEA_DEVELOPS_STOPS_EATING
DIARRHEA_DISCONTINUE
DIARRHEA_GONE
DIARRHEA_HAS_NOT_COME_BACK
DIARRHEA_OR_FOR_ANY_CONCERNS
DIARRHEA_OR_SIGN_OF_COLIC
DID_NOT_SEE_ANY_MORE_DIARRHEA
DISCONTINUE_USE_IMMEDIATELY_IF_SMOLE_SHOWS_SIGNS_OF_DIAR
RHEA
DISCUSSED_WITH_OWNER_POSSIBILITY_OF_DIARRHEA
DISPENSED_FOR_DIARRHEA_IN_FUTURE
DOES_NOT_HAVE_A_HX_OF_DIARRHEA
EGG_COUNT_-VE
EPG_-_NEEDS_WORMED_ADVISE_TO_USE_BIMECTIN
FROM_THE_PROBLEM_LIST_WE_CAN_REMOVE_FEVER_AND_DIARRHE
A
HAS_NOT_HAD_ANY_HISTORY_OF_DAIRRHEA
HAS_NOT_HAD_DIARRHEA_SINCE
HAS_NOT_HAD_WATERY_DIARRHEA
HE_MAY_BECOME_OVERLY_DROWSY_OR_DEVELOP_DIARRHEA
I.E._WATCH_FOR_DAIRRHEA
IE_RECURRENCE_DIARRHEA_OR_SIGN_OF
IF DIARRHEA DEVELOPSN
IF DIARRHEA IS OBSERVED
IF DIARRHEA IS SEEN
IF DIARRHEA SEEN
IF DIARRHEA SHOULD DEVELOP
IF DIARRHEA STARTS
IF_ANY_DIARRHEA_PLEASE_CALL
IF_ANY_SIGNS_OF_ULCERS_OR_DIARRHEA_PRESENT
IF_CONCERNED_CAN_CHECK_DUNG_SAMPLE
IF_DIARRHEA_BEGINS_AGAIN
IF_DIARRHEA_DEVELOPS
IF_DIARRHEA_INAPPETENCE_ETC
IF_DIARRHEA_OCCURS
IF_FOAL_GETS_DIARRHEA
IF_HE_BREAKS_WITH_DIARRHEA
IF_LOOSE_MANURE_DEVELIOPS
IF_LOOSE_MANURE_DEVELOPS
IF_THE_HORSE_DEVELOPS_DIARRHEA
IF_YOU_NOTCIE_ANY_DIARRHEA
IF_YOU_NOTICE_DIARRHEA

IF_YOU_NOTICE_LOOSE_MANURE
IF_YOU_NOTICE_SOFT_MANURE
KGSTOP_IF_DIARRHEA
LARVAL_COUNT_NEGATIVE
MAY_CAUSE_DIARRHEA
MONITOR_BOTH_HORSES_FOR_A_DECREASE_IN_APPETITE_DEPRESSI
ON_A_FEVER_ABOVE_102.5_DIARRHEA
MONITOR_DAILY_FOR_DAIRRHEA
MONITOR_FOR_ANY_DIARRHEA
MONITOR_FOR_COLIC_DIARRHEA_SORENESS_IN_FEET
MONITOR_FOR_COUGH_NASAL_DISCHAGE_DIARRHEA
MONITOR_FOR_DIARRHEA
MONITOR_FOR_INCREASED_FOOT_SORENESS_DIARRHEA
NOT_PASSED_ANY_DROPPINGS_SINCE_YESTERDAY
NO_COUGH_SNEEZE_DIARRHEA
NO_COUGHING_SNEEZING_DIARRHEA
NO_CYATH_TX._ADVISE_EQUESTS_FOR_MOST_PAN_GAURD
NO_D+
NO_D++
NO_DIARRHEA
NO_EVIDENCE_OF_DIARRHEA
NO_FEVER_OR_DIARRHEA
NO_HX_OF_DIARRHEA
NO_LOOSE_MANURE
NO_MANURE_DIARRHEA
NO_MORE_DIARRHEA_NOTED
NO_MORE_DIARRHEA_PRESENT
NO_NEW_MANURE_DIARRHEA
NO_NEW_MANURE_OR_DIARRHEA
NO_RECURRENCE_OF_THE_DIARRHEA
NO_SIGN_OF_DIARRHEA
NO_SIGNS_COLIC_OR_RECURRENT_DIARRHEA
NO_SIGNS_OF_ANY_DIARRHEA
NO_SIGNS_OF_COLIC_OR_DIARRHEA
NO_SIGNS_OF_DIARRHEA
NO_SIGNS_OF_LAMENESS_OR_DIARRHEA
NORMAL DEFECATION
NORMAL_MANURE
NORMAL_MANURE
NOT ISOLATED
PKSTOP_IF_DIARRHEA
PLANNING_
RCSTOP_IF_DIARRHEA
RGSTOP_IF_DIARRHEA
RISKS_WITH_OXYTET_INJECTIONS_INCLUDING_RESP._DISTRESS_COL
IC_DIARRHEA
SHOWS_SIGNS_OF_COLIC_OR_DIARRHEA
SIGNS_OF_LAMINITIS_WORSENING_SKIN_IRRITATION_DIARRHEA
SOFT_FAECES_PALPABLE_IN_RECTUM
SOFT_FECES_FILLING_THE_RECTUM

SPECTAM_SCOUR
STOPPED_SCOURING_YESTERDAY
SWAB_STANDARD_CHARCOAL
SWELLING_ABOVE_THE_WRAP_DECREASED_APPETITE_DIARRHEA
THERE_IS_SOME_RISK_OF_DIARRHEA
THIS_MEDICATION_CAN_CAUSE_DIARRHEA
THIS_MEDICATION_CAN_CAUSE_LOOSE_MANURE
WATCH_FOR_ANY_DIARRHEA_AND_CALL_IMMEDIATELY_AND_DISCO
NTINUE_MEDS_IF_IT_HAPPENS
WATCH_FOR_ANY_SIGN_OF_DIARRHEA
WATCH_FOR_DIARRHEA
WATCH_FOR_DIARRHEA
WATCH_FOR_DIARRHEA_LACK_OF_FECES
WATCH_FOR_DIARRHEA_TEETH_GRINDING_COLIC
WILL_LIKELY_PASS_SOME_SOFT_MANURE

CORTICOSTEROIDS INCLUSION DICTIONARY
ADCORTYL
AZIUM
AZIUM_GRANULES
BECLAMETHASONE
BECLAZONE
BECLOFORTE
BECLOMETHASONE
BECLOMETHOSONE
BESAMETHASONE
BETAMETASONE
BETSOLAN
BUTECORT
COLVASONE
CORT
CORTICOSTEROID
CORTICOSTEROIDS
CORTIFLEX
CORTIFLEX
CORTISONE
CORTIZONE
CORTRIL
DEPO
DEPOMDERONE
DEPOMED
DEPOMEDRAL
DEPOMEDROL
DEPOMEDROME
DEPOMEDRON
DEPOMEDRONE
DEX
DEXA
DEXA
DEXADRESON

DEXADRESON
DEXADRESSON
DEXADREXON
DEXAFORT
DEXAMET
DEXAMETH
DEXAMETHANANE
DEXAMETHASIONE
DEXAMETHASON
DEXAMETHASONE
DEXAMETHAZONE
DEXAMETHAZONE
DEXAMETHEASONE
DEXAMETHOSAONE
DEXAMETHOSONE
DEXAMOTHESOME
DEXASONE
DEXOMETHOSONE
DUPHACORT
DUPHACORT
DUPHADUOCORT
DXASON
EKYFLOGYL
FLIXOTIDE
FLUMETHASONE
FLUTICASONE
HYDROCORTISONE
HYRDROCORTISONE
ISOFLUPREDONE
LAURABOLIN
MAXITROLED
MEDRONE
METAPIRONE
METHILDEX
METHILPREDNISOLONE
METHILPREDNISONE
METHYLPRED
METHYLPREDNISALONE
METHYL-PREDNISOLONE
METHYLPREDNISOLONE
METILPREDNISOLONE
MODULITE
NANDROLONE
NAQUASOME
NAQUASONE
NAQUAZONE
NAQUSONE
OPTICORTEN
PRECORTISYL
PRED

PREDNI
PREDNICARE
PREDNID
PREDNIDALE
PREDNISALONE
PREDNISOLONE
PREDNISONE
PREDNIZONE
PREDSOL
RAPIDEXON
RAPIDREXON
SERETIDE
SOLUDELTA
SOLU-DELTA
SOLUDELTACORTEF
SOLUMEDRONE
SOLU-MEDRONE
STEORIDS
STERIODS
STEROIDAL
STEROIDALS
STEROIDS
SUMDEX
STEROID
TRIAMCINOLONE
TRIAMCINALONE
TRIMACINOLONE

SYSTEMIC CORTICOSTEROIDS INCLUSION DICTIONARY
AZIUM
AZIUM_GRANULES
COLVASONE
CORTISONE
CORTIZONE
DESADREXON
DEX
DEXA
DEXADRESON
DEXADRESSON
DEXADREXON
DEXAFORT
DEXAMET
DEXAMETH
DEXAMETHANANE
DEXAMETHASIONE
DEXAMETHASON
DEXAMETHASONE
DEXAMETHAZONE
DEXAMETHEASONE
DEXAMETHOSAONE

DEXAMETHOSONE
DEXAMOTHESOME
DEXASONE
DEXOMETHOSONE
DUPHACORT
EKYFLOGYL
HYDROCORTISONE
HYRDROCORTISONE
ISOFLUPREDONE
METHILDEX
METHILPREDNISOLONE
METHILPREDNISONE
METHYLPREDNISALONE
METHYL-PREDNISOLONE
METILPREDNISOLONE
NAQUASOME
NAQUASONE
NAQUAZONE
NAQUSONE
PRED
PREDNIDALE
PREDNISALONE
PREDNISOLONE
PREDNISONE
PREDNIZONE
RAPIDEXON
RAPIDREXON
SUMDEX
VOREN

FLUID THERAPY INCLUSION DICTIONARY
AMINOSYN
AQUAFORM
AQUAPHARM
AQUPHARM
BICARBONATE
CALCIJECT
CALCIUM GLUCONATE
DECTROSE
DEXTROSE
DRIP
EXPANDER
FLUIDS
GLUCOSE_40%
HARTMAN
HARTMANN
HARTMANNS
HARTMANS
HETASTARCH
HYPERIMUNE

HYPERMUNE
HYPERTONIC
HYPERTONIC_SALINE
ISOLEC
ISOTONIC
IVFT
IVISOLEC
IVSALIN
IVSALINE
MEGNIJECT
NACL
NORMOSAL
NORMOSOL
PENTASTARCH
PLASMALYTE
PLASMA_TRANS.
PLASMA_TRANSFUSION
PLASMOLYTE
POLYIMUNE
POLYMUNE
POTASSIUM_CHLORIDE
SODIUM_CHLORIDE
STARCH
STARCHES
VETIVEX
ZOLCAL
ZOLCAL-D

FLUID THERAPY EXCLUSION DICTIONARY
DEXTROSE POWDER
GLUCOSE - GLUCOMETER
GLUCOSE PANEL
GLUCOSE SHOWED
GLUCOSE TEST STRIP
SAFE STARCH

LAXATIVES INCLUSION DICTIONARY
EPSOM_SALT
EPSOM_SALTS
LIQUID_PARAFFIN
MAGNESIUM
MAGNESIUM_SULPHATE
MINERAL_OIL
OIL
NSAIDS_TOXICITY
BUTE_TOXICITY
COLITIS FOLLOWING NSAIDS
FROM_NSAID_TREATMENT
NSAIDS_INDUCED
NSAID_INDUCED

NSAID_INDUCED
NSAID_POISONING
NSAID_TOXICITY
NSAID_TOXICOSIS
NSAIDS INDUCED
NSAIDS_POISONING
NSAIDS_TOXICITY
NSAIDS_TOXICOSIS
POST_NSAID_ADMINISTRATION
RDC_EPISODE
RD_COLITIS
RIGHT_DORSAL_COLITIS
RIGHT_DORSAL,COLITIS
RIGHT_DORSAL_COLITIS,
RIGHT_DORSAL_COLITS,

ANTHELMINTIC DRUG INCLUSION DICTIONARY
ADVOCATE
AMPROLIUM
ANTHELCIDE
ANTIPARASITIC
ATIPAMAZOLE
BENZELMIN
BIMECTIN
CYDECTIN
DARAMECTIN
DAVAMECTIN
DECOMAX
DECOTMAX
DECTAMAX
DECTMAX
DECTO
DECTOMA
DECTOMAC
DECTOMAS
DECTOMAX
DECTOMEC
DECTOMEX
DECTOMICIN
DECTOMOX
DECTROMAX
DEECTOMAX
DEWORM
DEWORME
DEWORMED
DEWORMER
DEWORMERS
DEWORMING
DEXTOMAX
DORAMECTIN

EQUALAN
EQUALAN
EQUELL
EQUEST
EQUEST_PRAMOX
EQUIMAX
EQUIMAXTAB
EQUIMAXX
EQUITAPE
EQUVALAN
EQVALAN
EQVALEN
ERAQUELL
FEBENDAZOLE
FENBENDAZOLE
FIPRONIL
FLUBENVET
FRONTLINE
IVERMEC
IVERMECRTIN
IVERMECTI
IVERMECTIC
IVERMECTIN
IVOMEC
IVOMECTIN
LARVICIDAL
LEVADIN
LEVAMISOLE
LEVASURE
MILBEMAX
MOXIDECTIN
NILVERM
NOROMECTIN
OXFENDAZOLE
OXYBENDAZOLE
PANACUR
PANACURE
PANEQSYR
PANOMEC
PARAMOX
PIPERAZINE
POTOMACGUARD
PRAMOX
PRAZIQUANTAL
PRAZIQUANTEL
PRMECTIN
PYRANTAL
PYRANTEL
PYRATAPE
RYPOSECT

SAFEUARD
STONGID
STRONGID
STRONGIDPA
STRONGIDPG
STRONGID_P
TAPEWORMER
TELMIN
TETRAMISOLE
VECTIN
WAZINE
WORMER
ZIMECTERIN
ZIMECTIN
ZIMECTRIN

List of References

Akaike, H., 1998. Information Theory and an Extension of the Maximum Likelihood Principle, in: Parzen, E., Tanabe, K., Kitagawa, G. (Eds.), Selected Papers of Hirotugu Akaike, Springer Series in Statistics. Springer New York, pp. 199–213.

Akaike, H., 1974. A new look at the statistical model identification. IEEE Transactions on Automatic Control 19, 716–723.

Andrews, F., McConnico, R., 2009. Cause for concern: Evidence that therapeutic dosing of nonselective NSAIDs contributes to gastrointestinal injury. Equine Veterinary Education 21, 663–664.

Andrews, F., Reineimeyer, C.R., Longhofer, S.L., 2009. Effects of top-dress formulations of suxibuzone and phenylbutazone on development of gastric ulcers in horses. Veterinary Therapeutics 10, 113–120.

Anholt, R.M., Berezowski, J., Jamal, I., Ribble, C., Stephen, C., 2014a. Mining free-text medical records for companion animal enteric syndrome surveillance. Preventive Veterinary Medicine 113, 417–422.

Anholt, R.M., Berezowski, J., Ribble, C.S., Russell, M.L., Stephen, C., 2014b. Using Informatics and the Electronic Medical Record to Describe Antimicrobial Use in the Clinical Management of Diarrhea Cases at 12 Companion Animal Practices. PLoS ONE 9, e103190.

Appleby, S.B., Ristimäki, A., Neilson, K., Narko, K., Hla, T., 1994. Structure of the human cyclo-oxygenase-2 gene. Biochemical Journal 302, 723–727.

Bakhle, Y., Botting, R., 1996. Cyclooxygenase-2 and its regulation in inflammation. Mediators of Inflammation 5, 305–323.

Balas, E.A., Vernon, M., Magrabi, F., Gordon, L.T., Sexton, J., 2015. Big Data Clinical Research: Validity, Ethics, and Regulation., in: MEDINFO 2015: EHealth-Enabled Health: Proceedings of the 15th World Congress on Health and Biomedical Informatics. IOS Press, p. 448.

Barr, B.S., 2006. Infiltrative Intestinal Disease. Veterinary Clinics of North America: Equine Practice 22, e1–e7.

Barrett, E.J., Blair, C.W., Farlam, J., Proudman, C.J., 2005. Postdosing colic and diarrhoea in horses with serological evidence of tapeworm infection. Veterinary Record 156, 252–253.

Barton, M.H., 2010. Disorders of the Liver, in: Equine Internal Medicine. Saunders, pp. 939–975.

Barton, M. h., Paske, E., Norton, N., King, D., Giguère, S., Budsberg, S., 2014. Efficacy of cyclooxygenase inhibition by two commercially available firocoxib products in horses. Equine Veterinary Journal 46, 72–75.

Beretta, C., Garavaglia, G., Cavalli, M., 2005. COX-1 and COX-2 inhibition in horse blood by phenylbutazone, flunixin, carprofen and meloxicam: An in vitro analysis. Pharmacological Research 52, 302–306.

Bergh, M.S., Budsberg, S.C., 2005. The Coxib NSAIDs: Potential Clinical and Pharmacologic Importance in Veterinary Medicine. Journal of Veterinary Internal Medicine 19, 633–643.

Bjorkman, D., 1998. Nonsteroidal anti-inflammatory drug-associated toxicity of the liver, lower gastrointestinal tract, and esophagus. The American Journal of Medicine 105, 17S–21S.

Black, H.E., 1986. Renal toxicity of non-steroidal anti-inflammatory drugs. Toxicologic Pathology 14, 83–90.

Blain, H., Boileau, C., Lapicque, F., Nédélec, E., Lœuille, D., Guillaume, C., Gaucher, A., Jeandel, C., Netter, P., Jouzeau, J.-Y., 2002. Limitation of the in vitro whole blood assay for predicting the COX selectivity of NSAIDs in clinical use. British Journal of Clinical Pharmacology 53, 255–265.

Bland, J.M., Altman, D.G., 1986. Statistical methods for assessing agreement between two methods of clinical measurement. Lancet 1, 307–310.

Boden, L.A., Charles, J.A., Slocombe, R.F., Sandy, J.R., Finnin, P.J., Morton, J.M., Clarke, A.F., 2005. Sudden death in racing Thoroughbreds in Victoria, Australia. Equine Veterinary Journal 37, 269–71.

Boden, L.A., Parkin, T.D.H., 2008. Current guidelines on good reporting of analytical observational studies in epidemiology. Equine Veterinary Journal 40, 84–86.

Borer, L.R., Peel, J.E., Seewald, W., Schawalder, P., Spreng, D.E., 2003. Effect of carprofen, etodolac, meloxicam, or butorphanol in dogs with induced acute synovitis. American Journal of Veterinary Research 64, 1429-1437.

Botting, R.M., 2010. Vane's discovery of the mechanism of action of aspirin changed our understanding of its clinical pharmacology. Pharmacological Reports 62, 518–525.

Brideau, C., Van Staden, C., Chan, C.C., 2001. In vitro effects of cyclooxygenase inhibitors in whole blood of horses, dogs, and cats. American Journal of Veterinary Research 62, 1755–1760.

Brossette, S.E., Sprague, A.P., Hardin, J.M., Waites, K.B., Jones, W.T., Moser, S.A., 1998. Association rules and data mining in hospital infection control and public health surveillance. Journal of the American medical informatics association 5, 373–381.

Byars, T., Gonda, K., 2015. Equine history, physical examination, records, and recognizing abuse or neglect in patients, in: Large Animal Internal Medicine. Elsevier, St.Louis, pp. 13–22.

Cambridge, H., Lees, P., Hooke, R.E., Russell, C.S., 1991. Antithrombotic actions of aspirin in the horse. Equine Veterinary Journal 23, 123–127.

Cameron, N., Lederer, R., Bennett, D., Parkin, T., 2014. The prevalence of tail injuries in working and non-working breed dogs visiting veterinary practices in Scotland. Veterinary Record 174, 450–450.

Chandrasekharan, N., Dai, H., Roos, K., Evanson, N., Tomsik, J., Elton, T., Simmons, D., 2002. COX-3, a Cyclooxygenase-1 Variant Inhibited by Acetaminophen and Other Analgesic/Antipyretic Drugs: Cloning, Structure, and Expression. Proceedings of National Academy of Science 99, 13926–13931.

Chapman, W.W., Christensen, L.M., Wagner, M.M., Haug, P.J., Ivanov, O., Dowling, J.N., Olszewski, R.T., 2005. Classifying free-text triage chief complaints into syndromic categories with natural language processing. Artificial Intelligence in Medicine 33, 31–40.

Chen, E.S., Hripcsak, G., Xu, H., Markatou, M., Friedman, C., 2008. Automated Acquisition of Disease-Drug Knowledge from Biomedical and Clinical Documents: An Initial Study. Journal of the American Medical Informatics Association 15, 87–98.

Coakley, M., Peck, K.E., Taylor, T.S., Matthews, N.S., Mealey, K.L., 1999. Pharmacokinetics of flunixin meglumine in donkeys, mules, and horses. American Journal of Veterinary Research 60, 1441–1444.

Cohen, N.D., Carter, G.K., Mealey, R.H., Taylor, T.S., 1995. Medical Management of Right Dorsal Colitis in 5 Horses: A Retrospective Study (1987–1993). Journal of Veterinary Internal Medicine 9, 272–276.

Coleman, N., Halas, G., Peeler, W., Casaclang, N., Williamson, T., Katz, A., 2015. From patient care to research: a validation study examining the factors contributing to data quality in a primary care electronic medical record database. BMC Family Practice 16, 11-19

Collins, L.G., Tyler, D.E., 1985. Experimentally induced phenylbutazone toxicosis in ponies: description of the syndrome and its prevention with synthetic prostaglandin E2. American Journal of Veterinary Research 46, 1605–1615.

Collins, L.G., Tyler, D.E., 1984. Phenylbutazone toxicosis in the horse: a clinical study. Journal of the American Veterinary Medical Association 184, 699–703.

Cook, V.L., Meyer, C.T., Campbell, N.B., Blikslager, A.T., 2009a. Effect of firocoxib or flunixin meglumine on recovery of ischemic-injured equine jejunum. American Journal of Veterinary Research 70, 992–1000.

Cook, V.L., Shults, J.J., McDowell, M.R., Campbell, N.B., Davis, J.L., Marshall, J.F., Blikslager, A.T., 2009b. Anti-inflammatory effects of intravenously administered lidocaine hydrochloride on ischemia-injured jejunum in horses. American Journal of Veterinary Research 70, 1259-1268.

Cox, S., Dudenbostel, L., Sommardahl, C., Yarbrough, J., Saleh, M., Doherty, T., 2012. Pharmacokinetics of firocoxib and its interaction with enrofloxacin in horses. Journal of Veterinary Pharmacology and Therapeutics 35, 615–617.

Cox, S., Yarbrough, J., 2011. Determination of firocoxib in equine plasma using high performance liquid chromatography. Journal of Chromatography B Analytical Technologies in the Biomedical Life Sciences 879, 205–208.

Cuniberti, B., Odore, R., Barbero, R., Cagnardi, P., Badino, P., Girardi, C., Re, G., 2012. In vitro and *ex vivo* pharmacodynamics of selected non-steroidal anti-inflammatory drugs in equine whole blood. The Veterinary Journal 191, 327–333.

D'Arcy-Moskwa, E., Noble, G. k., Weston, L. a., Boston, R., Raidal, S. l., 2012. Effects of Meloxicam and Phenylbutazone on Equine Gastric Mucosal Permeability. Journal of Veterinary Internal Medicine 26, 1494–1499.

Davis, J.L., Marshall, J.F., Papich, M.G., Blikslager, A.T., Campbell, N.B., 2011. The pharmacokinetics and in vitro cyclooxygenase selectivity of deracoxib in horses. Journal of Veterinary Pharmacology and Therapeutics 34, 12–16.

De Bruijn, B., Martin, J., 2002. Getting to the core of knowledge: mining biomedical literature. International Journal of Medical Informatics 67, 7–18.

DeWitt, D.L., Smith, W.L., 1988. Primary Structure of Prostaglandin G/H Synthase from Sheep Vesicular Gland Determined from the Complementary DNA Sequence. Proceedings of the National Academy of Science 85, 1412–1416.

Dohoo, I., Martin, W., Stryhn, H., 2010. Veterinary Epidemiologic Research, 2nd ed. AVC Inc., Charlottetown.

Doucet, M.Y., Bertone, A.L., Hendrickson, D., Hughes, F., MacAllister, C., McClure, S., Reinemeyer, C., Rossier, Y., Sifferman, R., Vrins, A.A., White, G., Kunkle, B., Alva, R., Romano, D., Hanson, P.D., 2008. Comparison of efficacy and safety of paste formulations of firocoxib and phenylbutazone in horses with naturally occurring osteoarthritis. Journal of the American Veterinary Medical Association 232, 91–97.

Dowling, P.M., 2010. Introduction To Clinical Pharmacology, in: Equine Internal Medicine. Saunders, Philadelphia, pp. 148–204.

Dujardin, C.L.L., van Loon, J.P. a. M., 2011. Pain recognition and treatment in the horse: a survey of equine veterinarians in The Netherlands and Belgium. Tijdschr Diergeneeskd 136, 715–724.

Duz, M., Parkin, T.D., Cullander, R.M., Marshall, J.F., 2015. Effect of flunixin meglumine and firocoxib on *ex vivo* cyclooxygenase activity in horses undergoing elective surgery. American Journal of Veterinary Research 76, 208–215.

Ekiri, A.B., Morton, A.J., Long, M.T., MacKay, R.J., Hernandez, J.A., 2010. Review of the epidemiology and infection control aspects of nosocomial Salmonella infections in hospitalised horses. Equine Veterinary Education 22, 631–641.

Erstad, T.L., 2003. Analyzing computer based patient records: a review of literature. Journal of Healthcare Information Management 17, 51–57.

Faulkner, L.W., Erb, H.N., King, J.M., 1984. Renal papillary necrosis in equines. Bullettin of Environmental Contamination and Toxicology 33, 379–381.

Ferreira, S.H., Moncada, S., Vane, J.R., 1971. Indomethacin and aspirin abolish prostaglandin release from the spleen. Nature New Biology 231, 237–239.

FitzGerald, G.A., Patrono, C., 2001. The coxibs, selective inhibitors of cyclooxygenase-2. New England Journal of Medicine 345, 433–442.

Frawley, W.J., Piatetsky-Shapiro, G., Matheus, C.J., 1992. Knowledge discovery in databases: An overview. Ai Magazine 13, 57.

Friede, A., Blum, H.L., McDonald, M., 1995. Public Health Informatics: How Information-Age Technology Can Strengthen Public Health. Annual Review of Public Health 16, 239–252.

Friedlin, J., Grannis, S., Overhage, J.M., 2008. Using Natural Language Processing to Improve Accuracy of Automated Notifiable Disease Reporting. AMIA Annual Symposium Proceedings 2008, 207–211.

Frölich, J.C., 1997. A classification of NSAIDs according to the relative inhibition of cyclooxygenase isoenzymes. Trends in Pharmacological Sciences 18, 30–34.

Furst, D.E., 1999. Pharmacology and efficacy of cyclooxygenase (COX) inhibitors. American Journal of Medicine 107, 18S–22S; discussion 22S–26S.

Galvin, N., Dillon, H., McGovern, F., 2004. Right dorsal colitis in the horse: minireview and reports on three cases in Ireland. Irish Veterinary Journal 57, 467.

Garavito, R.M., DeWitt, D.L., 1999. The cyclooxygenase isoforms: structural insights into the conversion of arachidonic acid to prostaglandins. Biochimica et Biophysica Acta (BBA) - Molecular and Cell Biology of Lipids 1441, 278–287.

Garten, Y., Coulet, A., Altman, R.B., 2010. Recent progress in automatically extracting information from the pharmacogenomic literature. Pharmacogenomics 11, 1467–1489.

Gerbier, S., Yarovaya, O., Gicquel, Q., Millet, A.-L., Smaldore, V., Pagliaroli, V., Darmoni, S., Metzger, M.-H., 2011. Evaluation of natural language processing from emergency department computerized medical records for intra-hospital syndromic surveillance. BMC medical informatics and decision making 11, 50.

Gerring, E.L., Lees, P., Taylor, J.B., 1981. Pharmacokinetics of phenylbutazone and its metabolites in the horse. Equine Veterinary Journal 13, 152–157.

Gierse, J.K., Staten, N.R., Casperson, G.F., Koboldt, C.M., Trigg, J.S., Reitz, B.A., Pierce, J.L., Seibert, K., 2002. Cloning, expression, and selective inhibition of canine cyclooxygenase-1 and cyclooxygenase-2. Veterinary Therapeutics 3, 270–280.

Giraudel, J.M., Diquelou, A., Laroute, V., Lees, P., Toutain, P.-L., 2005. Pharmacokinetic/pharmacodynamic modelling of NSAIDs in a model of reversible inflammation in the cat. British Journal of Pharmacolology 146, 642–653.

Giraudel, J.M., Toutain, P.-L., King, J.N., Lees, P., 2009. Differential inhibition of cyclooxygenase isoenzymes in the cat by the NSAID

robenacoxib. Journal of Veterinary Pharmacology and Therapeutics 32, 31–40.

Giuliano, F., Warner, T.D., 1999. *Ex vivo* assay to determine the cyclooxygenase selectivity of non-steroidal anti-inflammatory drugs. British Journal of Pharmacology 126, 1824–1830.

Glaser, K., 1995. Cyclooxygenase selectivity and NSAIDs: Cyclooxygenase-2 selectivity of etodolac (LODINE). Inflammopharmacology 3, 335–345.

Goldman, J.A., Chu, W.W., Parker, D.S., Goldman, R.M., 1999. Term domain distribution analysis: a data mining tool for text databases. Methods of Information in Medicine 38, 96–101.

Gunson, D.E., Soma, L.R., 1983. Renal Papillary Necrosis in Horses after Phenylbutazone and Water Deprivation. Veterinary Pathology 20, 603–610.

Harman, D., 1996. The text retrieval conferences (trecs), in: Proceedings of a Workshop Held at Vienna, Virginia: May 6-8, 1996. pp. 373–410.

Harris, R.C., McKanna, J.A., Akai, Y., Jacobson, H.R., Dubois, R.N., Breyer, M.D., 1994. Cyclooxygenase-2 is associated with the macula densa of rat kidney and increases with salt restriction. Journal of Clinical Investigation 94, 2504–2510.

Hassey, A., 2001. A survey of validity and utility of electronic patient records in a general practice. British Medical Journal 322, 1401–1405.

Heath, M.F., Evans, R.J., Poole, A.W., Hayes, L.J., McEvoy, R.J., Littler, R.M., 1994. The effects of aspirin and paracetamol on the aggregation of equine blood platelets. Journal of Veterinary Pharmacology and Therapeutics 17, 374–378.

Heinze, D.T., Morsch, M.L., Holbrook, J., 2001. Mining free-text medical records. Procedings of AMIA Symposium 254–258.

Held, J.P., Daniel, G.B., 1991. Use of nonimaging nuclear medicine techniques to assess the effect of flunixin meglumine on effective renal plasma flow and effective renal blood flow in healthy horses. American Journal of Veterinary Research 52, 1619–1621.

Helmerhorst, H.J., Brage, S., Warren, J., Besson, H., Ekelund, U., 2012. A systematic review of reliability and objective criterion-related validity of physical activity questionnaires. International Journal of Behavioral Nutrition and Physical Activity 9, 103.

Hewetson, M., Cohen, N.D., Love, S., Buddington, R.K., Holmes, W., Innocent, G.T., Roussel, A.J., 2006. Sucrose concentration in blood: a new method for assessment of gastric permeability in horses with

gastric ulceration. Journal of Veterinary Internal Medicine 20, 388–94.

Higgins, A.J., Lees, P., Sharma, S.C., Taylor, J.B., 1987. Measurement of flunixin in equine inflammatory exudate and plasma by high performance liquid chromatography. Equine Veterinary Journal 19, 303–306.

Hinz, B., Brune, K., 2008. Can drug removals involving cyclooxygenase-2 inhibitors be avoided? A plea for human pharmacology. Trends in Pharmacological Sciences 29, 391–397.

Hotchkiss, J.W., Reid, S.W.J., Christley, R.M., 2007. A survey of horse owners in Great Britain regarding horses in their care. Part 1: Horse demographic characteristics and management. Equine Veterinary Journal 39, 294–300.

Hough, M., Steel, C., Bolton, J., Yovich, J., 1999. Ulceration and stricture of the right dorsal colon after phenylbutazone administration in four horses. Australian Veterinary Journal 77, 785–788.

House of Commons Committee of Public Accounts, 2011. The National Program for IT in the NHS: an update on the delivery of detailed care records systems - Forty-fifth Report of Session 2010–12 (No. HC 1070)

House of Commons Health Committee, 2007. The Electronic Patient Record - Sixth Report of Session 2006–07 (No. HC 422-I)

Hubbell, J.A.E., Saville, W.J.A., Bednarski, R.M., 2010. The use of sedatives, analgesic and anaesthetic drugs in the horse: An electronic survey of members of the American Association of Equine Practitioners (AAEP). Equine Veterinary Journal 42, 487–493.

Huber, L., 1998. Validation and Qualification in Analytical Laboratories. Taylor & Francis Ed.

Hugonnard, M., Leblond, A., Keroack, S., Cadoré, J.-L., Troncy, E., 2004. Attitudes and concerns of French veterinarians towards pain and analgesia in dogs and cats. Veterinary Anaesthesia and Analgesia 31, 154–163.

Iliopoulos, I., Enright, A.J., Ouzounis, C.A., 2001. Textquest: document clustering of Medline abstracts for concept discovery in molecular biology. Pacific Symposium of Biocomputing 384–395.

Jacobsen, S., Nielsen, J.V., Kjelgaard-Hansen, M., Toelboell, T., Fjeldborg, J., Halling-Thomsen, M., Martinussen, T., Thoefner, M.B., 2009. Acute Phase Response to Surgery of Varying Intensity in Horses: A Preliminary Study. Veterinary Surgery 38, 762–769.

Jaraiz, Rodriguez, San Andres, Gonzalez, San Andres, 1999. Pharmacokinetics and bioequivalence of two suxibuzone oral dosage forms in horses. Journal of Veterinary Pharmacology and Therapeutics 22, 247–254.

Jerie, P., 2006. [Milestones of cardivascular pharmacotherapy: salicylates and aspirin]. Casopís lékarů českých 145, 901–904.

Jones, P.H., Dawson, S., Gaskell, R.M., Coyne, K.P., Tierney, á., Setzkorn, C., Radford, A.D., Noble, P.-J.M., 2014. Surveillance of diarrhoea in small animal practice through the Small Animal Veterinary Surveillance Network (SAVSNET). The Veterinary Journal 201,412-418.

Jones, S.L., Davis, J., Rowlingson, K., 2003. Ultrasonographic findings in horses with right dorsal colitis: five cases (2000-2001). Journal of the American Veterinary Medical Association 222, 1248–51.

Joubert, K.E., 2001. The use of analgesic drugs by South African veterinarians. Journal of the South Africa Veterinary Association 72, 57–60.

Karcher, L.F., Dill, S.G., Anderson, W.I., King, J.M., 1990. Right Dorsal Colitis. Journal of Veterinary Internal Medicine 4, 247–253.

Kargman, S., Charleson, S., Cartwright, M., Frank, J., Riendeau, D., Mancini, J., Evans, J., O'Neill, G., 1996. Characterization of Prostaglandin G/H Synthase 1 and 2 in rat, dog, monkey, and human gastrointestinal tracts. Gastroenterology 111, 445–454.

Kay-Mugford, P., Benn, S.J., LaMarre, J., Conlon, P., 2000. In vitro effects of nonsteroidal anti-inflammatory drugs on cyclooxygenase activity in dogs. American Journal of Veterinary Research 61, 802–810.

King, J.N., Rudaz, C., Borer, L., Jung, M., Seewald, W., Lees, P., 2010. In vitro and *ex vivo* inhibition of canine cyclooxygenase isoforms by robenacoxib: A comparative study. Research in Veterinary Science 88, 497–506.

Krallinger, M., Valencia, A., Hirschman, L., 2008. Linking genes to literature: text mining, information extraction, and retrieval applications for biology. Genome Biology 9, S8.

Kreis, C., Gorman, P., 1997. Word frequency analysis of dictated clinical data: a user-centered approach to the design of a structured data entry interface. Proceedings AMIA Annual Fall Symposium 724–728.

Kurumbail, R.G., Stevens, A.M., Gierse, J.K., McDonald, J.J., Stegeman, R.A., Pak, J.Y., Gildehaus, D., Iyashiro, J.M., Penning, T.D., Seibert, K., Isakson, P.C., Stallings, W.C., 1996. Structural basis for

selective inhibition of cyclooxygenase-2 by anti-inflammatory agents. Nature 6610, 644-648

Kvaternick, V., Pollmeier, M., Fischer, J., Hanson, P.D., 2007. Pharmacokinetics and metabolism of orally administered firocoxib, a novel second generation coxib, in horses. Journal of Veterinary Pharmacology and Therapeutics 30, 208–217.

Lam, K.H., Parkin, T.D.H., Riggs, C.M., Morgan, K., 2007a. Use of free text clinical records in identifying syndromes and analysing health data. Veterinary Record 161, 547–551.

Lam, K.H., Parkin, T.D.H., Riggs, C.M., Morgan, K.L., 2007b. Evaluation of detailed training data to identify risk factors for retirement because of tendon injuries in Thoroughbred racehorses. American Journal of Veterinary Research 68, 1188–1197.

Lam, K.H., Parkin, T.D.H., Riggs, C.M., Morgan, K.L., 2007c. Descriptive analysis of retirement of Thoroughbred racehorses due to tendon injuries at the Hong Kong Jockey Club (1992–2004). Equine Veterinary Journal 39, 143–148.

Laneuville, O., Breuer, D.K., Dewitt, D.L., Hla, T., Funk, C.D., Smith, W.L., 1994. Differential inhibition of human prostaglandin endoperoxide H synthases-1 and -2 by nonsteroidal anti-inflammatory drugs. Journal of Pharmacology and Experimental Therapeutics 271, 927–934.

Lees, P., Creed, R.F.S., Gerring, E.E.L., Gould, P.W., Humphreys, D.J., Maitho, T.E., Michell, A.R., Taylor, J.B., 1983. Biochemical and haematological effects of phenylbutazone in horses. Equine Veterinary Journal 15, 158–167.

Lees, P., Giraudel, J., Landoni, M.F., Toutain, P.L., 2004a. PK–PD integration and PK–PD modelling of nonsteroidal anti-inflammatory drugs: principles and applications in veterinary pharmacology. Journal of Veterinary Pharmacology and Therapeutics 27, 491–502.

Lees, P., Higgins, A.J., 1985. Clinical pharmacology and therapeutic uses of non-steroidal anti-inflammatory drugs in the horse. Equine Veterinary Journal 17, 83–96.

Lees, P., Landoni, M.F., Giraudel, J., Toutain, P.L., 2004b. Pharmacodynamics and pharmacokinetics of nonsteroidal anti-inflammatory drugs in species of veterinary interest. Journal of Veterinary Pharmacology and Therapeutics 27, 479–490.

Lees, P., Michell, A.R., 1979. Phenylbutazone toxicity in ponies. Veterinary Record 105, 150–151.

Lees, P., Sedgwick, A.D., Higgins, A.J., Pugh, K.E., Busch, U., 1991. Pharmacodynamics and pharmacokinetics of meloxicam in the horse. British Veterinary Journal 147, 97–108.

Lee, W.M., 2003. Drug-Induced Hepatotoxicity. New England Journal of Medicine 349, 474–485.

Letendre, L.T., Tessman, R.K., McClure, S.R., Kvaternick, V.J., Fischer, J.B., Hanson, P.D., 2008. Pharmacokinetics of firocoxib after administration of multiple consecutive daily doses to horses. American Journal of Veterinary Research 69, 1399–1405.

Li, J.J., Anderson, G.D., Burton, E.G., Cogburn, J.N., Collins, J.T., Garland, D.J., Gregory, S.A., Huang, H.C., Isakson, P.C., Koboldt, C.M., 1995. 1,2-Diarylcyclopentenes as selective cyclooxygenase-2 inhibitors and orally active anti-inflammatory agents. Journal of Medical Chemistry 38, 4570–4578.

Longhofer, S.L., Reinemeyer, C.R., Radecki, S.V., 2008. Evaluation of the palatability of three nonsteroidal antiinflammatory top-dress formulations in horses. Veterinary Therapeutics 9, 122–127.

Love, S., 2003. Treatment and prevention of intestinal parasite–associated disease. Veterinary Clinics of North America: Equine Practice 19, 791–806.

Luong, C., Miller, A., Barnett, J., Chow, J., Ramesha, C., Browner, M.F., 1996. Flexibility of the NSAID binding site in the structure of human cyclooxygenase-2. Nature Structural Biology 3, 927–933.

MacAllister, C.G., Morgan, S.J., Borne, A.T., Pollet, R.A., 1993. Comparison of adverse effects of phenylbutazone, flunixin meglumine, and ketoprofen in horses. Journal of the American Veterinary Medical Association 202, 71–77.

MacKay, R.J., French, T.W., Nguyen, H.T., Mayhew, I.G., 1983. Effects of large doses of phenylbutazone administration to horses. American Journal of Veterinary Research 44, 774–780.

Mair, T.S., 1993. Recurrent diarrhoea in aged ponies associated with larval cyathostomiasis. Equine Veterinary Journal 25, 161–163.

Marshall, J.F., 2010. The Effect of Novel Anti-inflammatory Drugs on the Cyclooxygenase Enzymes and Recovery of Mucosal Barrier Function (PhD Thesis). North Carolina State University, Raleigh.

Marshall, J.F., Bhatnagar, A.S., Bowman, S.G., Howard, C.M., Morris, N.N., Skorich, D.A., Redding, C.D., Blikslager, A.T., 2011. Evaluation of the cyclooxygenase selectivity of robenacoxib and its effect on recovery of ischemia-injured jejunal mucosa in horses. American Journal of Veterinary Research 72, 226–232.

Mattin, M., O'Neill, D., Church, D., McGreevy, P.D., Thomson, P.C., Brodbelt, D., 2014. An epidemiological study of diabetes mellitus in dogs attending first opinion practice in the UK. Veterinary Record 174, 349–349.

McCann, M., Andersen, D., Brideau, C., Black, W., Zhang, D., Hickey, G., 2002. In vitro activity and in vivo efficacy of a novel COX-2 inhibitor in the horse, in: Research Abstract Program. Proceedings Annual ACVIM forum, Dallas, TX, p. 355.

McCann, M.E., Andersen, D.R., Zhang, D., Brideau, C., Black, W.C., Hanson, P.D., Hickey, G.J., 2004. In vitro effects and in vivo efficacy of a novel cyclooxygenase-2 inhibitor in dogs with experimentally induced synovitis. American Journal of Veterinary Research 65, 503–512.

McGorum, B.C., Pirie, R.S., 2009. Antimicrobial associated diarrhoea in the horse. Part 1: Overview, pathogenesis and risk factors. Equine Veterinary Education 21, 610–616.

Merlie, J.P., Fagan, D., Mudd, J., Needleman, P., 1988. Isolation and Characterization of the Complementary DNA for Sheep Seminal Vesicle Prostaglandin Endoperoxide Synthase (cyclooxygenase). Journal of Biological Chemistry 263, 3550–3553.

Meschter, C.L., Gilbert, M., Krook, L., Maylin, G., Corradino, R., 1990a. The effects of phenylbutazone on the intestinal mucosa of the horse: a morphological, ultrastructural and biochemical study. Equine Veterinary Journal 22, 255–263.

Meschter, C.L., Gilbert, M., Krook, L., Maylin, G., Corradino, R., 1990b. The effects of phenylbutazone on the morphology and prostaglandin concentrations of the pyloric mucosa of the equine stomach. Veterinary Pathology 27, 244–253.

Meschter, C.L., Maylin, G.A., Krook, L., 1984. Vascular pathology in phenylbutazone intoxicated horses. Cornell Vet 74, 282–297.

Mitchell, J.A., Warner, T.D., 1999. Cyclo-oxygenase-2: pharmacology, physiology, biochemistry and relevance to NSAID therapy. British Journal of Pharmacology 128, 1121–1132.

Monreal, L., Sabaté, D., Segura, D., Mayós, I., Homedes, J., 2004. Lower gastric ulcerogenic effect of suxibuzone compared to phenylbutazone when administered orally to horses. Research in Veterinary Science 76, 145–149.

Moreau, M., Dupuis, J., Bonneau, N.H., Desnoyers, M., 2003. Clinical evaluation of a nutraceutical, carprofen and meloxicam for the treatment of dogs with osteoarthritis. Veterinary Record 152, 323–329.

Moses, V.S., Bertone, A.L., 2002. Nonsteroidal anti-inflammatory drugs. Veterinary Clinics of North America: Equine Practice 18, 21–37.

Murray, M.J., Grodinsky, C., Anderson, C.W., Radue, P.F., Schmidt, G.R., 1989. Gastric ulcers in horses: a comparison of endoscopic findings in horses with and without clinical signs. Equine Veterinary Journal S7, 68–72.

Naylor, R.J., Taylor, A.H., Knowles, E.J., Wilford, S., Linnenkohl, W., Mair, T.S., Johns, I.C., 2014. Comparison of flunixin meglumine and meloxicam for post operative management of horses with strangulating small intestinal lesions. Equine Veterinary Journal 4, 427–434.

Nielsen, M., Mittel, L., Grice, A., Erskine, M., Graves, E., Vaala, W., Tully, R., French, D., Bowman, R., Kaaplan, R., 2013. AAEP Parasite Control Guidelines.

Noreen, Y., Ringbom, T., Perera, P., Danielson, H., Bohlin, L., 1998. Development of a Radiochemical Cyclooxygenase-1 and -2 in Vitro Assay for Identification of Natural Products as Inhibitors of Prostaglandin Biosynthesis. Journal of Natural Products 61, 2–7.

O'Conner, M.S., Steiner, J.M., Roussel, A.J., Williams, D.A., Meddings, J.B., Pipers, F., Cohen, N.D., 2004. Evaluation of Urine Sucrose Concentration for Detection of Gastric Ulcers in Horses. American Journal of Veterinary Research 65, 31–39.

Ogino, K., Hatanaka, K., Kawamura, M., Katori, M., Harada, Y., 1997. Evaluation of pharmacological profile of meloxicam as an anti-inflammatory agent, with particular reference to its relative selectivity for cyclooxygenase-2 over cyclooxygenase-1. Pharmacology 55, 44–53.

O'Neill, D., Church, D., Mcgreevy, P., Middleton, S., Summers, J., Thomson, P., Brodbelt, D., 2012a. Primary practice clinical data for companion animal surveillance, in: Book of Abstracts of the 13th International Symposium on Veterinary Epidemiology and Economics. Proceedings of ISVEE 13, Maastricht, p. 59.

O'Neill, D.G., Elliott, J., Church, D.B., McGreevy, P.D., Thomson, P.C., Brodbelt, D.C., 2013. Chronic Kidney Disease in Dogs in UK Veterinary Practices: Prevalence, Risk Factors, and Survival. Journal of Veterinary Internal Medicine 27, 814–821.

O'Neill, D., Hendricks, A., Summers, J., Brodbelt, D., 2012b. Primary care veterinary usage of systemic glucocorticoids in cats and dogs in three UK practices. Journal of Small Animal Practice 53, 217–222.

O'Neill, D., Summers, J., Middleton, S., Church, D., Brodbelt, D., McGreevy, P., Thomson, P., 2011. Disease surveillance project in pedigree dogs and cats. Veterinary Record 168, 414–414.

Orsini, J.A., Ryan, W.G., Carithers, D.S., Boston, R.C., 2012. Evaluation of oral administration of firocoxib for the management of musculoskeletal pain and lameness associated with osteoarthritis in horses. American Journal of Veterinary Research 73, 664–671.

Ortiz-Pelaez, Á., Pfeiffer, D.U., 2008. Use of data mining techniques to investigate disease risk classification as a proxy for compromised biosecurity of cattle herds in Wales. BMC Veterinary Research 4, 24.

Oswald, J., Love, S., Parkin, T.D.H., Hughes, K.J., 2010. Prevalence of cervical vertebral stenotic myelopathy in a population of thoroughbred horses. Veterinary Record 166, 82–83.

Pairet, M., 1998. Inhibition of cyclooxygenase-1 and cyclooxygenase-2 analysis of in vitro test systems and their clinical relevance. Journal Clinical Rheumatology 4, s17–25.

Papich, M.G., 2008. An update on nonsteroidal anti-inflammatory drugs (NSAIDs) in small animals. Veterinary Clinics of North America: Small Animal Practice 38, 1243–1266, vi.

Patrignani, P., Panara, M.R., Greco, A., Fusco, O., Natoli, C., Iacobelli, S., Cipollone, F., Ganci, A., Creminon, C., Maclouf, J., 1994. Biochemical and pharmacological characterization of the cyclooxygenase activity of human blood prostaglandin endoperoxide synthases. Journal of Pharmacology and Experimental Therapeutics 271, 1705–1712.

Payne, M., Baynes, R., Sundlof, S., Craigmil, A., Webb, A., Riviere, J., 1999. Drugs prohibited from extralabel use in food animals. Journal of the American Veterinary Medical Association 215, 28–32.

Peck, K.E., Matthews, N.S., Taylor, T.S., Mealey, K.L., 2002. Pharmacokinetics of sulfamethoxazole and trimethoprim in donkeys, mules, and horses. American Journal of Veterinary Research 63, 349–53.

Pellegrini-Masini, A., Poppenga, R.H., Sweeney, R.W., 2004. Disposition of flunixin meglumine injectable preparation administered orally to healthy horses. Journal of Veterinary Pharmacology and Therapeutics. 27, 183–6.

Petrova, M., Sutcliffe, P., Fulford, K.W.M. (Bill), Dale, J., 2012. Search terms and a validated brief search filter to retrieve publications on health-related values in Medline: a word frequency analysis study. Journal of the American Medical Informatics Association 19, 479–488.

Piatetsky-Shapiro, G., 2000. Knowledge discovery in databases: 10 years after. SIGKDD Explorations 1, 59–61.

Picot, D., Loll, P.J., Garavito, R.M., 1994. The X-ray crystal structure of the membrane protein prostaglandin H_2 synthase-1. Nature 6460, 243-249

Pringle, M., Ward, P., Chilvers, C., 1995. Assessment of the completeness and accuracy of computer medical records in four practices committed to recording data on computer. British Journal General Practice 45, 537–541.

Punke, J.P., Speas, A.L., Reynolds, L.R., Budsberg, S.C., 2008. Effects of firocoxib, meloxicam, and tepoxalin on prostanoid and leukotriene production by duodenal mucosa and other tissues of osteoarthritic dogs. American Journal of Veterinary Research 69, 1203–1209.

Radford, A.D., Noble, P.J., Coyne, K.P., Gaskell, R.M., Jones, P.H., Bryan, J.G.E., Setzkorn, C., Tierney, A., Dawson, S., 2011. Antibacterial prescribing patterns in small animal veterinary practice identified via SAVSNET: the small animal veterinary surveillance network. Veterinary Record 169, 310–310.

Read, W.K., 1983. Renal medullary crest necrosis associated with phenylbutazone therapy in horses. Veterinary Pathology 20, 662–669.

Richter, R.A., Freeman, D.E., Wallig, M., Whittem, T., Baker, G.J., 2002. In vitro anion transport alterations and apoptosis induced by phenylbutazone in the right dorsal colon of ponies. American Journal of Veterinary Research 63, 934–41.

Ricketts, A.P., Lundy, K.M., Seibel, S.B., 1998. Evaluation of selective inhibition of canine cyclooxygenase 1 and 2 by carprofen and other nonsteroidal anti-inflammatory drugs. American Journal of Veterinary Research 59, 1441–1446.

Roque, F.S., Jensen, P.B., Schmock, H., Dalgaard, M., Andreatta, M., Hansen, T., Søeby, K., Bredkjær, S., Juul, A., Werge, T., Jensen, L.J., Brunak, S., 2011. Using electronic patient records to discover disease correlations and stratify patient cohorts. PLoS Computational Biology 7, e1002141.

Ross, S.E., Duz, M., Rendle, D.I., 2015. Antimicrobial selection and dosing in the treatment of wounds in the United Kingdom. Equine Veterinary Journal doi: 10.1111/evj.12535. [Epub ahead of print]

Sabaté, D., Homedes, J., Salichs, M., Sust, M., Monreal, L., 2009. Multicentre, controlled, randomised and blinded field study

comparing efficacy of suxibuzone and phenylbutazone in lame horses. Equine Veterinary Journal 41, 700–705.

Sager, N., Lyman, M., Bucknall, C., Nhan, N., Tick, L.J., 1994. Natural language processing and the representation of clinical data. Journal of the American Medical Informatcs Association 1, 142–160.

Salton, G., 1971. The SMART Retrieval System - Experiments in Automatic Document Processing. Prentice-Hall, Inc., Upper Saddle River, NJ, USA.

Salton, G., Lesk, M.E., 1968. Computer Evaluation of Indexing and Text Processing. Journal of the ACM 15, 8–36.

Sanchez, L., 2010. Disorders of the gastrointestinal system, in: Equine Internal Medicine. Saunders, Elsevier, St.Louis, MO, pp. 777–938.

Sanchez, L.C., Robertson, S.A., 2014. Pain control in horses: What do we really know?: Pain control. Equine Veterinary Journal 46, 517–523.

Schmid, V. b., Seewald, W., Lees, P., King, J. n., 2010. In vitro and *ex vivo* inhibition of COX isoforms by robenacoxib in the cat: a comparative study. Journal of Veterinary Pharmacology and Therapeutics 33, 444–452.

Sellon, D., Wise, L., 2010. Disorders of the hematopoietic system, in: Reed SM, Bayly WM, Sellon DC, Eds. Equine Internal Medicine. Saunders, St. Louis, Missouri, pp. 730–776.

Simmons, T.R., Gaughan, E.M., Ducharme, N.G., Dill, S.G., King, J.M., Anderson, W.I., 1990. Treatment of right dorsal ulcerative colitis in a horse. Journal of the American Veterinary Medical Association 196, 455–458.

Singh, G., 1998. Recent considerations in nonsteroidal anti-inflammatory drug gastropathy. The American Journal of Medicine 105, 31S–38S.

Singh, G., Triadafilopoulos, G., 1999. Epidemiology of NSAID induced gastrointestinal complications. Journal of Rheumatology Suppl 56, 18–24.

Slingsby, L.S., Waterman-Pearson, A.E., 2001. Analgesic effects in dogs of carprofen and pethidine together compared with the effects of either drug alone. Veterinary Record 148, 441–444.

Smith, J.B., Willis, A.L., 1971. Aspirin selectively inhibits prostaglandin production in human platelets. Nature New Biology 231, 235–237.

Smith, T.J., 1998. Cyclooxygenases as the principal targets for the actions of NSAIDs. Rheumatic Diseases Clinics of North America 24, 501–523.

Snow, D.H., Bogan, J.A., Douglas, T.A., Thompson, H., 1979. Phenylbutazone toxicity in ponies. Veterinary Record 105, 26–30.

Snow, D.H., Douglas, T.A., Thompson, H., Parkins, J.J., Holmes, P.H., 1981. Phenylbutazone toxicosis in equidae: a biochemical and pathophysiological study. American Journal Of Veterinary Research 42, 1754–1759.

Soma, L.R., Uboh, C.E., Maylin, G.M., 2012. The use of phenylbutazone in the horse. Journal of Veterinary Pharmacology and Therapeutics 35, 1–12.

Staempfli, H., Oliver-Espinosa, O., 2015. Clinical chemistry tests, in: Large Animal Internal Medicine. Elsevier, St. Louis, pp. 350–373.

Steyerberg, E., 2009. Clinical Prediction Models: a practical approach to development, validation and updating, in: Clinical Prediction Models: A Practical Approach to Development, Validation and Updating. Springer, pp. 190–211.

Stone, E., 1763. An account of the success of the bark of the willow in the cure of agues. Philosophical transactions of the Royal Society 53, 195–200.

Streppa, H.K., Jones, C.J., Budsberg, S.C., 2002. Cyclooxygenase selectivity of nonsteroidal anti-inflammatory drugs in canine blood. American Journal Of Veterinary Research 63, 91–94.

Tazawa, R., Xu, X.M., Wu, K.K., Wang, L.H., 1994. Characterization of the Genomic Structure, Chromosomal Location and Promoter of Human Prostaglandin H Synthase-2 Gene. Biochemical and Biophysical Research Communications 203, 190–199.

Thrusfield, M., 2007. Veterinary epidemiology, 3rd ed. Blackwell Science Ltd., Oxford.

Tolman, K.G., 1998. Hepatotoxicity of non-narcotic analgesics. The American Journal of Medicine 105, 13S–19S.

Tomlinson, J.E., Blikslager, A.T., 2005. Effects of cyclooxygenase inhibitors flunixin and deracoxib on permeability of ischaemic-injured equine jejunum. Equine Veterinary Journal 37, 75–80.

Toutain, P.L., Autefage, A., Legrand, C., Alvinerie, M., 1994. Plasma concentrations and therapeutic efficacy of phenylbutazone and flunixin meglumine in the horse: pharmacokinetic/pharmacodynamic modelling. Journal of Veterinary Pharmacology and Therapeutics 17, 459–469.

Toutain, P.L., Reymond, N., Laroute, V., Garcia, P., Popot, M.A., Bonnaire, Y., Hirsch, A., Narbe, R., 2004. Pharmacokinetics of

meloxicam in plasma and urine of horses. American Journal of Veterinary Research 65, 1542–7.

Vago, T., Bevilacqua, M., Norbiato, G., 1995. Effect of nimesulide action time dependence on selectivity towards prostaglandin G/H synthase/cyclooxygenase activity. Arzneimittelforschung 45, 1096–1098.

Valat, J.P., Accardo, S., Reginster, J.Y., Wouters, M., Hettich, M., Lieu, P.L., 2001. A comparison of the efficacy and tolerability of meloxicam and diclofenac in the treatment of patients with osteoarthritis of the lumbar spine. Inflammation Research 50 Suppl 1, S30–34.

Vane, J.R., 2000. The fight against rheumatism: from willow bark to COX-1 sparing drugs. Journal of Physiology Pharmacology 51, 573–586.

Vane, J.R., 1971. Inhibition of prostaglandin synthesis as a mechanism of action for aspirin-like drugs. Nature New Biology 231, 232–235.

Vane, J.R., Bakhle, Y.S., Botting, R.M., 1998. Cyclooxygenases 1 and 2. Annual Review of Pharmacology and Toxicology 38, 97–120.

Vane, J.R., Botting, R.M., 1995. New insights into the mode of action of anti-inflammatory drugs. Inflammation Research 44, 1–10.

Ward, M., Kelman, M., 2012. Disease surveillance in dogs and cats: a practitioner-based system, in: Book of Abstracts of the 13th International Symposium on Veterinary Epidemiology and Economics. Proceedings of ISVEE 13, Maastricht, p. 57.

Warner, T.D., Giuliano, F., Vojnovic, I., Bukasa, A., Mitchell, J.A., Vane, J.R., 1999. Nonsteroid drug selectivities for cyclo-oxygenase-1 rather than cyclo-oxygenase-2 are associated with human gastrointestinal toxicity: A full in vitro analysis. Proceedings of the National Academy of Sciences of the United States of America 96, 7563–7568.

Watson, A.D., Nicholson, A., Church, D.B., Pearson, M.R., 1996. Use of anti-inflammatory and analgesic drugs in dogs and cats. Australian Veterinary Journal 74, 203–210.

Weeber, M., Klein, H., Aronson, A.R., Mork, J.G., de Jong-van den Berg, L.T., Vos, R., 2000. Text-based discovery in biomedicine: the architecture of the DAD-system. Proceedings AMIA Symposium 903–907.

Weese, J.S., Giguère, S., Guardabassi, L., Morley, P.S., Papich, M., Ricciuto, D.R., Sykes, J.E., 2015. ACVIM Consensus Statement on Therapeutic Antimicrobial Use in Animals and Antimicrobial Resistance. Journal of Veterinary Internal Medicine 29, 487–498.

Whitelaw, F.G., Nevin, S.L., Milne, R.M., Taylor, R.J., Taylor, M.W., Watt, A.H., 1996. Completeness and accuracy of morbidity and repeat prescribing records held on general practice computers in Scotland. British Journal General Practice 46, 181–186.

Wilson, A.E., Pollock, C., Weekes, T., Dowell, A., 1995. Can general practice provide useful information? Evaluation of a primary health care information project in northern England. Journal of Epidemiology and Community Health 49, 227–230.

Wilson, E.B., 1927. Probable Inference, the Law of Succession, and Statistical Inference. Journal of the American Statistical Association 22, 209–212.

Wilson, J.E., Chandrasekharan, N.V., Westover, K.D., Eager, K.B., Simmons, D.L., 2004. Determination of expression of cyclooxygenase-1 and -2 isozymes in canine tissues and their differential sensitivity to nonsteroidal anti-inflammatory drugs. American Journal of Veterinary Research 65, 810–818.

Wolfe, M.M., Lichtenstein, D.R., Singh, G., 1999. Gastrointestinal toxicity of nonsteroidal antiinflammatory drugs. New England Journal of Medicine 340, 1888–1899.

Wooten, J.G., Blikslager, A.T., Ryan, K.A., Marks, S.L., Law, J.M., Lascelles, B.D.X., 2008. Cyclooxygenase expression and prostanoid production in pyloric and duodenal mucosae in dogs after administration of nonsteroidal anti-inflammatory drugs. American Journal of Veterinary Research 69, 457–464.

Workman, T.E., Stoddart, J.M., 2012. Rethinking information delivery: using a natural language processing application for point-of-care data discovery. Journal of Medical Library Association 100, 113–120.

Yocum, D., Fleischmann, R., Dalgin, P., Caldwell, J., Hall, D., Roszko, P., 2000. Safety and efficacy of meloxicam in the treatment of osteoarthritis: a 12-week, double-blind, multiple-dose, placebo-controlled trial. The Meloxicam Osteoarthritis Investigators. Archives of Internal Medicine 160, 2947–2954.

Yokoyama, C., Takai, T., Tanabe, T., 1988. Primary structure of sheep prostaglandin endoperoxide synthase deduced from cDNA sequence. FEBS Letters 231, 347–351.