A pilot study to explore the feasibility of building a cohort to investigate factors associated with dental caries in children

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Abstract

Dental caries is the most common chronic disease of childhood, worldwide and in Scotland. There has been a continuous steady improvement in the dental health of young children in Scotland since 2003. However, a clear gradient across all levels of deprivation remains, with those at the lower end of the socioeconomic spectrum suffering the greatest burden of caries. *Childsmile* is the national oral health improvement programme for young children in Scotland that involves a range of active interventions commencing from infancy. One of the key challenges *Childsmile* faces is in tackling oral health inequalities.

This study aimed to explore the feasibility of conducting a cohort study among preschool children, following them up one year later, to examine factors associated with dental caries and their role in explaining socioeconomic inequalities in caries.

A dental examination was carried out according to the British Association for the Study of Community Dentistry criteria, saliva & plaque samples collected and heights & weights measured according to the Royal College of Paediatrics & Child Health guidelines in nursery/school settings. Parents completed questionnaires on their child's habitual diet, early feeding habits, oral hygiene practices, use of dental services; and on their own attitudes & beliefs around oral health and socioeconomic position (SEP). Binary logistic regression was used to produce odds ratios and 95% Confidence Intervals for caries experience according to putative risk factors and socioeconomic position (SEP); and to estimate the relative index of inequality (RII) for caries experience by SEP and attenuation by the aforementioned factors.

Consent was obtained for 219 children (35%) and complete data (clinical and questionnaire) was available for 165 children (75%) in Sweep 1. One hundred and seventy five children (80%) were examined a year later in Sweep 2, of which 144 (66%) had complete data in Sweep 1. Mean [SD] age of the children at baseline was 4.8 [0.4] years and 47% were girls. The prevalence of caries experience was 35.8% at baseline and 47.7% at follow-up and showed a strong social gradient with both area- and individual-based measures of SEP. The level of *S.mutans* in

saliva and six other variables relating to current diet, early feeding habits, oral hygiene practices, use of dental services and SEP were found to be independently associated with caries experience at follow-up. All measures of SEP demonstrated high RIIs for caries experience. Despite some attenuation in the RII after adjustment for relevant risk factors, the socioeconomic gradient in caries persisted, suggesting that perhaps other more distal factors may be important in causing inequalities.

It was feasible to recruit a large number of preschool children and their parents from the most deprived areas of Glasgow via nursery schools and collect clinical and questionnaire data that were of sufficient quality over time across educational establishments. There is a strong socioeconomic gradient in the prevalence of caries, partially attenuated when adjusted for some important risk factors. Explaining the socioeconomic gradient is key to addressing oral health inequalities. The findings from this pilot study will be used to design a larger birth cohort study to robustly investigate modifiable risk factors of caries, identify ‘causes of the causes’, and design and test potential interventions to improve oral health of children overall and tackle and reduce oral health inequalities in the future.

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List of Abbreviations

AI/AN	American Indian and Alaska Native
ALSPAC	Avon Longitudinal Study of Parents and Children
AMPs	Anti-Microbial Proteins
AMR	Active/Carious dentinal decay, Missing and Restored
ANOVA	Analysis of Variance
BASCD	British Association for the Study of Community Dentistry
BMI	Body Mass Index
CDC	Centre for Disease Control and Prevention
CERT	*Childsmile* Research and Evaluation Team
CHP	Community Health Partnership
CHSP	Child Health Surveillance Programme
CHSP - PS	Child Health Surveillance Programme - Primary School
CI	Confidence Interval
CRF	Common Risk Factor
CS	*Childsmile*
DASH	Determinants of young Adult Social well-being and Health
$d_3$mft	Mean number of decayed, missing and filled teeth
DEPCAT	Carstairs and Morris' deprivation category
DHS RU	Dental Health Services Research Unit
DHSW	Dental Health Support Worker
DMF	Decayed, Missing, Filled
ECC	Early Childhood Caries
EDDNs	Extended Duty Dental Nurses
FFQ	Food Frequency Questionnaire
FSAS	Food Standards Agency Scotland
GA	General Anesthesia
GDHS	Glasgow Dental Hospital and School
GUS	Growing Up in Scotland
HEAT	Health Improvement, Efficiency, Access to Services and Treatment
HIC	Health Informatics Centre
ICDAS	International Caries Detection and Assessment System
IRAS	Integrated Research Application System
ISD	Information Statistics Division
LB	Lactobacillus species
LMS	Lambda Mu Sigma
LoC	Locus of Control
MCS	Millennium Cohort Study
MRC	Medical Research Council
MS	Mutans streptococci
NDIP	National Dental Inspection Programme
NHANES	National Health and Nutrition Examination Survey
NHS	National Health Service
NHS NSS	National Health Service National Services Scotland
NHSGGC	National Health Service Greater Glasgow & Clyde
NIDCR	National Institute of Dental and Craniofacial Research

NIHR	National Institute for Health Research
NMES	Non-Milk Extrinsic Sugars
NS SEC	UK National Statistics Socioeconomic Classification
NSHD	National Survey of Health and Development
ONS	Office for National Statistics
OPCS	Office for Population Censuses and Surveys
OR	Odds Ratio
PAC	Privacy Advisory Committee
PAR	Population Attributable Risk
PIL	Participant Information Leaflets
QPCR	Quantitative Polymerase Chain Reaction
RCPCH	Royal College of Paediatrics & Child Health
REC	Research Ethics Committee
RGSC	British Registrar General's Social Class
RII	Relative Index of Inequality
ROC	Receiver Operating Characteristic
SCS	Salimetrics Children's Swab
S-ECC	Severe Early Childhood Caries
SEG	Socio-economic Groups
SEM	Structural Equation Modelling
SEP	Socioeconomic position
SHBDEP	Scottish Health Boards' Dental Epidemiological Programme
SiC	Significant Caries Index
SIgA	Salivary IgA antibodies
SII	Slope Index of Inequality
SIMD	Scottish Index of Multiple Deprivation
SOC	Standard Occupation Classification
UK	United Kingdom
USA	United States of America
WHO	World Health Organization
WIC	Women, Infants and Children programme

Chapter 1: Introduction

The dental health of children in Scotland has shown continuous improvement in the past decade. Data from the National Dental Inspection Programme (NDIP) shows a sustained increase in the proportion of five-year-olds with no obvious decay experience, rising from 45% in 2002/3 to 68% in 2013/14 and a decrease in the mean number of decayed, missing and filled teeth ($d_3$mft), falling from 2.8 in 2002/03 to 1.3 in 2013/14 (NDIP, 2014). Similar trends have been observed among three-year-olds from the Greater Glasgow and Clyde NHS Board (McMahon et al., 2011). Furthermore, this improvement has been evident across the socioeconomic spectrum (McMahon et al., 2011, NDIP, 2014). The relative improvements in the caries rates across all social groups have largely been attributed to the national child oral health improvement programme, *Childsmile* (Macpherson et al., 2013). *Childsmile* adopts a life-course approach commencing in infancy with interventions applied universally, but in proportion to the level of disadvantage (Macpherson et al., 2010a, Marmot et al., 2010). Despite these improvements, large inequalities persist, with only 53% of the five-year-olds in the most deprived areas showing no obvious decay experience compared to 83% in the least deprived (NDIP, 2014). A clear gradient across all levels of deprivation remains (NDIP, 2014).

Dental caries has a complex multifactorial aetiology with the role of diet, oral hygiene, cariogenic bacteria and fluoride well established. However, the mechanism by which these factors act indirectly or interactively is not well understood. A systematic review of risk factors for caries in children called for more high quality, longitudinal studies using validated tools that took into account the impact of parental attitudes and beliefs across socioeconomic groups (Harris et al., 2004). Most studies on caries have almost exclusively focussed on either biological or behavioural factors (Watt, 2007). Studies that explain social gradients in health behaviours and the complex interlinking causal pathways which vary with time to understand ‘causes of the causes’ are few (Marmot and Bell, 2011). With the pathways between the various social factors not teased out, risk factors remain isolated, obscuring how they relate to one another and to caries (Watt, 2007, Sheiham et al., 2011).

Some efforts are underway in Australia, employing a longitudinal design and a multidisciplinary team to investigate risk factors in the development of caries in children to inform interventions (de Silva-Sanigorski et al., 2010). However Scotland is unique within its phase of improving oral health and evidence-based interventions, which comprise the *Childsmile* programme (Macpherson et al., 2010a). All children have the opportunity for daily-supervised toothbrushing in nurseries, fluoride toothpaste distributed for home use and targeted fluoride varnish programmes, home-support & clinical caries prevention interventions. Nevertheless, the prevailing gap in dental health between the most and least deprived identifies a need to investigate the independent effects and interaction of risk factors implicated.

This thesis aims to explore the feasibility of conducting a cohort study among young Scottish children, following them up one year later, to examine factors associated with dental caries and their role in explaining socioeconomic inequalities in dental health. Methods, tools, experiences and data collected from this study will be used to design a larger cohort study that will inform and evaluate the contribution of *Childsmile* interventions to oral health improvement, and reduction in oral health inequalities in Scotland into the future.

Chapter 2: Literature Review

2.1 Literature search strategy

An initial search was carried out on Ovid Medline 1996-2010 and Embase 1996-2010 to get an overall breadth of the dental caries inequalities literature using the search terms

i. [dental or oral or dental caries and infant* or toddler* or pre?school or young child*]
ii. [inequalit* or deprivation or deprived or disparit* or under privileged or poverty or socio?economic* or social class or low income*]
iii. i and ii were combined

A similar strategy using terms [aetiology* or etiolog or cause* or risk or diet or sugar* or sweet* or fizzy or life?style or oral hygiene or tooth?brushing or fluorid* or behavio?r*] were combined with the previous search to explore literature on risk factors; and inequalities related to risk factors in dental caries. Studies were limited to English language; and studies on fluorosis were excluded. Full texts of relevant studies assessed through titles and abstracts were accessed through the University of Glasgow library. Permanent alerts were set up for relevant authors using Ovid AutoAlert. The Cochrane library was searched for relevant systematic reviews and the bibliography of relevant papers were also used to identify papers missed through database searches. Additionally, dental public health textbooks and Google were used to undertake searches in the grey literature.

2.2 Dental Caries

2.2.1 The disease process

Dental caries is ‘the process of tooth decay’ (Johnson, 1991), characterised by localized demineralization and destruction of the hard tissues of the tooth (enamel, dentine and cementum) (Fejerskov and Kidd, 2008). It is caused by an imbalance in favour of demineralization in the demineralization-remineralization cycle within the oral cavity (Ismail, 2004).

Consumption of fermentable carbohydrates enable acidogenic bacteria in the oral cavity to produce organic acids, including lactic, formic, acetic, and propionic acids (Hicks et al., 2004). Some oral cariogenic species also synthesize intracellular polysaccharides that may be metabolized in the absence of exogenous carbohydrates for continued acid production (Balakrishnan et al., 2000). The acids produced lower the plaque pH to a point below the critical value of enamel dissolution (demineralization) resulting in diffusion of calcium and phosphate ions. Tooth enamel demineralizes at a pH below the 'critical pH'; the critical pH for demineralization varying between 5.2 to 5.5 among individuals (Dawes, 2003). The rate of demineralization depends on the absolute pH decrease as well as the length of time the pH remains below the critical pH. Demineralization initially presents as a white-spot lesion that is non-cavitated (i.e. macroscopically has intact enamel), but further exposure to reduced plaque pH results in sub-surface enamel softening (Featherstone, 2008) to form a carious cavity, showing a distinct discontinuity in the surface integrity (Fejerskov, 2006).

Remineralization is the body's natural repair process for sub-surface non-cavitated carious lesions (Featherstone, 2008). Calcium and phosphate ions primarily from saliva and other topical sources diffuse into the porous tooth surface and, with the help of fluoride, deposit a new veneer in a non-cavitated lesion (Featherstone, 2004). Additionally, fluoride binds to hydroxyapatite crystals to form fluoroapatite, which is relatively resistant to demineralization and also stimulates remineralization (Featherstone, 2004). Other factors that help remineralization by increasing the pH of the oral environment include the absence of substrate for bacterial metabolism, lowering the percentage of bacterial plaque, an increased rate of salivary secretion, an increased buffering capacity of saliva, antibacterial components of saliva and rapid food clearance (Hicks et al., 2004).

The cycle of demineralization-remineralization occurs numerous times daily in the oral cavity following intake of fermentable carbohydrates. The demineralization process may reverse through remineralization to completely heal the demineralized dental tissue or stop to preserve the damaged tissue (Ismail, 2004).

2.2.2 Nomenclature and definition

The term ‘dental caries’ or ‘caries’ is used to describe the signs and symptoms of a bacteria-mediated transmissible disease of multifactorial aetiology (Harris et al., 2004). With more evolving understanding of the disease process, it has been defined as a ‘continuum of disease states’ that increases in severity and tooth destruction ranging from ‘sub-clinical, sub-surface changes at the molecular level to lesions with dentinal involvement’ (Selwitz et al., 2007). The terms are used to identify both the disease process and the carious lesion (cavitated or non-cavitated) that is formed as a result of the process (Fejerskov, 2006).

Dental caries in primary teeth of young children before they begin school is commonly referred to as early childhood caries. Various terms and definitions have been used in the past to describe dental caries in primary teeth of young children, including ‘nursing bottle mouth’, ‘baby bottle tooth decay’, ‘nursing bottle syndrome’, ‘nursing caries’, ‘night bottle mouth’ or ‘bottle caries’ (Ismail and Sohn, 1999). After drawing criticism for the inconsistencies and failure to reflect the ‘multifactorial’ nature of the disease and misrelating the causal behaviour exclusively to improper feeding practices, the Centre for Disease Control and Prevention (CDC) workshop (1994) proposed the term, Early Childhood Caries (ECC) to describe all forms of caries in infants and preschool children up to 71 months of age (Drury et al., 1999).

The term, ECC faces objections in it not describing the characteristics of the condition, its rampant nature, risk factors, prevention (Vadiakas, 2008) and for it not being universally understood by parents (Narvey and Shwart, 2007). Additionally, agreement is yet to be reached on a universal case definition for epidemiological studies of ECC and those used have varied across epidemiological studies making international epidemiological data comparison difficult. Cases in this body of work were defined as the presence of visual caries into dentine (d_3 diagnostic threshold – ***See Section 2.2.3.2.1***) as used throughout the National Dental Inspection Programme (NDIP) and British Association for Study of Community Dentistry (BASCD) surveys (Pitts et al., 1997). This convention ensured comparability with other surveys within the UK and specifically excluded all carious surfaces with ‘white spot’ or ‘brown spot’ in enamel. As the case definition of ECC proposed by the National Institute of Dental and Craniofacial Research (NIDCR) (Drury et al., 1999) to standardize the collection and reporting of data in research projects includes non-cavitated lesions (including lesions in enamel presenting as demineralized white spots), which were not measured in this body of work, this thesis will use the term ‘dental caries’ when addressing caries in young (preschool and primary school) children throughout the review and when reporting results.

2.2.3 Measurement of dental caries

Dental caries is a dynamic disease, which clinically manifests in a number of forms (Featherstone, 2008). It is first observed as ‘a white spot’ in the enamel, which left unchecked advances to involve the dentine and later the pulp. The disease at the enamel and dentine level may present ‘non-cavitated’ or ‘cavitated’; or if treated; ‘filled/restored’ or ‘removed/extracted’. Caries measurement involves quantifying, grading and recording defined stages of the caries process (Pitts, 2004). Epidemiological quantification of the occurrence of such a dynamic disease in the population is governed by various criteria ***(See section 2.2.3.2.1)*** for the conditions under which caries is judged to be present. Dental caries requires to be measured by intensity and not by prevalence alone, since the latter cannot discriminate between degrees of severity. Caries experience, constituting past and present caries activity (treated or untreated) per individual is measured using the DMF index.

2.2.3.1 DMF/dmf index

The Decayed, Missing, Filled (DMF) index is the key measure of caries experience in dental epidemiology (Broadbent and Thomson, 2005). The index is applied to a Tooth as a whole- designated as DMFT, or applied per tooth Surface- designated as DMFS. A DMFT/S score for an individual is the arithmetic sum of the number of teeth/surfaces of teeth that are decayed (D), missing due to caries (M) and filled due to caries (F).

The DMF index when used for permanent teeth is always written in uppercase letters; the equivalent index for the primary dentition, ‘def’ index and its modifications, written in lowercase letters. The ‘e’ component of the def index

emphasises the exclusion of teeth that may have naturally exfoliated or are missing from trauma and only include teeth that are extracted due to caries (Cappelli and Mobley, 2008).
Modifications of the def index include
(a) dmf for use in children before ages of exfoliation,
(b) dmf applied only to the primary molar teeth after nine years
(c) the df index in which missing teeth are ignored, often used after exfoliation begins.

In the case of the mixed dentition when children are between six and ten years, DMF and def scores are computed separately and not added together.

The dmf/DMF is a cumulative caries experience index that indicates the total lifetime caries experience and its sequel of an individual. It is calculated for 20 primary teeth or 28 permanent teeth, excluding the third molars or according to the World Health Organization (WHO), modification for 32 permanent teeth (World Health Organization, 1997). Epidemiological studies of dental caries commonly present caries prevalence and severity as a population percentage with dmft=0 and as mean dmft value of all members of the population respectively.

Limitations
In spite of its widespread use and universal acceptance, owing to its simplicity and reliability when examiners have been trained (Fejerskov and Kidd, 2008), the DMF/dmf index has well-documented limitations (Spencer, 1997, Broadbent and Thomson, 2005). As a missing tooth makes it necessary to allocate a number of surfaces as having been decayed, researchers have been cautioned against estimation errors when computing DMFS score- assigning the maximum value for the 'M' component of DMFS leading to overestimation of an individual's caries experience. However, assigning a minimum value for the 'M' component causes an underestimation (Broadbent and Thomson, 2005). Broadbent and Thomson (2005) suggest assigning the same number of affected surfaces as recorded at the most recent examination before the tooth was lost to account for the 'M' component in longitudinal studies with short follow up period; and three surfaces for cross sectional studies (Broadbent and Thomson, 2005).

Additionally, overestimation is likely with the ‘F’ component as it carries the assumption that all filled surfaces were carious prior to the filling. However, evidence show wide variation in dentists' treatment decisions (Bader and Shugars, 1993, Bader and Shugars, 1997).

While mean dmf scores are frequently used to summarize severity of caries in a population, issues may arise when the sample of individual scores is positively skewed. This can obscure ‘tails’ of a distribution, making the mean dmf value not accurately reflect the burden of disease as a population descriptor (Campus et al., 2003, Armfield, 2005). This is so especially in developed countries where the typical dmf distribution is positively skewed and has a high proportion of zero scores (Marthaler, 2004, Armfield, 2005, Ditmyer et al., 2011). The use of the Significant Caries index (Bratthall, 2000), zero-inflated Poisson and zero-inflated negative-binomial modelling have been recommended in overcoming the skew (Ditmyer et al., 2011, Lewsey and Thomson, 2004). Further limitations of the dmf index include its inability to account for sealed teeth and its limited use for estimating treatment needs, as criteria used to diagnose caries in epidemiological surveys are usually different from those used by dentists in formulating a patient’s treatment plan (Fejerskov and Kidd, 2008, Honkala et al., 2011). Assessing treatment needs for a population based only on DMFT, without radiographs has shown to underestimate the need for restorative care by as much as 44%, particularly due to early proximal caries (Becker et al., 2007).

2.2.3.2 Caries detection criteria

Caries detection involves an objective method of determining whether or not disease is present (Pitts and Stamm, 2004). With progress in the understanding of the caries process, the original dmf index that scores ‘d’ only at the level of cavitation has led way to various caries detection systems which record caries at various stages, from the earliest enamel caries through to cavitation; under varying examination conditions. The degree of generalizability of the epidemiological caries data available on a global basis depends on the diagnostic thresholds and criteria employed, as they show a significant impact on the estimates of caries prevalence and severity.

2.2.3.2.1 Caries diagnostic thresholds & Detection criteria for recording caries

'Diagnostic threshold' is a term that describes the cut-off level used in making a decision of what is classified as 'diseased' or 'sound' (Pitts, 2004). The caries diagnostic threshold has been excellently represented in the form of an iceberg by Pitts (2004) (Appendix A). Clinically detectable lesions are graded from 'd_1'- enamel caries lesions with intact surfaces to 'd_4'- lesions extending into the pulp. The peak of the iceberg represents gross or frank dentine caries- the d_4 and more limited d_3, caries into dentine lesions. Resting below is less extensive d_2- cavitated enamel lesions and further limited d_1- white- or brown- spot enamel caries lesions with intact surface.

Caries epidemiological studies have traditionally recorded caries at the d_3 (caries into dentine) diagnostic threshold. It includes d_3 and d_4 grades of lesion and excludes other less extensive d_2 and d_1 lesions; and early signs of caries. This threshold is followed by the WHO criteria (World Health Organization, 1997), Radike criteria in the USA (Radike, 1968) and diagnostic criteria for surveys in the UK (Pitts et al., 1997). In the USA, and other countries that adopt the WHO criteria (World Health Organization, 1997), epidemiological recording of caries at the d_3 level takes place only at macroscopic cavitation. However, in 1992 the British Association for the Study of Community Dentistry criteria for assessing caries in the UK were amended to also include visual caries at the d_3 level (non-cavitated dentine lesions) (Pitts et al., 1994). The proportion of the population classified as 'caries free' in studies where data are collected and/or reported at the d_3 threshold warrants caution. It has been suggested the term 'no obvious caries experience' be used as using 'caries free' may mistakenly convey complete absence of disease (Pitts et al., 1997).

The rationale for recording only dentinal lesions at the d_3 threshold has primarily been justified on the grounds of better examiner reproducibility as differences in clinical opinion may be marked in the case of non-cavitated lesions (World Health Organization, 1997). It has also been reported that clinical examination of non-cavitated lesions under 'field conditions' may be challenging compared to a 'dental operatory', which may result in inconsistent data (Burt, 1997). Furthermore, some early carious lesions may not always progress (lesion arrest) or even reverse and recording these lesions may overestimate caries experience in a population (Neilson and Pitts, 1991). However, the need for detecting and measuring non-cavitated enamel lesions has been argued and proposed over many years for better estimates of disease, improving possibility for successful preventive intervention, assessing change in caries status and predicting future caries (Ismail and Sohn, 1999, Ismail, 2004). It is recommended that non-cavitated lesions are detected and monitored in studies that involve the natural history of caries and its treatment- clinical trials (Pitts and Stamm, 2004).

More recent systems developed for recording caries, like the International Caries Detection and Assessment System (ICDAS), record caries at various levels of severity from early enamel lesions to those involving the pulp (Ismail et al., 2007). This system requires cleaning of the tooth surface (using toothbrush or prophylaxis cup) and the use of compressed air to dry teeth prior to visual inspection, which is difficult under field conditions. While the ICDAS criteria has been feasibly applied in epidemiological surveys of dental caries in young children and clinical trials (Braga et al., 2009), the mean examination time has been reported to be almost twice as long as when using the WHO criteria (World Health Organization, 1997). Although inter examiner reliability has been reported to be higher using the WHO criteria, the ICDAS has given acceptable kappa values in the primary dentition (Braga et al., 2009, Ismail et al., 2009). Some other approaches to caries measurement differentiate between actively progressing and inactive carious lesions (Ekstrand et al., 1998, Nyvad et al., 1999).

This body of work focussed on caries in the primary dentition of young children. The review from this point forward will reveal the literature in relation to caries in young children, particularly five to six years of age for which representative data is available.

2.2.4 Prevalence and severity of dental caries in the primary dentition- Global

Although there have been numerous epidemiological surveys conducted in young children below the age of five to six years worldwide, few can be considered national with large samples representative of the population. Additionally, the epidemiological data on the dental health of younger age groups (two to four-year-olds) have not been documented to the same extent as the dental health of school-children, mainly due to the difficulty in examining (Tinanoff and Reisine, 2009) and accessing these age groups for data collection (Vadiakas, 2008). Furthermore, available data are often grouped into broad age categories and direct comparison of the numbers reported across studies/surveys is difficult due to varying caries diagnostic criteria, calibration procedures, examination conditions and indicator age groups in individual countries.

Most studies reviewed in the following sub-sections were epidemiological data that collected data at the d_3 threshold within their definition. However, given the diversity in reporting caries among young children, the definition of 'dental caries' and diagnostic threshold as described in the study will be quoted if otherwise.

2.2.4.1 Geographic

The extent of epidemiological data on the dental health of young children varies widely between countries. While some countries have repeated cross-sectional data of representative samples, many others have little or out-dated data (Pitts et al., 2011). Overall, dental caries stands as the most common chronic disease of childhood (Gussy et al., 2006, Centers for Disease Control and Prevention, 2012). The highest rates are reported from a few developing countries, particularly those of the South East Asian continent like Thailand and Macau, with caries affecting more than 85% of the five to six-year-olds and showing a high severity (mean dmft) of 6.0 and 4.5 respectively (World Health Organization, 2012). Small areas of high prevalence have also been reported within developed western countries. According to the most recent Australian Child Dental Health survey, caries affects 67% of the five to six-year-olds in the northern territory with a dmft of 3.8 (Mejia et al., 2012). Across Europe, there is wide between-country variation in caries prevalence reported, with some of the highest rates recorded in Central and Eastern Europe, and generally lower rates in Western European countries (Marthaler, 2004). In the United States, the prevalence of dental caries was reported to be 40% among five-year-olds in the National Health and Nutrition Examination Survey (NHANES) (Vargas et al., 2014).

2.2.4.2 Race/Ethnicity

Significant variation in the distribution of caries by race/ethnicity have been noted from some countries, with native (indigenous) young children suffering from poorer dental health (Parker et al., 2010). The majority of these studies are reported from the USA: while 26% of the non-Hispanic white young children showed caries experience, that percentage increased to 43% among Mexican-American children and 29% among non-Hispanic black children in the NHANES (Vargas et al., 2014). Minority children have also presented increasing severity of the disease; the dft in the NHANES being 0.7 for non-Hispanic whites, 1.0 for non-Hispanic blacks and 1.7 for Mexican-Americans (Vargas et al., 2014). Similar variation in dental health status has been reported from regional studies of California; with Hispanic Asian and Latino children, independent of socioeconomic position, showing the highest prevalence of caries (30%) compared to their peer non-Hispanic whites (13%) (Shiboski et al., 2003). The American Indian and Alaska Native (AI/AN) children demonstrate the highest prevalence of caries ever documented in the US (Douglass et al., 2003): the prevalence in a recent community-based sample of five-year-olds from the population was 75%, with a mean dmft of 5.7 (Phipps et al., 2012). Other studies using convenience or state level samples have demonstrated similar higher levels among AI/AN young children than their counterparts from other racial/ethnic backgrounds (Tang et al., 1997, Hardison, 2006, Klejka et al., 2011). It has also been documented that the families of these children live relatively isolated, with lower levels of educational attainment and in greater deprivation (Phipps et al., 2012). However, an investigation of the underlying causes and how much of it might possibly be socioeconomically related remains unexplored.

Other countries reporting similar findings include Australia, New Zealand and Canada (Peressini et al., 2004, Parker et al., 2010, Christian and Blinkhorn, 2012). Aboriginal five-year-olds in Australia and Canada are recognised as having, on average, more than twice the caries experience as other non-Aboriginal children (Jamieson et al., 2007, Schroth et al., 2007), with caries levels in some communities being five times (Australian Institute of Health and Welfare, 2011b, Christian and Blinkhorn, 2012) and a mean dmft over three times (4.3 compared to 1.9) that of non-Indigenous children of the same age (Jamieson et al., 2007). Similar findings have been reported among first generation infants born to migrant mothers in European countries (Ferro et al., 2007). Social exclusion of minority people has been theorized as one possible explanation of the observed significant differences by race/ethnicity (Marmot, 2005).

2.2.4.3 Socioeconomic position (SEP)

Regardless of the case definition used, there is overwhelming evidence of caries levels being higher in socioeconomically deprived children, irrespective of race/ethnicity or culture, in both developed and developing countries (Bernabé and Hobdell, 2010, Marmot and Bell, 2011).

Among developed countries, dental caries in children have been reported to be closely associated to income inequality (Bernabé and Hobdell, 2010, Do et al., 2010). In the US, young children from disadvantaged backgrounds suffer more than twice the caries experience of their more affluent peers: 39.7% compared to 17.2%, when SEP was measured by parental educational attainment (Dye et al., 2004). Additionally, children living below the poverty line (annual income of $17,000 for a single family of four) have presented with increasing severity of the disease and these differences follow into adolescence (Bagramian et al., 2009). The greatest difference in caries prevalence among SEP groups in NHANES III were seen in children showing six or more carious surfaces, 11% of all children below the poverty line compared to 2% of those above the poverty line. Similarly, five to six-year-olds from deprived areas of Australia show a higher prevalence of caries (54.3%) than their affluent peers (39.3%), with a mean dmft of 2.6 and 1.5 respectively (Australian Institute of Health and Welfare, 2011a). Comparable findings have been reported in New Zealand (Thomson et al., 2004) and some European countries (Campus et al., 2009, Oulis et al., 2012), with children from deprived backgrounds carrying a disproportionate burden of caries experience.

2.2.5 Annual caries prevalence studies for children- UK/Scotland

In the UK, dental epidemiological surveys are regularly undertaken and documented to establish the state of the dental health of children, and examine trends over time. These surveys use the visual examination method developed by BASCD that assess caries at the d_3 threshold (Pitts et al., 1997).

2.2.5.1 The Children's Dental Health Survey

The Children's Dental Health Survey in the UK has been undertaken decennially by the Office for National Statistics (ONS) (formerly the Office for Population Censuses and Surveys (OPCS)). The survey commissioned by the four United Kingdom Health Departments has been carried out since 1973 in England and Wales and since 1983 in the whole of the UK including Scotland and Northern Ireland (Pitts et al., 2006). However, Scotland did not participate in the most recent survey of 2013. The survey provides information on the dental health of children in the United Kingdom with a representative sample of children aged 5, 8, 12 and 15 years of age attending Local Authority and independent schools in the UK. In addition, it provides information on children's oral hygiene, experiences of dental care and treatment; and measures changes in oral health since previous surveys (Pitts et al., 2006).

2.2.5.2 UK annual caries prevalence studies of children coordinated by BASCD

The British Association for the Study of Community Dentistry (BASCD) in association with the National Health Service (NHS), co-ordinates annual caries prevalence studies of children in the UK. It includes data from Scotland, collected under the National Dental Inspection programme (NDIP) ***(See Section 2.2.5.3)***. They commenced in 1985/6 across Wales and parts of England (Dowell, 1988), followed by Scotland in 1987 and Northern Ireland in 1994 (Nugent, 1997). Whilst the ONS surveys examine a relatively smaller number of children once in 10 years and report data on a range of oral health indicators at country level, the BASCD surveys report the prevalence and severity of dental caries from examining larger numbers of five to six and 11 to 12-year-old children annually. Consequently, they provide detail at a local and regional level (Pitts et al., 2007). However, limited comparison of results is possible with Scotland since 2007/8 following the introduction of positive parental consent (opt-in) in England and Wales (Davies et al., 2011).

2.2.5.3 National Dental Inspection programme (NDIP)

Since 1987, caries prevalence surveys have been undertaken in Scotland, first by the Scottish Health Boards' Dental Epidemiological Programme (SHBDEP) and then in 2003 by the National Dental Inspection programme (NDIP). The NDIP collects dental data annually using a Basic Inspection intended for all children of five and eleven years of age attending Local Authority schools and a Detailed Inspection for a representative sample of one of the aforementioned specific age groups in alternate years (NDIP; 2014). Passive parental consent (opt-out) underpins participation in the NDIP since its beginning.

The detailed inspection determines caries experience at the d_3/D_3 threshold, levels of oral hygiene and additionally illustrates the impact of deprivation on dental health (NDIP, 2014). The data can also be used for evaluating existing oral health improvement initiatives and highlighting areas requiring further work. The NDIP programme protocol allows local NHS Boards to undertake supplementary dental inspections for additional age groups as necessary within the population. Consequently, the Greater Glasgow and Clyde NHS Board (NHSGGC) has added an additional age group, three-year-olds attending nursery, to the standard NDIP schedule of dental inspections since 2006/7 (McMahon et al., 2011).

2.2.6 Prevalence and severity of dental caries in the primary dentition- UK/Scotland

Pitts & Topping (2005) reviewed the UK dental health trends among five-year-olds from 1973 to 2004 and concluded significant statistical and clinical reductions in caries from 1973 to 1983, that slowed in the 1990s and halted by 2003. However, a pattern of true continuous steady progress in dental health has been reported in Scotland since 2003 (Davies et al., 2011, NDIP, 2014). In England and Wales, the consent arrangements for the surveys changed in 2007/8 and the resultant response bias renders datasets not directly comparable with the Scottish data (Davies et al., 2011).

2.2.6.1 Scotland in recent years

In Scotland, throughout the time of SHBDEP, more than 50% of the five-year-olds showed obvious caries experience, the highest being 61.8% in 1993 and the lowest being 54.9% in 1999, with mean $d_3$mft of 3.20 and 2.55 respectively (Pitts et al., 2000, NDIP, 2014). Improvements in dental health over this period had slowed down and the distribution of caries was markedly skewed, indicating regional geographic and socioeconomic inequality (Sweeney et al., 1999, Pitts et al., 2000). In its final report in 2000, the Grampian Health Board showed a mean $d_3$mft of 1.89, whilst that of Glasgow was 3.51 (Pitts et al., 2000).

Scotland has come a long way since then with both the prevalence and severity of dental caries among five-year-olds having significantly reduced over time (NDIP, 2014). The proportion of five-year-olds with obvious caries experience, has fallen from 55.4% in 2002/03 to 32% in 2013/14 and the respective mean $d_3$mft, declining from 2.8 to 1.3 (NDIP, 2014). Identical trends have been reported among three-year-olds in Greater Glasgow with those showing obvious caries experience being 26% in 2006/7 and 17% in 2009/10 and a mean $d_3$mft of 1.1 and 0.4 respectively (McMahon et al., 2011). Additionally, this improvement has been evident across the socioeconomic spectrum (McMahon et al., 2011, NDIP, 2014). The relative improvements in caries rates across all socioeconomic groups have largely been attributed to the Scotland-wide child oral health improvement programme, *Childsmile* ***(See Section 2.3.9.1)*** (Macpherson et al., 2013). However, a gradient in the distribution of caries towards socioeconomically deprived children persists. A recent study also reported better dental health among five-year-olds in remote and rural areas of Scotland when compared with those living in cities (Levin et al., 2010). The prevalence of caries in cities was 46.5% and in remote rural areas was 34.7%, with a mean $d_3$mft of 2.2 and 1.4 respectively. Nonetheless, this difference became non-significant after adjusting for area-based deprivation.

Despite dental health improvements, more than a quarter of Scottish five-year-old children still suffer from caries and the distribution is skewed towards those from deprived backgrounds. Although largely preventable, it remains the most prevalent chronic disease of childhood compared to obesity and asthma (ISD Scotland, 2013a). Individuals remain susceptible to the disease throughout dentate life and for those affected, it inflicts a considerable social and economic burden with implications for public health policy.

2.2.7 Impact of caries in children

Caries affects the wellbeing and quality of life in children by causing pain, disturbed speech and eating patterns and absence at school (Sheiham, 2006). It has also been shown to affect nutrition and growth, particularly among young children (Floyd, 2009, Watt and Rouxel, 2012). Furthermore, premature loss of primary teeth can affect the permanent occlusion, impacting appearance, self-esteem and confidence.

Caries inflicts a significant burden on health care services with a total of £65 million spent on children's dental care in Scotland in 2013 (ISD Scotland, 2013c). Treatment for severe caries may involve extraction of teeth, which remains the most common reason for Scottish children to be admitted to hospital for an elective procedure. While there has been a reduction in general anaesthetics use for dental extractions in the past few years, 1810 children aged up to four years have been reported to have had teeth extracted under general anaesthesia (GA) in 2010/11 (ISD Scotland, 2012). Children referred for dental care under GA due to caries have been shown to have a high need for retreatment (MacCormac and Kinirons, 1998). In addition, such extractions can lead to risks of GA, development of dental anxiety and phobias, resulting in poorer dental attendance in adulthood (Hosey et al., 2006, Nunn, 2006). In the longer run, children who experience caries are likely to develop further new carious lesions in both the primary and permanent dentitions (Ismail et al., 2009).

2.2.8 Risk factors for dental caries

2.2.8.1 The terminology

Risk factors are defined as environmental, behavioural, or biological factors confirmed by temporal sequence, that when present, directly increase the probability of a disease and when absent or removed reduce the probability of the disease occurring (Beck, 1998). They are either part of the causal chain or expose the host to the causal chain (Beck, 1998, Burt, 2005). Strictly speaking, a risk factor must establish that the exposure has occurred before the outcome. Thus, prospective studies are necessary to establish risk factors for dental caries, while a cross-sectional study can only provide evidence of 'risk indicators'. The term 'risk indicators' is used to differentiate factors that are putative risk factors (Beck, 1998). Although the definition does not specify whether a risk factor has to be modifiable, general usage implies it does (Burt, 2005). The term, 'demographic risk factors', previously called 'background characteristics' have often been used for non-modifiable risk factors that are social and demographic characteristics of age, gender, and race/ethnicity (Beck, 1998).

2.2.8.2 Caries risk factor models for children

Conceptual models describe various potential influences on dental health and help in understanding the wider determinants that cause disease. Several theoretical models have been proposed conceptualizing the influences on the dental health of young children (Holst et al., 2001, Pine et al., 2004b, Fisher-Owens et al., 2007, Seow, 2012). All models suggest a multilevel approach to describing the complex causal pathways between social structure and health, interlinking material, psychosocial and behavioural pathways **(*See section 2.3.6.1*).** The comprehensive model, based on a thorough review of major population and oral health literatures by Fisher-Owens et al. (2007) depicted the complex interplay of an extensive list of factors acting at multiple levels including the effect of time on the oral health of children. The model included child, family and community-level influences, incorporating five key domains, namely: genetic and biological factors, health behaviours, dental/medical care, social and physical environment (Figure 2-1). However, the model does not specify pathways by which factors are linked to child oral health. The authors called for testing the model empirically, acknowledging the necessity of perhaps simplifying the model. An adequately powered prospective cohort study with a conceptual disease model combining qualitative and quantitative research methods and sophisticated statistical modelling techniques (multilevel modelling, path analysis or structural equation modelling) has been advocated in disentangling the mechanism underlying the social gradient in caries (Newton and Bower, 2005, Aleksejūnienė et al., 2009).

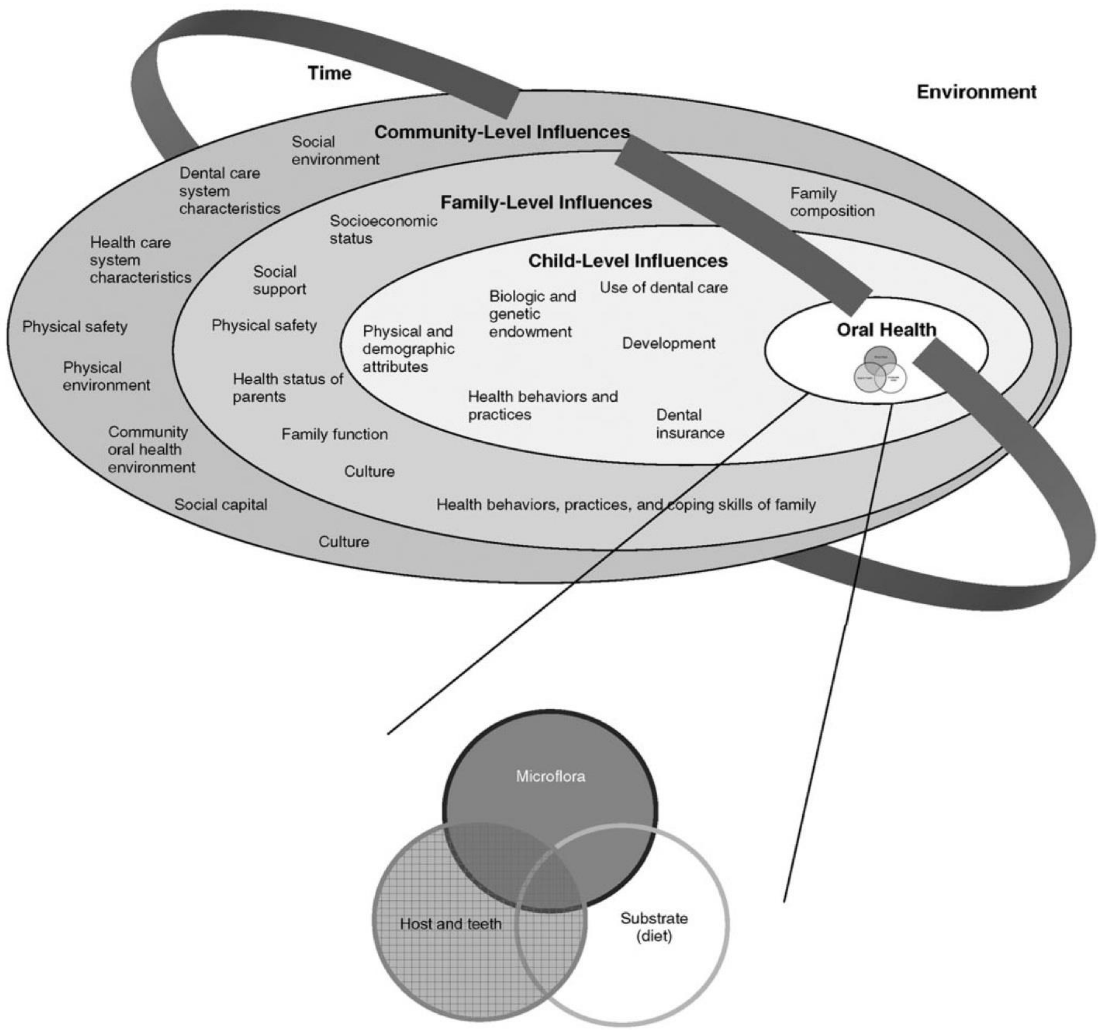

Figure 2-1: Conceptual model depicting the multilevel influences on oral health (Fisher-Owens et al., 2007)

2.2.8.3 Risk factors for dental caries among young children

Risk factors for dental caries are multiple and interrelated which makes the disease complex and the literature extensive. Those factors implicated in the initiation and progression of dental caries in young children have been reviewed systematically (Harris et al., 2004) and in a number of other narrative reviews (Gussy et al., 2006, Vadiakas, 2008, Selwitz et al., 2007).

While the main factors involved in the disease initiation and progression are cariogenic bacteria, fermentable carbohydrates, a susceptible tooth/host and time, a systematic review of risk factors associated with caries in children under six years of age found 106 factors related to caries risk (Harris et al., 2004). The numerous factors were categorised under oral flora, dietary, infant feeding, oral hygiene, socio-demographic and other factors. The risk of caries thus being sensitive to the interaction of multiple factors translating a complex disease aetiology (Harris et al., 2004).

A thorough review of the literature on all of the risk factors and caries is beyond the scope of the present body of work. However, risk factors that have shown consistent evidence will be reviewed briefly in the following sub-sections in the order they are hypothesized to appear in a conceptual model, beginning with proximal child-level influences followed by family-level influences (Figure 2-1) (Fisher-Owens et al., 2007).

2.2.8.3.1 Early Colonization by Cariogenic Bacteria

The oral cavity of a new-born is a sterile environment, but within two days it begins to get colonized by various microbial species (Pearce et al., 1995). Colonization is thought to be due to direct transmission from the main carer, usually the mother (Li et al., 2005).

Mutans Streptococci

The presence of mutans streptococci, namely *Streptococcus mutans (S. mutans)* and *Streptococcus sobrinus (S.sobrinus)* in plaque and saliva have been used as an indicator of caries risk (Parisotto et al., 2010). *S. mutans* has been associated with initiating dental caries, while *S. sobrinus* is thought to progress the lesion (Parisotto et al., 2010).

A systematic review of risk factors associated with caries in children under six years of age found consistent evidence of a young child being most likely to develop caries if they acquired *S. mutans* at an early age (Harris et al., 2004). A second systematic review investigated *S. mutans* count in plaque and saliva as a risk factor for caries in young children, aged two to five years (Thenisch et al., 2006). In contrast to the Harris et al. (2004) review, a quantitative synthesis and meta-analysis (where possible) was carried out. The results showed a pooled risk ratio and [95% confidence interval (CI)] of 2.11 [1.47 to 3.02], indicating higher salivary *S. mutans* counts to be associated with a considerable increase in caries risk. A more recent systematic review that investigated the relationship between *S. mutans* and childhood caries concluded it as a strong 'risk indicator' for caries in young children (Parisotto et al., 2010). However, all of the reviews called for well-designed longitudinal studies to confirm mutans streptococci levels as a 'risk factor'(Parisotto et al., 2010) in childhood caries using validated measures (Harris et al., 2004, Thenisch et al., 2006), with appropriate adjustment for potential confounders (Thenisch et al., 2006).

Poor socioeconomic conditions have been identified as a risk factor for early colonization with *S. mutans* (Li et al., 2005). Additionally, young children (five to six-year-olds) from poorer socioeconomic backgrounds have been shown to have cariogenic bacteria (mutans streptococci (MS) and lactobacillus species (LB)) at levels significantly higher than their more affluent peers (Boyce et al., 2010). The counts of these bacteria were found to substantially mediate the social gradient in caries (Boyce et al., 2010).

Other bacterial species

Molecular-based studies of caries have documented caries in the absence of *S. mutans* (Becker et al., 2002, Aas et al., 2008). Additionally, *S. mutans* have been detected in the absence of caries (Radford et al., 2000; Wan et al., 2001). A variety of bacterial species other than *S. mutans* present in the oral cavity can produce acids from fermentable carbohydrates, such as *S. gordonii, Veillonella spp., Actinomyces spp., Bifidobacterium spp., S. mitis, S. oralis* and *S. anginosus* (Becker et al., 2002). These studies highlight the complexity in the oral microflora associated with caries initiation and progression.

Limited studies have investigated how the immune system deals with the complex microflora associated with caries in young children (Malcolm, 2013). Antimicrobial peptides together with other innate and adaptive immune mediators like secretory immunoglobulin (sIgA) in saliva may act as a protective factor against cariogenic bacteria (Malcolm, 2013).

2.2.8.3.2 Oral Hygiene Practices and the Role of Fluorides

Reduced frequency of toothbrushing is known to be a key behavioural risk factor in caries. It is recommended that children brush their teeth at least twice daily using a fluoride toothpaste as soon as teeth erupt (SIGN, 2014) as fluoride controls the initiation and progression of caries (Marinho et al., 2003). Other significant risk indicators with respect to oral hygiene practices that have shown associations to caries in young children include higher age at commencement of regular toothbrushing (Sheehy et al., 2008), unsupervised toothbrushing (Marinho et al., 2003), rinsing with water after brushing (Sjögren et al., 1995) and the use of a non-fluoridated over a fluoride toothpaste (Marinho et al., 2003).

Interactions between oral hygiene and diet are known to affect bacterial cariogenic potential. Harris et al. (2004) point to the interaction between the two factors so that if there is a balance between the deleterious effects of sugar consumption and the benefit of toothbrushing with fluoride toothpaste, the development of caries may be controlled. This is in line with the findings of a systematic review of observational studies from 1980 to 2000, investigating the relationship between dental caries and sugar consumption (Burt and Pai, 2000). The review found that the relationship between dental caries and total sugar consumption or frequency of sugar consumption was less strong particularly when there was adequate fluoride exposure (Burt and Pai, 2000).

In Scotland, toothbrushing frequency has been reported to differ by gender and SEP, with girls (74% among girls vs 63% among boys) and family affluence (80% in the least deprived areas vs 55% in the most deprived) associated with a higher frequency of toothbrushing (Maes et al., 2006, Bradshaw et al., 2008, Masson et al., 2010). Younger children have also been reported to brush their teeth more frequently than older children (76% in three to seven-year-olds vs 63% in 12 to 17-year-olds) (Bradshaw et al., 2008, Masson et al., 2010). This may be credited to the *Childsmile* core component that promotes toothbrushing at home, in nurseries and some schools. Socioeconomic inequalities in toothbrushing may no longer be so evident among this youngest age group as a reduction in absolute inequalities have been reported, with the mean $d_3$mft reducing by 1.71 in the most deprived children and by 0.43 in the least deprived after the roll out of the toothbrushing programme (Macpherson et al., 2013).

The Role of Fluorides

Fluoride has a protective effect on teeth, through its ability to remineralize enamel and modify the metabolism of plaque bacteria by inhibition of acid production and reduction of extracellular polysaccharides (Bowden, 1990). Cochrane reviews have demonstrated the effectiveness of brushing with fluoride toothpaste for preventing caries among children and its effective use as part of a dental public health programme (Marinho et al., 2003, Walsh et al., 2010b), with additional effectiveness in high caries incidence populations from supervised brushing, higher frequency of use and higher concentrations of fluoride (Walsh et al., 2010). It is recommended that children use a toothpaste with fluoride concentrations of 1000 to 1500 ppm, with children at increased risk of developing caries advised to use a toothpaste with higher concentrations of fluoride (SIGN, 2014).

Additional benefits have also been demonstrated in relation to using other forms of topically applied fluoride preparations like gels, varnishes and mouth-rinses, in combination with fluoride toothpaste in children (Marinho et al., 2003a). A recent systematic review of randomised controlled trials has confirmed the substantial caries preventive effect of fluoride varnishes in children, in both permanent and primary dentition (Marinho et al., 2013). Consequently, the SIGN (2014) guidelines recommend fluoride varnish applications at least twice yearly in all children. The Scottish Government (2010) set a **H**ealth Improvement, **E**fficiency, **A**ccess to Services and **T**reatment **(**HEAT) target aiming to achieve at least two applications of fluoride varnish per year in 60% of three and four-year-olds in each SIMD quintile by March 2014. The overall levels of Scottish three and four-year-olds receiving at least two fluoride varnish applications in 2013 were 20% and 26% respectively (ISD Scotland, 2013a).

There has been some debate over potential risks of fluorosis among young children (Stookey, 1994). However, a recent Cochrane review, assesing the risks of fluoride toothpastes among children under six years of age found no evidence for frequency and amount of fluoride toothpaste used and fluorosis. Weak, but ‘unreliable’ evidence of an increased risk of fluorosis was found for starting the use of fluoride toothpaste in children under 12 months of age. Given the caries-preventive effects of fluoride toothpastes to increase with increasing concentrations of fluoride concentration, the review recommended use of toothpastes with fluoride concentrations <1000 ppm among young children when the development of fluorosis was a prime concern (eg: when residing in an area with fluoridated water supply) (Wong et al., 2011).

2.2.8.3.3 Diet

Diet, particularly those rich in sugars, plays an important role in the aetiology of dental caries as it determines the level of bacterial activity in the plaque (Watt and Rouxel, 2012).

Types of sugars

The term ‘sugars’ includes all mono and disaccharides, the most common being sucrose, glucose, fructose, maltose and lactose (Moynihan, 1998). In the UK, dietary sugars are classified as intrinsic sugars (sugars within the cellular structure of foods) as in fruits and vegetables, and extrinsic sugars (sugars located outside the cell structure); including Milk sugars and Non-milk extrinsic (NME) sugars (Department of Health, 1989). NMES include fruit juices, honey and added sugars (table sugars and recipe sugars).

Non-milk extrinsic (NME) sugars

NMES are more readily metabolised by oral bacteria than intrinsic sugars and milk sugars; and are implicated as a causative factor in dental caries, obesity and diabetes (Department of Health, 1989, Watt and Rouxel, 2012). Consequently, The Department of Health (1989) and more recently, the revised Scottish Dietary Goals (The Scottish Government, 2013) recommend no more than 11% of total energy intake (approx. 33 g per day for young children (Watt and Rouxel, 2012)) shall come from NMES. However, a systematic review of the literature from 1995 to 2006, looking at the harmful effects of sugar intake on a number of health conditions, gave mixed evidence and no clear associations between NMES and most chronic health conditions. The reviewers did not support a single quantitative sugar guideline covering all health issues (Ruxton et al., 2009).

Relationship between sugars and dental caries

Anderson et al. (2009) systematically reviewed papers published from 1856-2007 to determine any relationship between sugar consumption and dental caries. Out of the 31 valid studies that were included in the final analysis, six studies showed a positive significant association between sugar quantity and dental caries, and 19 provided evidence of an association between frequency of sugar consumption and dental caries. It was concluded that the relationship of sugars to caries is present for frequency, but weak. More recently, a systematic review and meta-analysis, to inform the updating of the WHO sugar consumption guidelines, analysed all available published data from 1950 relating to the amount of sugar consumption and levels of dental caries for both adults and children. The review identified ‘moderate quality evidence’ supporting a relationship between the amount of sugar intake and the development of dental caries across age groups. Furthermore, they recommended limiting intake of NMES to <5% energy intake to reduce the risk of caries (Moynihan and Kelly, 2014). However, this recommendation did not follow a reliable evidence of effect from intervention studies. The evidence backing the recommendation came from three Japanese population surveys, published in 1959 and 1960, showing lower caries levels in children who consumed less than five percent of energy intake in the form of sugar. Additionally, all three studies were in populations with low fluoride exposure. Although meta-analysis was limited, the analysis indicated a large effect size for the impact of sugar intake on dental caries; standardized mean difference for DMFT being 0.82 (95% CI 0.67 to 0.97), and when measured as caries prevalence the risk ratio being 7.15 (95% CI 2.82 to 8.14) (Moynihan and Kelly, 2014). Nevertheless, Ruxton et al. (2009)’s systematic review (as described previously) found that the combination of sugar amount/frequency, fluoride exposure, and food adhesiveness were more reliable predictors of caries risk than the amount of sugar alone.

NMES intake among children in Scotland

There is a high intake of NMES among Scottish children. The latest survey of sugar intake among children in Scotland (Sugar Survey) commissioned by the Food Standards Agency Scotland (FSAS) in 2010 found a high mean intake of NMES (Masson et al., 2012). Fifteen percent of daily food energy was derived from NMES among three to seven-year-olds; considerably higher than the recommended population averages (Masson et al., 2012). The same survey in 2006 showed children who reported having received treatment for dental decay had significantly higher intakes of NMES (Masson et al., 2010). Additionally, it showed self-reported treatment for dental decay to increase with increasing deprivation in both sexes aged three to 17 years (Sheehy et al., 2008). The Growing Up in Scotland (GUS) study of four to five-year-olds reported similar findings, with younger mothers, single mothers, mothers with less educational qualifications and those with lesser household incomes, more likely to report their children eating sugary snacks and soft drinks (Bradshaw et al., 2008). However, the concurrent association of socio-economic position with the risk of developing dental caries, considering all of the behavioural risk factors, has been understudied.

The relationship between diet and dental caries in young children is complex and confounded by many variables, such as mutans streptococci, oral hygiene behaviours, fluoride exposure and modified by socioeconomic circumstances. Although evidence suggests most of the risk factors being socially patterned, fewer studies have studied how the interactions between different risk factors change along the socioeconomic spectrum. In addition to the frequency and amount consumed; physical consistency of the food that influences the oral clearance rate, intake pattern (in between snacking) and timing are other important factors associated with caries (Marshall et al., 2005, Marshall et al., 2007).

Other dietary factors

Snacking increases the numbers of eating events in a day which in turn has been associated with higher rates of dental caries, particularly when snacks are high in NMES (NHS Health Scotland, 2012). In Scotland, analysis of 156 children aged five to 17 years who completed a 4-day non-weighed diet diary in the Sugar

Intake Survey 2006 showed the median number of snacks per day was two, with 98% of children having a snack and 77% snacking 'biscuits, cakes and pastries', which are high in NMES (Macdiarmid et al., 2009). Frequent snackers had higher daily intakes of NMES; with thirty nine percent of the NMES intake accounted for by snacking and this did not vary by sex, age, body mass index (BMI) and socioeconomic circumstances (measured by SIMD- ***See section 2.3.2.2***) (Macdiarmid et al., 2009).

The GUS study of four to five-year-olds reported a quarter of the children (23%) eating crisps at least once daily (Bradshaw et al., 2008). Although 'crisps and savoury snacks' contributed less than one percent to NMES intake, children in the Sugar Survey aged three to 17 years, with medium/higher intake of these snacks were reported to have significantly higher risk of having received treatment for decay (Sheehy et al., 2008). These foods are sticky and leave residue on teeth that prolongs exposure to carbohydrate. Notwithstanding this, children from more deprived areas have been reported to derive a higher proportion of energy from crisps and savoury snacks than children from less deprived areas (Sheehy et al., 2008).

2.2.8.3.4 Early feeding practices

While breastfeeding is well evidenced to provide numerous health benefits to infants and mothers (Salone et al., 2013); prolonged on-demand and night time breastfeeding has been reported to be a potential risk factor for caries in young children (Harris et al., 2004). However, systematic reviews that investigated the risk of developing caries in infants with continuation of breastfeeding beyond six months have shown no evidence of an independent association (Valaitis et al., 1999, White, 2008). The evidence is inconsistent, and is primarily based on cross-sectional studies and a few longitudinal studies that have failed to adequately measure and control for confounding variables such as oral hygiene practices, fluoride use and dietary factors (White, 2008). On the contrary to prolonged breastfeeding, some studies also show early weaning from breastfeeding, relating to earlier introduction to solid foods to be associated with earlier acquisition of mutans streptococci (Wan et al., 2001, Seow, 2012).

Frequent and/or prolonged exposure to sugars using a bottle in bed, or during the day is implicated as one of the major risk factors of caries among infants (Petersen, 2003). During sleep, there is a reduction in salivary flow, thus decreasing salivary neutralization capacity that in turn cause food stagnation and prolonged exposure to fermentable carbohydrates. Other notable risk factors associated with bottle use cited in the literature include the use of the bottle beyond 12 months and sweetened contents in bottles (Vadiakas, 2008).

2.2.8.3.5 Use of dental services

The use of dental services appears to be an important factor in predicting dental caries in young children (Harris et al., 2004). *Childsmile* aims to identify young children at highest risk of caries and encourage dental attendance from six months of age for primary dental care (Macpherson et al., 2010a). Additionally, it helps the child to get accustomed to visiting a dental practice, alleviating dental anxiety.

Dental registrations of three to five-year-olds in Scotland is now at 92%; nonetheless, registration of infants up to two-year-olds remains low at 47% (ISD Scotland, 2013b). This is of concern because evidence suggests younger age at first dental visit and regular attendance to be associated with lower levels of caries (Al Ghanim et al., 1998). Children who were reported to be treated for decay in the Sugar Survey were more likely to have been older when they first attended the dentist and attended the dentist for the first time because of trouble with their teeth (Sheehy et al., 2008). Children with higher rates of caries have also been reported to be more likely to have fewer routine annual dental check-ups and have parents who do not regularly visit a dentist (Hooley et al., 2012).

Families engaged in the early years of *Childsmile* Practice ***(See Section 2.3.9.1.2***) have been reported to have a failure to attend rate of 32%, with highest failure to attend rates concentrated in areas of deprivation (Deas et al., 2010). Dental attendance pattern has been found to be a factor contributing to the socioeconomic gradient in dental health among Singaporian young children, with SEP affecting dental attendance both directly and indirectly (Gao et al., 2010). The indirect effect of SEP on dental attendance is demonstrated to be through parental knowledge, attitudes including anxiety (Milsom et al., 2003) and beliefs regarding the importance of anticipatory dental attendance (Finlayson et al., 2007, Gao et al., 2010). Positive parental attitudes which are associated with better dental attendance (Tickle et al., 2000) and better dental health among children have been shown to be held by higher socio-economic groups (Skeie et al., 2010, Van den Branden et al., 2012).

2.2.8.3.6 Parental knowledge, attitudes and beliefs

Parents play a central role in the early years of children by the choices they make for their children and in shaping health promoting behaviours (Hooley et al., 2012). Parental knowledge, attitudes and beliefs about dental health have been shown to predict oral health related behaviours like diet, toothbrushing and routine dental attendance; and in turn dental caries in their children (Pine et al., 2004a). Parental attitudes and beliefs around oral health have been more closely associated with caries experience in young children than parental reports of toothbrushing practices or sugar-snacking habits (Pine et al., 2004a). Negative attitudes towards diet among parents were related to caries increment between ages three and five, with children whose parents had a negative attitude at both ages experiencing the highest risk of caries (OR = 6.0, 95% CI [2.7–13.4]) (Skeie et al., 2008). Parents of young children in the UK have been reported to have varying knowledge and attitudes towards dental health based on education, ethnicity and area of residence (Williams et al., 2002).

Parental self-efficacy

Parental self-efficacy is the parents' belief in their perceived ability to effectively implement actions required to prevent caries in their children. This concept is thought to determine behaviours (Pine et al., 2004b, Finlayson et al., 2007). It has been measured in relation to important behavioural risk factors like tooth-brushing and diet, encapsulating ideas around parental knowledge, logistical barriers and ability to control and discipline the child (Pine et al., 2004a, Finlayson et al., 2007).

Higher parental self-efficacy has been associated with more frequent toothbrushing (by parent and child), more frequent visits for a dental check-up (Finlayson et al., 2007) and lower sugar intake (Litt et al., 1995). The most significant variable predicting a child’s caries experience in a large international study involving 17 countries was parents’ perceived ability to perform toothbrushing for their child and not the child’s brushing behaviour itself. Furthermore, this effect persisted in children from deprived socioeconomic backgrounds (Pine et al., 2004a). Nonetheless, there is no clear evidence that traditional oral health promotion approaches of health education and development of parenting skills may reduce caries in children from deprived backgrounds (Watt, 2007).

Ownership

Ownership is a concept related to ‘parental self-efficacy’, with its routes based on the theory of ‘locus of control’ (LoC). It is the extent to which parents believe it is their responsibility to prevent caries in their children. A person is determined to have an external LoC, when one's health is believed to depend on luck, fate, chance or the influence of other persons. They have an internal LoC, when they believe that health is determined by one's own behaviour.

A clear linear association has been demonstrated between parental LoC and child's caries status, with increased internal parental LoC showing a higher probability for children to have no caries, independent of sociodemographic variables of parents (Lenčová et al., 2008). Among low income African-American families in Detroit, maternal levels of dental fatalism almost tripled their child’s risk of caries and showed a higher two-year caries increment among young children (Ismail et al., 2009). Fatalistic beliefs have also been reported in the UK, in Wales, with more than a quarter of the parents in a study believing ‘tooth decay runs in families’ and accepting ‘dental decay as bad luck’ (Karki et al., 2011).

2.2.8.3.7 Socioeconomic circumstances

There is overwhelming evidence of a strong association between socioeconomic circumstances and caries among young children, with disproportionate rates of caries found among children from deprived backgrounds (Reisine and Psoter, 2001, McMahon et al., 2010, NDIP, 2014, Campus et al., 2009, Oulis et al., 2012, Thomson et al., 2004). Studies that explore the relationship of caries prevalence/incidence with SEP were systematically reviewed (from 1990 up to 2000) by Reisine & Poster (2001). Studies in the review were ‘primarily’ cross-sectional surveys that used one or more indicators to measure SEP including- education completed, total family income in the past year, occupation, poverty status, Medicaid recipient, eligibility for Head Start, and/or eligibility for the special supplemental nutrition programme for Women, Infants and Children (WIC). They note a consistent and statistically significant inverse relationship between SEP and caries in children under six years. The relationship was weaker but still significant among older children.

A lifecourse study of a birth cohort analysing social mobility identified childhood SEP (parental occupation) at five years of age to predict adult caries risk (Thomson et al., 2004). Those with persisent high SEP and upward social mobility showed significantly reduced caries risk compared to those with low SEP in childhood through to adulthood. The dental health of five-year-olds have also been highly correlated with maternal levels of education (Finlayson et al., 2007, Wigen et al., 2011) and neighbourhood deprivation (Ismail et al., 2009), both with potential to explain oral health related attitudes and behaviours (Parkes and Wight, 2011).

Earlier acquisition of cariogenic bacteria, greater intake of NMES, inadequate oral hygiene practices, poor parental self-efficacy, and irregular dental attendance pattern have all been suggested as possible reasons for high caries rates among children from disadvantaged backgrounds (Arora et al., 2011). However, the underlying pathways that account for socioeconomic inequalities in caries remains largely unexplored.

The Role of stress

It has been suggested that the relationship between low SEP and caries in young children may be influenced by parental stress (Sisson, 2007). Evidence supports chronic stress to potentially influence health promoting behavioural decision making and additionally affect the immune response (Sisson, 2007, Boyce et al., 2010).

Salivary cortisol has been used as a surrogate measure of stress to explain SE inequalities among young children aged five to six years of age (Boyce et al. 2010). Low SEP (parental level of education), higher salivary cortisol levels in children and increased levels of cariogenic bacteria were each significantly and independently associated with caries. In addition, low SEP was associated with higher salivary cortisol levels, increased levels of cariogenic bacteria and a higher prevalence of caries (Boyce et al., 2010). The authors proposed two, social and psychobiological pathways through which lower SEP was associated with increased levels of salivary cortisol and cariogenic bacteria. However, findings from multilevel modelling analysis and structural equation modelling among low income families have reported parental stress along with parental self-belief in implementing regular twice daily toothrushing as protective against caries (Litt et al., 1995, Finlayson et al., 2007)

2.2.8.3.8 Immigrant background

Ethnic origin is not a variable routinely collected in the UK and Scottish oral health surveys. Watt and Sheiham (1999) argue that it may no longer be relevant in the UK and may divert attention from more relevant socioeconomic variables. However, Conway et al. (2007) reported five-year-old Pakistani children in Glasgow showing higher $d_3$mft levels than white Scottish children, with only 25% of the Pakistani children showing no obvious decay experience compared to 48% of white children. These differences persisted after controlling for socioeconomic deprivation. This echoes the findings from several other European countries (Grindefjord et al., 1996, Ferro et al., 2007, Wigen and Wang, 2010). Although a part of the difference in dental status among children of immigrant background is shown to reflect differences in SEP (Gao et al., 2010), results from some studies show that after controlling for parental SEP, oral health behaviours and attitudes, preschool children of non-western background have higher caries experience than their western peers (Conway et al., 2007, Wigen and Wang, 2010). Greater caries experience among immigrant children has been associated with 'cultural' differences in child rearing and immigrant parents' perceptions and beliefs towards dental health (Adair et al., 2004).

2.2.8.3.9 Age & Gender

Caries experience varies with time due to the additive effect of dental tissues being exposed to the oral environment. It is thus recommended that reporting of caries incidence/prevalence be age restricted and any analyses adjusted for the effect of age on caries experience (Levin, 2005).

Varying associations between gender and caries in young children have been reported, from no relationship in developing countries (Al Ghanim et al., 1998) to a significant relationship in developed countries- boys showing a higher caries experience (Dasanayake et al., 2002). Data on gender is routinely collected in UK caries surveys and only used to adjust for potential confounding effects. However, the relationship of gender to caries is complex due to interactions with other risk factors, particularly behavioural (Dasanayake et al., 2002). As previously reported in ***Section 2.2.8.3.2*** gender inequality in daily toothbrushing frequency have been reported among Scottish children (Levin and Currie, 2009, Masson et al., 2010) and various other European countries (Maes et al., 2006).

2.2.8.3.10 Family structure, marital and smoking status

Family structural characteristics have been associated with caries experience among young children, with negative risk indicators being mother's young age, household overcrowding (Rodrigues and Sheiham, 2000) and having had a change in marital status from pregnancy to five years of age (Mattila et al., 2000). Five-year-old children living in one-parent families were twice (OR 2.0, 95% CI 1.1 to 3.4) as likely to experience caries than children from two-parent families (Wigen et al., 2011).

Studies have also suggested maternal smoking as a significant risk indicator in caries risk among young children, with children's tobacco smoke exposure measurement ranging from parental self-reports of smoking status (Williams et al., 2000) to measuring serum cotinine levels (Aligne et al., 2003). Maternal smoking is reported to be significantly higher in single parent households (Leroy et al., 2008) and substantially attenuating the socioeconomic gradient in the UK (Williams et al., 2000).

While the above risk factors have been strongly associated with caries, only with additional data on SEP have they predicted the gradient in caries. Clearly there is difficulty in interpreting the interaction between the various risk factors above (younger maternal age, marital status and maternal smoking) which may stem from low SEP. While low SEP is a strong predictor of smoking; overcrowding and marital status have been used as a proxy measure of SEP (Galobardes et al. 2006b).

2.3 Socioeconomic position and Inequalities

2.3.1 Definition

A variety of terms have been used in the health literature to describe socioeconomic conditions, including ‘social class’, ‘social stratification’, ‘social’, ‘socioeconomic status’ and ‘socioeconomic position’ (Galobardes et al., 2007). To a large extent, the wide range reflect their roots in various disciplines and historical concepts (Lynch and Kaplan, 2000). However, these terms have often been used interchangeably in spite of their differing theoretical backgrounds and therefore, interpretations (Galobardes et al., 2007). Lynch and Kaplan (2000) suggested the use of the term, ‘socioeconomic position’ to mean ‘the social and economic factors that influence what position(s) individuals or groups hold within the structure of a society’. This thesis will follow the same consensus of using the term ‘socioeconomic position’ (SEP) rather than the commonly used phrase ‘socioeconomic status’ which has been argued to ‘blur distinctions between two different aspects of socioeconomic position: i) actual resources, and ii) status, meaning prestige’ (Krieger et al., 1997). In addition, the phrase ‘socioeconomic circumstances’ will be used when addressing both individual- and area-level indicators of socioeconomic position, which seems to more fully reflect the multidimensional complex construct of social and economic factors covering class, status, relative income etc.

2.3.2 Measures of socioeconomic position

Various measures of socioeconomic circumstances used in the health literature have been thoroughly reviewed by several authors (Krieger et al., 1997, Lynch and Kaplan, 2000, Galobardes et al., 2006b, Galobardes et al., 2006a, Galobardes et al., 2007). This section will discuss the major individual- and composite area-based measures of socioeconomic position, which are used in epidemiological research, and the measures of SEP that will be used in this body of work.

2.3.2.1 Individual-level measures of SEP

Individual-level measures of SEP used in health research measure some form of individual wealth or asset and to some extent are correlated because they all measure aspects of the underlying socioeconomic stratification, acting at various stages of the life course (Galobardes et al., 2007). However, comparing measures between countries and cultures is often difficult as levels may be country or culturally specific.

2.3.2.1.1 Education

Education is one of the most frequently used measure of SEP (Galobardes et al., 2006a). It attempts to capture the knowledge-associated assets of an individual and can in part measure early life SEP in a life course approach. It can be measured as a categorical variable based on definite educational levels achieved and/or as a continuous variable based on the number of years in formal education. The strengths of education as a measure of SEP include its relative simplicity of measurement in self-administered questionnaires, its relevance to people regardless of age, gender, ethnicity or working status, relatively less stigmatisation (compared to income) and that it remains broadly stable over a life course (Krieger et al., 1997, Galobardes et al., 2006a). However, limitations involve the potential of obscuring social mobility as more individuals in recent years have higher years of education compared to the older cohorts (Galobardes et al., 2006a). When considering the span of educational levels, the range for income and/or wealth is far-reaching, thus making it a relatively less sensitive measure in assessing the magnitude of socioeconomic inequalities in health (Krieger and Fee, 1994). This measure of SEP may pose some challenges in international studies due to differences between educational systems in various countries.

The importance of measuring parental educational level as a measure of childhood SEP affecting health has been emphasised (Krieger et al., 1997). More specifically, educational attainment has been attributed to the acquisition of health related knowledge, attitudes and beliefs, lifestyle behaviours and optimal use of health services (Galobardes et al., 2006a), parental characteristics highly associated with health (Finlayson et al., 2007).

2.3.2.1.2 Occupational- based measures

Occupation-based measures are traditionally used for measuring SEP in the UK, where social stratification has historically been conceptualized in terms of occupation (Galobardes et al., 2007).

There are various indicators based on occupation, with most studies using the current or the longest held occupation as a measure of adult SEP. The oldest and the best known official measure of social class in the UK until 2001 was the British Registrar General's social class (RGSC) which summarized occupations to represent ''social grades'' (1990).

Galobardes et al. (2006b) outlines the limitations of the RGSC for its weak theoretical basis, considering the subjectivity of assigning individuals into categories based on prestige. In addition, it does not take into account continuous changes in the occupational structure, such as the reduced number of people working in unskilled and semi-skilled manual occupations, or the increased number of working women. It also struggles with categorizing groups outside the active labour force (non-retired unemployed, homemakers, retired adults etc.) and its limited recognition of differences between individuals in the same occupation group in terms of both education and income (Krieger et al., 1997). Nevertheless, the RGSC has widely been used to describe socioeconomic inequalities in health mainly due to its past widespread use in the UK in many censuses and surveys over a long period and its adaptations used in many European countries, making comparability between studies easier (Galobardes et al., 2006b).

Due to the limitations discussed above, in 2001 the Office for National Statistics in the UK adopted the UK National Statistics socioeconomic classification (NS SEC) as its official occupation classification (Table 2-1) (2005). The NS-SEC is based on the Goldthorpe Schema (Erikson and Goldthorpe, 2002) and measures employment relations and conditions of occupations as opposed to skill and social standing. It can be derived based on the level of detail of the employment status available (full, reduced and simplified) as a categorical measure. Additionally, there are procedures for classifying the unemployed (2005). While there is an order to the occupational groups in the NS-SEC classification, it is not in a strict hierarchical order.

One of the strengths to using NS-SEC is its theoretical base, which may help in the development of causal narratives to explain a part of the frequently observed socioeconomic gradient in health.

Table 2-1: The National Statistics Socio-economic Classification Analytic Classes

1	Higher managerial and professional occupations
1.1	Large employers and higher managerial occupations
1.2	Higher professional occupations
2	Lower managerial and professional occupations
3	Intermediate occupations
4	Unskilled
5	Small employers and own account workers
6	Lower supervisory and technical occupations
7	Semi-routine occupations
8	Routine occupations
9	Never worked and long-term unemployed

There are various other occupational social classification schemes detailed by Lynch and Kaplan (Lynch and Kaplan, 2000) and Galaborades et al. (2006b). However, most have not been updated regularly.

2.3.2.1.3 Income

Galobardes et al. (2006a) considers income as the most direct measure of material circumstances with a cumulative effect over the life course, measured through direct reporting of monthly or yearly income. However, it is considered to be a ‘sensitive’ indicator as individuals may be reluctant to provide such information. This measure also has limitations of being most likely to change over a time period and might only partly capture SEP, as it does not include assets, inherited wealth etc. It is more difficult than education to be compared across countries. Furthermore, income for young and older adults may be a less reliable indicator of their true SEP as it varies with age. To be comparable across households, it is recommended that household rather than individual income is collected along with family size and number of dependents under the reported income.

2.3.2.1.4 Benefits claimant

Individuals claiming/entitled to certain benefits or whose income is fully derived from benefits may be used as an indicator of low income and therefore low SEP. The main strength of this measure is the availability of robust sources for this data that are regularly updated; for e.g. The Department for Work and Pensions. However, this measure only gives a picture about those at one end of the socioeconomic spectrum. In addition, only those who claim benefits may be identified and not those who are eligible for benefits. They are also arbitrary indicators of low income that do not have a scientific definition, but projecting rules defined by the Government (Shaw et al., 2007).

2.3.2.1.5 Housing tenure

Housing tenure is a marker of material circumstances (Galobardes et al. 2006a) and differences in housing tenure have shown patterns of inequalities in health in Scotland previously (Macintyre et al., 1998). It is measured by checking if the housing of the individual is owned (owned/being bought with a mortgage), or rented from a private or social landlord (Galobardes et al. 2006a). Although it has the advantage of being relatively easy to collect, it has limitations of area specificity, consequently proving difficult to compare across studies (Shaw et al., 2007).

2.3.2.1.6 Other measures

A vast number of other measures of individual SEP exists; they include: a range of country-specific occupational indices, housing conditions, household amenities (Galobardes et al. 2006a), indicators of wealth (Lynch and Kaplan, 2000) etc. Other ‘proxy’ measures of individual SEP described by Galobardes et al. (2006a) include: number of siblings, marital status and some health measures (e.g. infant or maternal mortality)

2.3.2.2 Area-based measures (Indices of deprivation)

Area-based measures utilise data from census or other administrative databases to aggregate the data at a small area level (Galobardes et al., 2006b) and classify individuals by the socioeconomic properties of their area of residence (usually based on postcode). However, there is a potential for misclassifying individuals, as all individuals in an area may not necessarily be of the same SEP.

2.3.2.2.1 Carstairs index

The Carstairs score and its deprivation categories (DEPCAT) has been a commonly used indicator of deprivation in Scotland which is calculated from various census variables- Overcrowding, male unemployment, car ownership and low social class (IV & V). Geographical areas are based on postcode sectors with an average population of 5,000. A DEPCAT score is calculated for each postcode sector, classified from DEPCAT 1 (most affluent) to DEPCAT 7 (most deprived) (Carstairs and Morris, 1990).

2.3.2.2.2 Scottish Index of Multiple Deprivation

The Scottish Executive, in response to the August 2003 report 'Measuring Deprivation in Scotland: Developing a Long-Term Strategy', developed the Scottish Index of Multiple Deprivation (SIMD) which identifies small area concentrations of multiple deprivation across Scotland. The SIMD 2009 used in this body of work combines 38 indicators across seven domains, namely: income, employment, health, education, skills and training, housing, geographic access and crime at the level of 'data zones'. Data zones are groups of 2001 census output areas and have populations of between 500 and 1,000 residents with arguably similar social characteristics. SIMD ranks are divided into quintiles, with quintile 1 covering the most deprived 20% of Scottish postcode areas (The Scottish Government, 2009b). Criticisms against SIMD arise from the use of an area-based measure to identify individuals as some who are materially disadvantaged live in areas that are not particularly deprived in terms of SIMD and vice-versa (Shaw et al., 2009). SIMD 2009 has now been superseded by SIMD 2012. However, it is considered appropriate to use the SIMD most close to data collection.

2.3.3 Choice of SEP indicator

SEP is a multidimensional construct that has been measured in numerous ways across studies due to strengths and limitations of each measure. Thus, the inclusion of multiple measures of SEP is recommended and those chosen should be based upon their conceptual links with the health outcome (Shavers, 2007). Additionally, it helps with validation and avoids residual confounding. The correlation between the measures, education, occupation and income in developed countries was found to be relatively weak (0.3-0.6) suggesting that each measure explains a different aspect of SEP which contributes differently to health inequalities (Braveman et al., 2005). It is recommended that, where possible, both individual- and area-based measures are used as each appears to have a role in explaining the degree and causal mechanism of dental health inequalities (Thomson and Mackay, 2004).

2.3.4 Socioeconomic inequalities in health

Health inequality is defined as variations in health status across individuals in a population (Gakidou et al., 2000). Social and economic differences within a population is known to cause a gradient in health status worldwide, with those from lower socioeconomic position experiencing poorer health (Marmot and Bell, 2011). These inequalities influenced by SEP are shaped by education, occupation, income, gender and ethnicity (Marmot and Bell, 2011).

In Scotland, inequalities have long been credited to relative deprivation in the city of Glasgow (McLoone and Boddy, 1994). The socio-economic composition of Glasgow differs from Scotland, as a whole. Glasgow is the most economically disadvantaged area, with almost half (45.3%) of the 15% most deprived datazones in Scotland (The Scottish Government, 2009b). Marmot and Bell (2011) took Glasgow as an example of stark health inequities, describing men living in the most disadvantaged part of Glasgow to have ‘dramatically worse health than the Indian average’. The unexplained patterns of poor health and low life expectancy compared to the rest of equally deprived former industrial cities such as Birmingham, Liverpool and Manchester has been conceptualized as due to some unknown 'Glasgow effect' (Walsh et al., 2010a). Gray and Leyland (2009) showed that part of the ‘Glasgow effect’ was attributable to socioeconomic factors.

2.3.5 Socioeconomic inequalities in dental caries among Scottish young children

Although Schou and Uitenbroek (1995) demonstrated the stronger association between caries and SEP than toothbrushing behaviour and sugar consumption, Sweeney et al. (1999) using the Carstairs 1991 score were the first to show the full extent of the socioeconomic inequality among five-year-olds in Scotland. Children from the most disadvantaged DEPCAT category showed mean $d_3$mft scores of 4.87 compared to 1.48 in the most advantaged DEPCAT category and the percentages with obvious caries experience were 80.2% and 37.6% respectively (Sweeney et al. 1999). The linear gradients in the distribution of the disease across the seven DEPCAT categories emphasised the need for 'multi-agency working, precise and intensive focusing on the target group, and consumer involvement' for improvements (Sweeney et al. 1999). Successive SHBDEP and NDIP reports are in line with these findings. Socio-economic inequality in the prevalence and mean $d_3$mft reduced over time from 1993 to 2003 (Levin et al., 2009). However, inequality in the amount of disease experience for those affected increased. In 2003, the odds of a child in DEPCAT 7 showing caries were 4.60 (95%CI, 3.47 to 6.14) compared to peers in DEPCAT 1 (Levin et al., 2009).

Marked improvements have been evident across the socioeconomic spectrum since 2003, with the extent of disease continuing to fall in those most affected by caries (NDIP, 2014). The SiC^{10}[1] decreasing from 11.6 to 9.0 through 1993-94 to 2007-08 and from 13.0 to 9.8 for Glasgow (Blair et al., 2013). However, large inequalities persist with only 53% of the five-year-olds in the most deprived areas showing no obvious decay experience compared to 83% in the least deprived (NDIP, 2014). Children from the most deprived quintile (SIMD 1) are yet to meet the 2010 National Target of 60% with no obvious decay experience.

2.3.6 Explanations of socioeconomic inequalities

Sisson (2007) reviewed contemporary explanations for inequalities in oral health; namely materialist, behavioural, psychosocial and the life course perspective.

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[1] The SiC is calculated as the mean dmft in the third (33%) of children with the highest caries experience (dmft). The SiC index has been modified to use alternative cut offs of 25%, 20% and 10% (SiC^{25}, SiC^{20} and SiC^{10}).

The materialist explanation considers how materialist aspects like cost of treatment, cost of accessing treatment and purchasing health promoting foods and products may be limited to individuals from lower socioeconomic backgrounds. An alternative behavioural and psychosocial theory proposes the likelihood of people from lower socioeconomic circumstances suffering from poorer health due to engaging in ‘health damaging behaviours’ and higher levels of psychological stress respectively (Sisson, 2007). The life course perspective is the most recent theory and is thought to provide the best explanation for socioeconomic inequalities in oral health as it combines materialist, behavioural and psychosocial factors over time in understanding causation. Socioeconomic and biological factors during childhood have been demonstrated to be significantly related to caries experience in adolescence (Nicolau et al., 2003) and adulthood (Thomson et al., 2004). This suggests a need for longitudinal, multifactorial designed studies to investigate risk factors in the development of caries in Scottish children in the current age of improving oral health, to inform interventions that tackle the determinants of inequalities from infancy (or earlier).

2.3.6.1 Socioeconomic explanations of dental caries risk- Interaction between the materialist and behavioural theories

Whilst a number of studies indicate that oral health-related behaviours explain part of the SE disparities in oral health outcomes in adolescents (Mashoto et al., 2010, Jung et al., 2011) and adults (Sanders et al., 2006b, Donaldson et al., 2008) worldwide, only one study has been conducted among young children (Slade et al., 2006). Although the results showed strong associations between oral health outcome and behaviours, behaviours did not fully account for the socio-economic gradient in oral health. None of the studies have attempted to fully explain the socioeconomic gradient by including explicit variables on all important oral health-related behaviours while using validated measures. It is suggested that SEP has a direct effect on oral-health outcomes, independent of the proximal determinants of oral-health-related behaviours. An analysis of young children’s (one and a half to four and a half years of age) data from the National Diet and Nutrition Survey indicated the strength of association between SEP and caries to exceed that of sugar consumption and toothbrushing (Gibson and Williams, 1999). It is unknown if bringing material, behavioural and psychosocial factors together in a longitudinal study will fully explain the SE inequalities in oral health (Sisson, 2007).

2.3.7 Measurement of socioeconomic health inequalities

A wide variety of measures have been used to quantitatively measure socioeconomic inequalities in health and the choice of measure primarily depends on the research question (absolute or relative inequalities) and the available data (individual- or area-based measures and the level of measurement of variables) (Mackenbach and Kunst, 1997). This section will review common summary measures of health inequality under the headings ‘simple measures’ and ‘sophisticated measures’ (Mackenbach and Kunst, 1997).

2.3.7.1 Simple Measures

Range

Range is the most commonly used measure of health inequality and involves comparing the experience between the lowest and highest socioeconomic groups. This may be presented in the form of the range (Absolute Range), taking the difference in outcome between the lowest and highest socioeconomic (reference) groups, but more often presented as the relative difference or ratio between the reference group and the other (Relative Range). However, the range ignores the experience of the in-between socioeconomic groups and does not represent inequalities well when there is no clear linear gradient between the lowest and highest socioeconomic groups.

Population attributable risk (PAR)

PAR is a measure of reduction in disease if the study population were to have the rate of those in the reference group. It is expressed as a fraction or proportion between the overall disease rate and the rate in the reference group. The main limitation of PAR is its inapplicability in comparative studies where the reference socioeconomic group may not represent the same proportion of individuals to the comparative populations. PAR applied to Scottish epidemiological data of five-year-olds suggested that overcoming relative deprivation would have reduced 22.8% of the caries experience in the population in 2007/08 (Blair et al., 2013)

2.3.7.2 Sophisticated (complex) measures

These include numerous tests from statistics and income economics. The Odd's ratio (OR) is the ratio of the odds of a health outcome in the reference group to the odds of the health outcome in the other group. However, it has been cautioned against use when the frequency of the health outcome is higher than 0.20 due to the OR overestimating the size of the relation (Regidor, 2004)

Receiver Operating Characteristic (ROC) curve plots the sensitivity to '1-specificity' for an exposure to predict presence or absence of disease. The c-index is the area under ROC curve (Altman and Bland, 1994). A c value of 0.50 indicates no predictive ability where as a variable with perfect predictive power has a c value of 1.0. ROC plots have been used to examine microbial carriage (Radford et al., 2001), prior caries experience and to determine the extent of varying oral health due to deprivation (McMahon et al., 2010).

Slope index of inequality (SII) and Relative index of inequality (RII) are regression based methods that involve calculating the mean health status of each socioeconomic group and then ranking groups by their SEP. SII is interpreted as the absolute difference in health by moving from the lowest socioeconomic group to the highest. The SII can be divided by the mean level of health to estimate relative differences- called the RII.

SII and RII are appropriate to measure how health varies with increasing SEP as opposed to comparisons between SEP groups. These indices take into account the differences in the proportions of the population in the different categories of a SEP variable, thus enabling direct comparison between SEP variables (Mackenbach and Kunst, 1997). RII is considered useful for making comparisons between geographic regions or cohorts.

Recently SII was applied to Scottish epidemiological data of five-year-olds from 1993/94 to 2007/08. A downward trend in SII demonstrated improvements in absolute inequality (Blair et al., 2013). These findings echoed with deciduous caries inequality trends observed in Australia (Do et al., 2010).

The Concentration index is calculated as twice the area between the concentration curve and the line of equality. The concentration curve models the cumulative distribution of a health outcome against the cumulative proportion of individuals by SEP, ranked from lowest to highest. This curve is then compared to the line of equality. If the concentration curve coincides with the line of equality, all individuals have the same level of health. If the curve is under the diagonal, health is concentrated in persons of higher SEP, and vice-versa (Regidor, 2004).

2.3.8 Approaches to address inequality

Universal population approaches or targeted individual approaches on their own are considered inadequate for improving dental health and reducing inequalities (Shaw et al., 2009). Instead a combination of the two approaches- 'directed population approach' has been suggested. This follows the 'proportionate universalism' strategy proposed for tackling health inequalities by which actions are applied universally, but with the scale and intensity matched to the level of disadvantage (Marmot et al., 2010). This essentially acknowledges health inequalities as a gradient over extreme differences between the well off and worst off. Another key recommendation has been prioritising investment in prevention and health promotion from early years (Marmot et al., 2010).

There is some evidence that interventions focusing on individual behaviour (individualistic approach) alone may have limited effect in reducing inequalities or even increase them, due to greater uptake among more affluent groups (Schou and Wight, 1994). These narrowly focused preventive and educational 'downstream' interventions have been criticized for 'victim blaming' and over-simplifying the problem, by neglecting the wider circumstances in which people live and behaviours are learnt (Watt, 2007). Consequently, Watt (2007), following on from the WHO (1986) Ottawa Charter and the more recent WHO Commission on Social Determinants of Health (2008), advocates a more 'upstream action'- to develop public health strategies and policies which address the underlying social determinants of oral diseases (Watt, 2007). A combination of both upstream and downstream interventions has been suggested to best address inequalities (Scottish Governement, 2008). The combination ensures that the root causes of inequalities are addressed while those who are victims to health-compromising behaviours are supported. The common risk factor (CRF) approach has also been recommended to address the shared risk factors (behavioural-shared risk and common social determinants) associated with various chronic health conditions (like cardiovascular diseases, obesity and oral diseases) (Watt and Sheiham, 2012). The prevalence of most chronic health conditions is highest among the deprived and adopting the CRF approach means reducing inequalities, and creating supportive environments for promoting health in the whole population. It also avoids duplication, increasing effectiveness and efficiency of health promotion.

2.3.8.1 Other chronic health condition with shared risk factors

Some risk factors associated with dental caries; namely diet and socioeconomic position overlap with that of the obesity epidemic (Spiegel and Palmer, 2012). Childhood obesity is an important predictor of adult obesity and other chronic diseases including heart disease, certain cancers, type-2 diabetes and stroke. Additionally, it has important social and psychological consequences. The prevalence of obesity and overweight in children in the UK remains one of the highest amongst industrialised countries, and the most recent figures for Scotland report 14.9% of P1 children overweight, obese and severely obese (ISD Scotland, 2013a). Additionally, data from the Child Health Surveillance Programme (CHSP) (ISD Scotland, 2013a) and the 2011 Scottish Health Survey indicate obesity to be strongly associated with deprivation (SIMD), although it did not follow any particular pattern (The Scottish Government, 2012). Results from recent systematic reviews are equivocal of a relationship between the two conditions for the primary dentition (Hayden et al., 2013). There is a need to understand the relationship between dental caries and obesity and any potential pathways of the association if one exists, so that common approaches to prevention can be identified (Sheiham and Watt, 2000).

2.3.9 Policy context- Scotland

The Scottish Office Home and Health Department published in its *Health Education in Scotland- A national Policy Statement (1991)* the first dental health target for Scottish children of 60% of children to have no obvious decay experience by 1995. Later on, publications of White Papers on Health (The

Scottish Office, 1999, Scottish Executive, 2003) emphasised new approaches towards child health improvement with an emphasis on reduction of health inequalities by focusing on early years interventions. The key policy document that has influenced oral health in children within Scotland in recent years is the *Action Plan for Improving Oral Health and Modernising Dental Services in Scotland* (also called the *'Scottish Dental Action Plan')* (Scottish Executive, 2005). This action plan outlined a range of oral health improvement measures in children and a number of ambitious milestones to achieve, including the three times unachieved target of 60% of children to have no obvious decay experience, this time by 2010. This target was met for five-year-olds for Scotland as a whole in 2010 (Macpherson et al., 2010b), with all NHS boards meeting the target in 2012 (Macpherson et al., 2012).

2.3.9.1 Childsmile

Childsmile is the Scottish national oral health improvement programme for children, initiated in 2006 in response to the *Scottish Dental Action Plan* (Scottish Executive, 2005). It is based around the principles of the WHO Ottawa Charter for health promotion (World Health Organization, 1986) ('building healthy public policy', 'creating supportive environments', 'strengthening community action', 'developing personal skills', and 're-orientating health services'); and informed by published clinical guidelines (SIGN, 2000, SIGN, 2005) and previous pilot oral health improvement programmes in Scotland (Blair et al., 2006).

Childsmile aims to improve the oral health of Scottish children and reduce inequalities both in dental health and in access to dental services from infancy through a universal and targeted approach (Macpherson et al., 2010a, Marmot et al., 2010). *Childsmile* consists of four components- *Childsmile* Core, *Childsmile* Practice, *Childsmile* Nursery and *Childsmile* School (Macpherson et al., 2010a).

2.3.9.1.1 Childsmile Core

The Core component is universal: every child in Scotland is provided with fluoride toothpaste and toothbrushes on at least six occasions until the age of 5 years. The Core programme also includes provision for the daily-supervised toothbrushing of every three and four-year-old in all nurseries and additionally to targeted Primary-1 and Primary-2 children attending schools in the 20% most deprived areas in each Health Board in Scotland (Macpherson et al., 2010a).

2.3.9.1.2 Childsmile Practice

The Practice component focuses on children's oral health from birth and helps to link families to Primary Care Dental Services by six months of age. All children are invited to take part in the Practice Programme. Families are risk assessed by health visitors around eight weeks old to determine whether they need additional support from a Dental Health Support Worker (DHSW) (Macpherson et al., 2010a). DHSWs visit these families in their homes to facilitate attendance at *Childsmile* dental practices, reinforce oral health messages and link families to other community activities that support oral health. Parents are encouraged to take their child to a dental practice at least every six months, where toothbrushing demonstrations and dietary advice are given. Children over two years are able to receive fluoride varnish applications and fissure sealants at the dental practice. Fissure sealants seal off the pit and fissure surfaces of the teeth that are particularly susceptible to caries. Dental practices are paid by the NHS to provide these preventive interventions (Scottish Government, 2011).

2.3.9.1.3 Childsmile Nursery and Childsmile School

This component of *Childsmile* provides fluoride varnish applications, delivered by Extended Duty Dental Nurses (EDDNs) trained through *Childsmile* in targeted nurseries and schools (located in the 20% most deprived areas in each Health Board) and identify children with dental care needs to facilitate dental attendance.

2.3.9.2 Evaluation of complex health interventions

Childsmile is a complex intervention involving multiple aims, interacting components, targets, processes and outcomes and stakeholders; informed by evidence, theories of change and relying on multiple behaviours from those delivering or receiving the intervention. Additionally, there is local tailoring within NHS boards influencing its delivery and possible effectiveness (Craig et al., 2008, Macpherson et al., 2010a, Turner et al., 2010). Addressing its complexity, *Childsmile* has been subject to a comprehensive evaluation (*Childsmile* Research and Evaluation Team (CERT), 2009) following the Medical Research Council (MRC) guidance (Craig et al., 2008). The MRC guidance suggests developing interventions systematically using the best available evidence and theory, then to test them using a phased approach starting with a series of pilot studies targeted at each of the uncertainties in the design, and moving on to an exploratory and then a definitive evaluation (Figure 2-2). The results should be disseminated widely, with further research to facilitate and monitor the process of implementation (Craig et al., 2008).

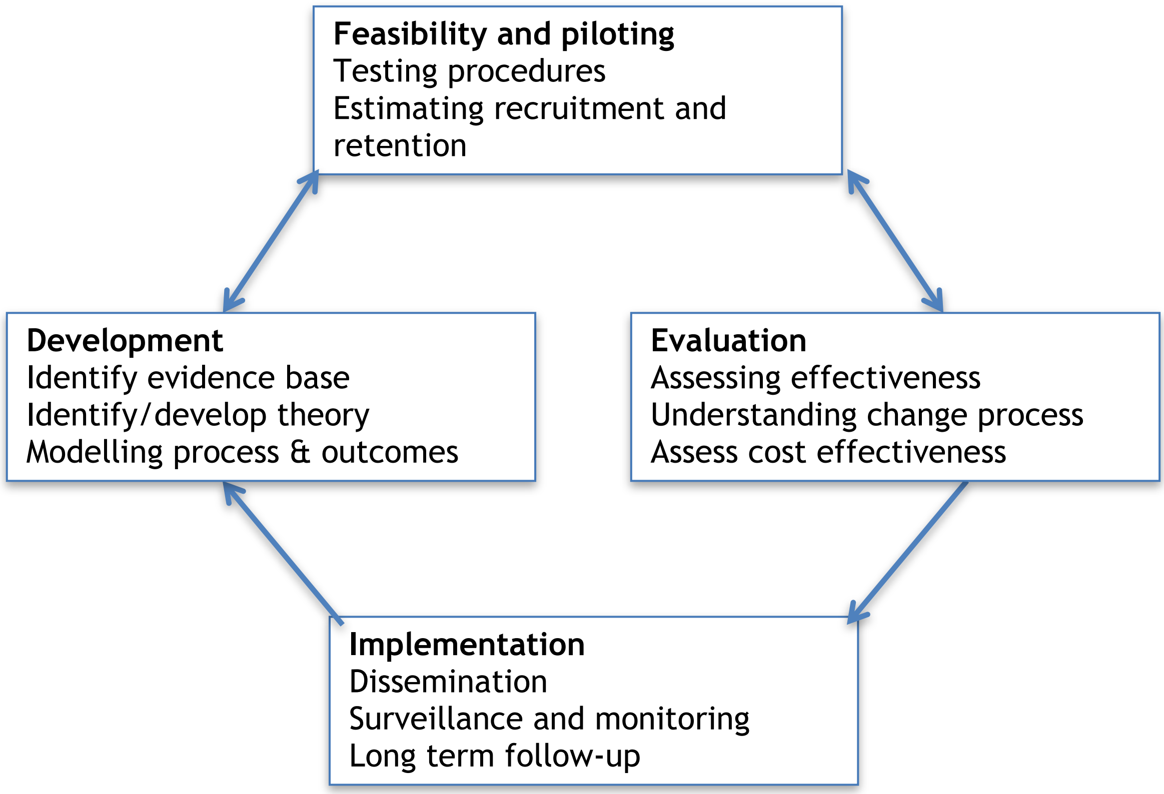


Figure 2-2: Elements of development and evaluation process of a complex intervention (Adapted from Craig et al., 2008)

2.4 Cohort studies

2.4.1 Introduction

A well-designed cohort study is a vital method for evaluating causality in epidemiology as it can indicate the temporal framework of exposure and outcome, providing strong scientific evidence (Level II evidence). Although the need for a large sample size and potentially long follow-up duration might have cost and time implications, cohort studies are particularly advantageous in establishing risk factors as exposure is always assessed prior to disease development (Song and Chung, 2010). In addition, incidence and relative risk can be assessed in the exposed and the unexposed. They also have the power to examine rare exposures and examine multiple disease outcomes simultaneously. A prospective cohort study is the ideal design to test causal life-course hypotheses (Nicolau et al., 2007). Although biases from loss to follow-up; and those from outcome information being influenced by knowledge of exposures are inevitable limitations of cohort studies, baseline data on those lost to follow-up can be compared with those remaining in the cohort for evaluating effect of attrition on estimates (Song and Chung, 2010).

2.4.2 Cohort studies exploring risk factors for dental caries in young children

A thorough consideration of the literature on all cohort studies exploring risk factors for dental caries in young children is beyond the scope of this section. Numerous factors have been associated with caries development in young children, but there are few quality longitudinal studies confirming ‘risk factors’ (Harris et al., 2004). This section will briefly review and systhesize results from major cohort studies that were thought to be generaliziable and/or relevant to the population under study, assessed multiple risk factors, used multivariable statistical analysis and had an acceptable attitrition rate.

Grindefjord et al. (1995) assessed risk factors for dental caries development among 692 Swedish two and half-year-olds. They found 92% of those with caries at baseline developed new lesions during the one-year period and high sugar consumption, mother’s educational attainment, immigrant background and mutans streptococci colonization at baseline were associated with caries development and progression at three and a half years (Grindefjord et al., 1996). The four risk factors were found to be cumulative; with the probability of caries developing by three and a half years estimated to be 80%. Another study of 289 Swedish children examined at one, two and three years of age, and their parents interviewed about the children's oral hygiene and dietary habits at one and two years of age highlighted the balance between sugar consumption and oral hygiene practices in caries development (Wendt et al., 1996). Children engaged in caries-risk dietary behaviour like frequent consumption of a sugary drink at one year of age showed an increased likelihood of no caries experience at three years when strict oral hygiene practices (including the use of fluoride toothpaste) were observed at two years of age. Similar findings were reported among 135 Finnish children followed up prospectively from three to six years of age (Karjalainen et al., 2001). The combination of sweet intake more than once a week and the presence of visible plaque at three years gave 1.7 fold increase in caries risk at six years as compared to children with neither habit (Karjalainen et al., 2001). These two risk factors have shown to have a significant positive correlation with the mutans streptococci levels in plaque (Habibian et al., 2002). In a cohort study of 163 English infants, children who did not brush regularly by 12 months of age and those who ate/drank more frequently during the day were more likely to show mutans streptococci in their plaque at 12 months of age (Habibian et al., 2002).

Litt et al. (1995) employing a multidimensional model followed 184 low income Connecticut three to four-year-olds for a year to evaluate caries development. The study collected interview data from parents regarding sociodemographics, dental knowledge, child's sugar intake, oral hygiene practices and some infant feeding practices in addition to collecting saliva samples from children to assess *S. mutans* levels. The study found baseline *S. mutans* levels strongly predictive of caries at five years of age, and baseline *S. mutans* levels strongly predicted by sugar intake levels, which in turn was predicted by ethnicity and parental self-efficacy. Negative parental attitudes towards diet have been related to caries increment between ages three and five, with children of parents with negative attitude at both ages experiencing the highest risk of caries (OR = 6.0, 95% CI [2.7 to 13.4]) (Skeie et al., 2008). While dental attitudes improved in parents over a two years' period, children of mothers with low educational attaintment and immigrant status maintained negative dental attitudes (Skeie et al., 2010).

A longitudinal study of 642 Iowa children collected caries risk factor data from one to five-year-olds and examined children for caries from four to seven years of age (Marshall et al., 2003). The study found that older age at dental examination, lower family income, less frequent toothbrushing and higher non diet soft-drink consumption were factors associated with caries. Identical findings were found when 128 six months olds from low SEP in Iowa were followed up after a year (Warren et al., 2009). However, socioeconomic position was not found to be associated with caries in this study, possibly due to reduced variation in SEP in the sample.

Whilst there are no longitudinal data on the dental health of young children in the UK, a number of birth cohort studies are ongoing starting from the National Survey of Health & Development (NSHD) in 1946. Noteworthy are the more recent Avon Longitudinal study of Parents and Children (ALSPAC) (Golding, 1990) and the Millenium Cohort Study (Plewis and Ketende, 2006). The ALSPAC has followed more than 14,000 children of Avon (England) since 1991 from their mothers' pregnancies onwards. Duncan (2000), using the data from ALSPAC, examined 10% of the sample for caries status at two and a half, three and a half and five years of age. Children who lived in council or rented property and whose mothers were educated only up to O-level standard were more likely to show caries. A strong association between the frequency of consumption of chocolate at six months and 15 months and caries experience at five years was also shown. Although multivariable analyes was not pursued in this study (Duncan, 2000), the ALSPAC have shown poor dietary habits (diet based on high-fat and sugar content, processed and convenience foods) at four and seven years of age significantly more likely among children with mothers of low educational attaintment and where the child had more siblings (Northstone and Emmett, 2005). These findings are in line with the GUS study, a large-scale longitudinal survey tracking Scotland's children and their families from infancy through to their teens since 2005. The GUS study of four to five-year-olds found indicators of socioeconomic circumstances associated with eating patterns and toothbrushing frequency (Bradshaw, 2008). Children from the lowest income households were twice as likely to report lower frequency of brushing compared to those in the highest income households (Bradshaw, 2008). In a large cohort (1,500) study of infants examined and sampled annually from one to four years of age in Dundee, caries prevalence has been shown to increase with age and social deprivation. Social deprivation (measured as DEPCAT) and lactobacilli were correlated, independently of caries status, among three and four-year-olds. Correlations between mutans streptococci and deprivation were found among two-year-olds with enamel lesions and in three-year-olds with dentine lesions (Radford et al., 2000, Radford et al., 2001). Consequently, a risk assessment model was developed, which showed that the two significant risk factors at age one year for the child having at least three carious teeth at age four years was living in council housing and the health visitor's opinion of the child's risk of caries (sensitivity=65%, specificity=69%) (MacRitchie et al., 2012). However, there is limited evidence that the use of a caries risk assessment tool aids caries prevention.

The most recent UK-wide Birth Cohort, Life Study aims to recruite over 80,000 babies and their families from a wide range of backgrounds and follow them from their pregnancies to adulthood (Dezateux et al., 2013). The study will collect a large amount of data- social, environmental, behavioural and biological samples. The study is promising in that it can explore the interplay between various factors and the mechanisms operating through the lifecourse. In addition, the study has implications for genetic studies which may partly explain the increased predisposition of individuals from deprived backgrounds to develop chronic diseases, through as yet unknown mechanisms.

2.4.3 Summary

In summary, a combination of socioeconomic factors [family income (Fontana et al., 2011), maternal education (Grindefjord et al., 1995, Grindefjord et al., 1996, Wigen et al., 2011), neighbourhood disadvantage (Ismail et al., 2009)], immigrant status (Grindefjord et al., 1996, Wigen et al. 2011), dental health beliefs and fatalism (Ismail et al., 2009), sugar consumption (Karjalainen et al., 2001), oral hygiene factors (Grindefjord et al., 1995, Wendt et al. 1996, Rodrigues and Sheiham, 2000) and mutans streptococci levels (Warren et al., 2009) feature as significant risk factors for childhood caries.

Comprehensive cohort studies are yet to be undertaken to reveal the relative importance of each risk factor and the extent of the relationship between each other. The above is not a comprehensive review of all cohort studies, but all of the studies reviewed above except one (Litt et al., 1995) have carried out analyses at one level, thus neglecting the sequence of direct and indirect influences and interaction patterns. This also means that all risk factors are treated equally, often underestimating the effect of the wider determinants of caries. There is clearly a lack of focus on the causal pathways between the various risk factors for caries in young children, which may suggest new explanations for the gradient in caries. Furthermore, relatively fewer studies have been carried out on young children under three years of age on a large birth cohort. Complex concepts like SEP have not been measured multidimensionally, which means the wider social determinants which impact on dental health remain under-studied. Consequently, the development of caries in very young children and the mechanisms underlying the socioeconomic inequalities in the dental health of young children remains less understood.

There is a scarcity of well designed cohort studies, conducted in very young children initiated as early in life as possible. In order to understand the natural history of caries development and to identify which biological, behavioural, psychosocial and socioeconomic factors in early life may increase risk or protect against caries, it is crucial that a longitudinal study on a birth cohort is designed. Such a study will need to be large by design to test multiple associations, interactions and directions of effect, whilst adjusting for putative confounding factors.

2.4.4 Importance of pilot studies

The MRC explicitly recommends the use of feasibility and pilot studies for avoidance of problems associated with sample size calculation, recruitment and retention, acceptability and compliance that impede evaluations (Craig et al., 2008). Additionally, they can test and validate measures, assess time and budget problems and potential personnel and data management issues (van Teijlingen and Hundley, 2001). However, caution must be exercised when interpreting pilot studies results when making assumptions about the numbers required when the evaluation is scaled up as effects may be more variable and response rates lower when the intervention is rolled out across wider settings (Craig et al., 2008).

2.5 Summary of Literature review

Dental caries is the dissolution of tooth surface caused due to acids produced by bacteria after fermenting sugar (Hicks et al., 2004). Caries in young children is a major public health problem worldwide, with recent studies reporting an increase in caries prevalence and a skewed distribution in many developed countries (Bagramian et al., 2009). It stands as the most common chronic disease of childhood, worldwide and in Scotland (Centers for Disease Control and Prevention, 2012, ISD Scotland, 2013a). It has a significant impact on children's general health, future oral health and quality of life, and results in a substantial economic burden for the health service (Sheiham, 2006).

Data from oral health inspections of young children in Scotland show a continuous steady improvement in dental health since 2003 (NDIP, 2014). Despite these improvements across Scotland, there is a significant number of children experiencing caries, with those at the lower end of the socioeconomic spectrum suffering the greatest burden of disease (NDIP 2014). The relative improvements in the caries rates across all social groups have largely been attributed to the national child oral health improvement programme, *Childsmile* (Macpherson et al., 2013). *Childsmile* adopts a life-course approach commencing in infancy offering a range of interventions- some applied universally, others in proportion to the level of disadvantage (Macpherson et al., 2010a, Marmot et al., 2010).

A wide range of risk factors have been found to be associated with caries in young children from the extensive literature available (Harris et al., 2004), with strong evidence of a young child being most likely to develop caries if they acquire S. *mutans* at a young age (Parisotto et al., 2010). However, limited studies have completely acknowledged the competing behaviour and interplay of a number of factors (diet, oral hygiene practices, fluoride, parental attitudes & beliefs and socioeconomic factors) that directly/indirectly influences the level of bacterial activity and in turn the development of caries. While family and broader socio-demographic influences on oral health have had an increased focus of attention, little research has examined how biological, behavioural and psychosocial factors interact to determine the onset and progression of caries in young children. There is a shortage of high quality studies using a longitudinal design (Harris et al., 2004) carried out in the youngest age groups using validated tools. Consequently, the development of caries in very young children remains less understood.

There is substantial evidence of socioeconomic inequalities in dental health commencing from childhood, worldwide and in Scotland (Reisine and Psoter, 2001; NDIP, 2014). Although various theories have been discussed to explain how inequalities form (Sisson, 2007), there is limited knowledge about the mechanisms that sustain the gradient. Whilst a number of studies have empirically tested the extent to which the socioeconomic gradient in oral health was explained by behaviours in adults and adolescents, only one study has been conducted among young children (Slade et al., 2006). Although behaviours did not fully account for the socio-economic gradient in oral health, none of the studies included explicit variables on all important oral health-related behaviours while using validated measures. There is speculation that material, behavioural and psychosocial factors need to be taken into account in a longitudinal study in explaining the SE inequalities in oral health (Sisson, 2007).

There is a need for preventive interventions to commence early in life, which requires a thorough understanding of the causes of the disease, and how these interact with socioeconomic factors in the youngest age group. In order to estimate the precise extent of the relation between specific risk factors and caries and their interaction with socioeconomic circumstances, a coherent disease model is required that evaluate both upstream and downstream factors. This model should also permit multivariable analyses to control for confounders and interactions and allow direction of effects to be modelled. Only with such a disease model will it be possible to investigate the relation between the occurrence of a determinant and dental caries, and to estimate the extent of this relation. In addition, it remains unknown how *Childsmile* interventions delivered at different times through childhood impact on these relationships and disease outcomes. This requires a longitudinal study design collecting data at the individual level prospectively. Cohort studies have to be large by design to test associations and directions of effect, whilst adjusting for putative confounding factors. The MRC recommends a feasibility and piloting phase to test the methods and procedures before using them on a large-scale to avoid methodological and logistical problems and adequately power future cohorts.

This study aims to explore the feasibility of building a cohort of Scottish young children to design a larger cohort study that will inform and evaluate the contribution of *Childsmile* interventions to oral health improvement and reduction of oral health inequalities in Scotland into the future.

Chapter 3 – Research Aims and Objectives

3.1 Aims

To assess the feasibility of recruiting and following up a cohort of preschool children and collecting data to investigate behavioural, biological and social factors associated with dental caries.

3.2 Objectives

1. To develop procedures and pilot tools and methodologies to be used in a large cohort study within *Childsmile* in the future, and identify challenges associated with such a study. In particular
   i. To assess recruitment, consent and participation metrics
   ii. To assess the feasibility of undertaking a detailed oral examination and plaque and saliva collection in nursery and school settings
   iii. To assess the feasibility of measuring heights and weights in addition to oral health assessment within nursery and school settings
   iv. To determine the response rate and completeness of questionnaires completed by parents
2. To produce Standard Operating Procedures for successful methods
3. To produce statistical models for associations between biological, behavioural and social factors and dental caries
   i. Risk models
   ii. Models explaining socio-economic gradient
4. To use these models to consider sample size requirements for a cohort study.

Chapter 4 – Methods and Materials

4.1 Study design

A longitudinal cohort study design was used to collect data prospectively on children aged four to five years followed-up one year later.

4.2 Hypothetical pathway

The study design, data collection and data analyses were driven by a hypothetical pathway. The hypothetical pathway shown in Figure 4-1 considered the multiple influences on caries, adapted from Fisher-Owens et al. (2007). Pathways were hypothesized based on previous findings from studies utilising sophisticated statistical techniques like structural equation modelling in other populations (Litt et al., 1995; Gao et al., 2010; Qiu et al., 2014).

This hypothetical pathway acknowledges the influence of the social drivers in dental health inequalities, the causes of the causes, as the key cause of caries. These social factors that are distal (farther in the pathway) to the outcome, pattern the more proximal influences on dental health such as oral health related behaviours through intermediary psychosocial pathways (Petersen, 2003).

According to the conceptual model in Figure 4-1, biological and behavioural factors may be considered as proximal risk factors, whereas social factors (SEP) may be considered a distal risk factor. The relationship of oral health related behaviours with socioeconomic position on one hand and caries on the other suggests that oral health related behaviours may play an important role in the observed socioeconomic inequalities in caries. The biological, behavioural, psychosocial and socioeconomic factors of interest will hence be collected in Sweep 1 and the main outcome measure, caries experience at Sweep 2.

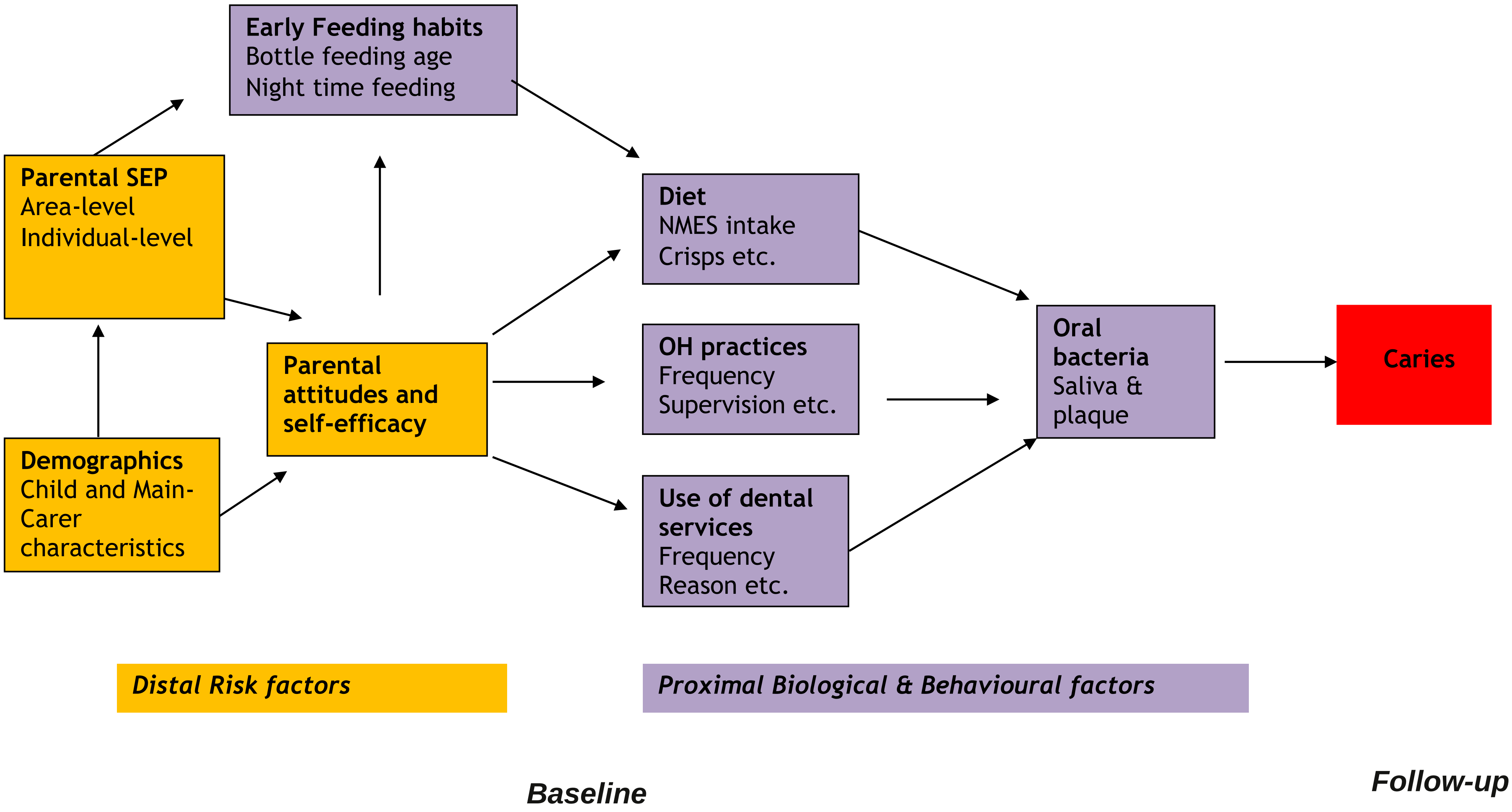


Figure 4-1: Hypothetical pathway related to the development of caries in young children

4.3 Study sample, sample size & setting

A random sample of Local Authority nursery schools within Glasgow City, who previously participated in the NDIP in 2009/2010 were stratified by the Scottish Index of Multiple Deprivation (SIMD) and caries prevalence. All children in the preschool year of nursery (aged four to five years typically) and their parents were eligible to participate in the study. The sample size for this pilot cohort study was approximately 200 preschool children and their parents. This was considered a reasonable sample size on which to conduct the pilot study over two academic years, with a single researcher and limited funding, whilst ensuring a reasonable amount of variation in key measures. Based on a recruitment rate of 40% from previous *Childsmile* study experience (Tsakos et al., 2012), to recruit 200 families, 600 families were to be invited. In addition, based on an average preschool nursery class roll of 40, we estimated the need to approach 14 nursery schools.

4.4 Ethical approval

4.4.1 Research Ethics Committee approval

The Research Ethics Committee (REC) application was completed online using the Integrated Research Application System (IRAS). The application was sent to the West of Scotland REC, along with the study protocol, participant invitation letter, participant information leaflets (PIL), consent forms and draft versions of the questionnaires that were developed for the purpose of the study. Following a meeting with REC 4 at the Glasgow Royal Infirmary in December 2010, attended by the principal supervisor and the author; ethical approval was granted with minor amendments for the conduct of a longitudinal study (REC reference- 10/S0704/1/62). The minor amendments included changes to the wordings of the PIL and format of the consent form. The ethical application included details of how informed signed consent was to be sought from parents and information on the inclusion and exclusion criteria for the study.

Inclusion Criteria

1. Attendance at a local authority nursery school that has undergone NDIP 2009/2010

2. Based in pre-school year
3. Positive consent from parent (return of signed consent form)

Exclusion Criteria
1. No positive consent from parent
2. Known chronic illness.

On liaison with the NDIP team coordinator, Dr Yvonne Blair after the first sweep (Sweep 1) of data collection it was apparent that not all of the participants in this study were sampled to undergo a detailed NDIP dental exam in the primary school year (2012) and will therefore have no outcome data in Sweep 2. A major amendment was thus applied for in December 2011. This amendment described a change in the Sweep 2 data collection process, from record-linking to the National Dental Inspection programme (NDIP) databases to carrying out a clinical dental examination within primary schools by the primary researcher. The committee gave a favourable opinion and approval was granted the same month (REC reference- 10/S0704/1/62).

Permission was also sought from the Education Services of the Glasgow City Council to approach nurseries within Glasgow City and invite them to take part in the study.

4.4.2 Sponsorship

Dr Debra Stuart from the University of Glasgow, Clinical Trials Unit acted as the Research Governance Sponsor for the study.

4.4.3 Funding

The study was funded by *Childsmile*; funded by the Scottish Government. Some minor costs (e.g. printing and postage of participant information packs) were borne by the University of Glasgow.

4.4.4 Recruitment and Consent

4.4.4.1 Sweep 1- Nursery Phase: Cross-sectional (March–June 2011)

Based on an average preschool class size of 40, head teachers at 14 nursery schools within Glasgow City were sent letters inviting their nursery to take part in the study and to attend a meeting at the Glasgow Dental Hospital and School (GDHS). The Head teachers' meeting explained the nature of the study and what would be involved. All nurseries were offered a contribution towards their funds as a token of appreciation (incentive). Following the meeting, names, dates of birth and postcodes of all preschool children from each nursery was requested. Figure 4-2 outlines the recruitment and data collection process that was involved in Sweep 1.

Information packs were delivered (by hand) to head teachers/assigned (March-June 2011) at the nurseries requesting distribution to all preschool children. The information pack contained an invitation letter and a participant information leaflet (Pack 1- Appendix B) addressed to the parent/guardian of each child based in the preschool year of the selected nurseries. This pack invited potential participants to take part and briefly explained what they would be asked to do. A week later a consent form and prize draw slip (Pack 2- Appendix C) were handed out by the nursery-staff in sealed envelopes addressed to the parent/guardian of each preschool child in the participating nurseries, which was to be returned to the nursery if they chose to take part. Children whose parents signed and returned the signed informed consent forms were included in the study. Parents/guardians were given a period of 2 weeks to return consent forms before a reminder leaflet was sent. Additionally, the head teachers at nurseries discussed participation with parents/guardians- some by sending text messages, others verbally.

On return of the consent form, through the nursery, three questionnaires were sent to the child's parent/guardian along with a covering letter, 'assistance for questionnaire completion' leaflet and free-post return envelope (Pack 3- Appendix D), requesting completion and return to the nursery. The leaflet offered to provide help in completing questionnaires either face-to-face with the researcher or over the phone. If help was needed, parents were asked to contact the researcher using the contact details that were given on the leaflet. A visit was arranged to the nursery school after agreeing with the nursery-staff for data collection and questionnaire collection.

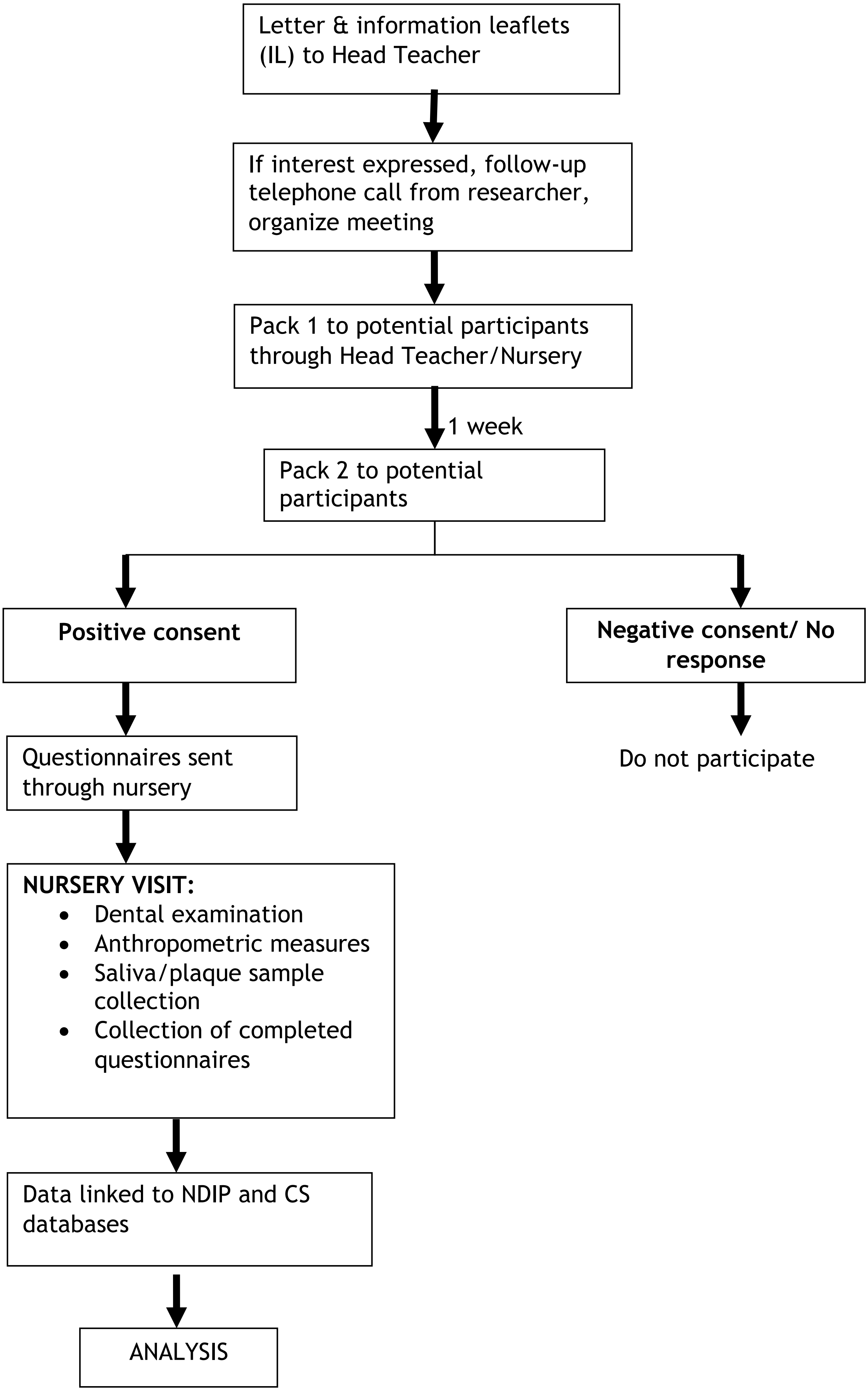


Figure 4-2: Flow diagram of the recruitment and data collection procedures in Sweep 1

4.4.4.2 Sweep 2- Primary School Phase: Longitudinal (April-June 2012)

Destination primary schools (53 primary schools) for all children who had parental consent were traced from nursery records and crosschecked at the Glasgow Education Services. Figure 4-3 outlines the follow-up and data collection procedures that were involved in Sweep 2.

All children who had a signed parental consent (n=219) were contacted for follow-up a year later (April- June 2012). Of particular focus were children who had complete data (clinical and questionnaire data) in Sweep 1. One of the following routes were used to follow-up children:

- Inviting the parent and child for a data collection session organized at the GDHS
- Visiting the child at their respective primary schools (located within Glasgow City only)
- Offering home visits for those parent-child dyads who had difficulty commuting to the Dental hospital or had relocated out with Glasgow city.

Head teachers at 32 primary schools within Glasgow City were sent emails introducing the study and requesting an opportunity to examine participating children in the primary-1 year who were a part of the study. This was followed up by a phone call from the researcher to explain the nature of the study and what would be involved.

Information packs were sent by post to head teachers at the schools, requesting distribution to participating children. The information pack contained a letter and a participant information leaflet (Pack 1) addressed to the parent/guardian of each child based in the primary-1 year of the schools. This pack informed parents about the follow-up data collection and briefly explained what they would be asked to do. It was made clear to parents that their participation was voluntary and they may withdraw from the study at any time. A week later, the children were seen by the researcher at their schools for data collection.

On the day of data collection, another pack (Pack 3) was distributed to the children. This pack contained a covering letter, ‘help’ leaflet, three questionnaires and a postage paid envelope addressed to the parent/guardian.

The letter requested questionnaire completion and return to the GDHS using the postage paid envelope. The leaflet offered to provide help in completing the questionnaires either face-to-face with the researcher or over the phone. If help was needed, parents were asked to contact the researcher using the contact details that were provided on the leaflet.

Attempts were made to follow-up children at the remaining 21 primary schools over the phone. The parent and child were invited for a scheduled data collection session organized at the GDHS or offered home visits. Parents were given the flexibility to choose a day and time of their choice; and offered travel reimbursements by bus or train if they chose to attend the organized event at GDHS.

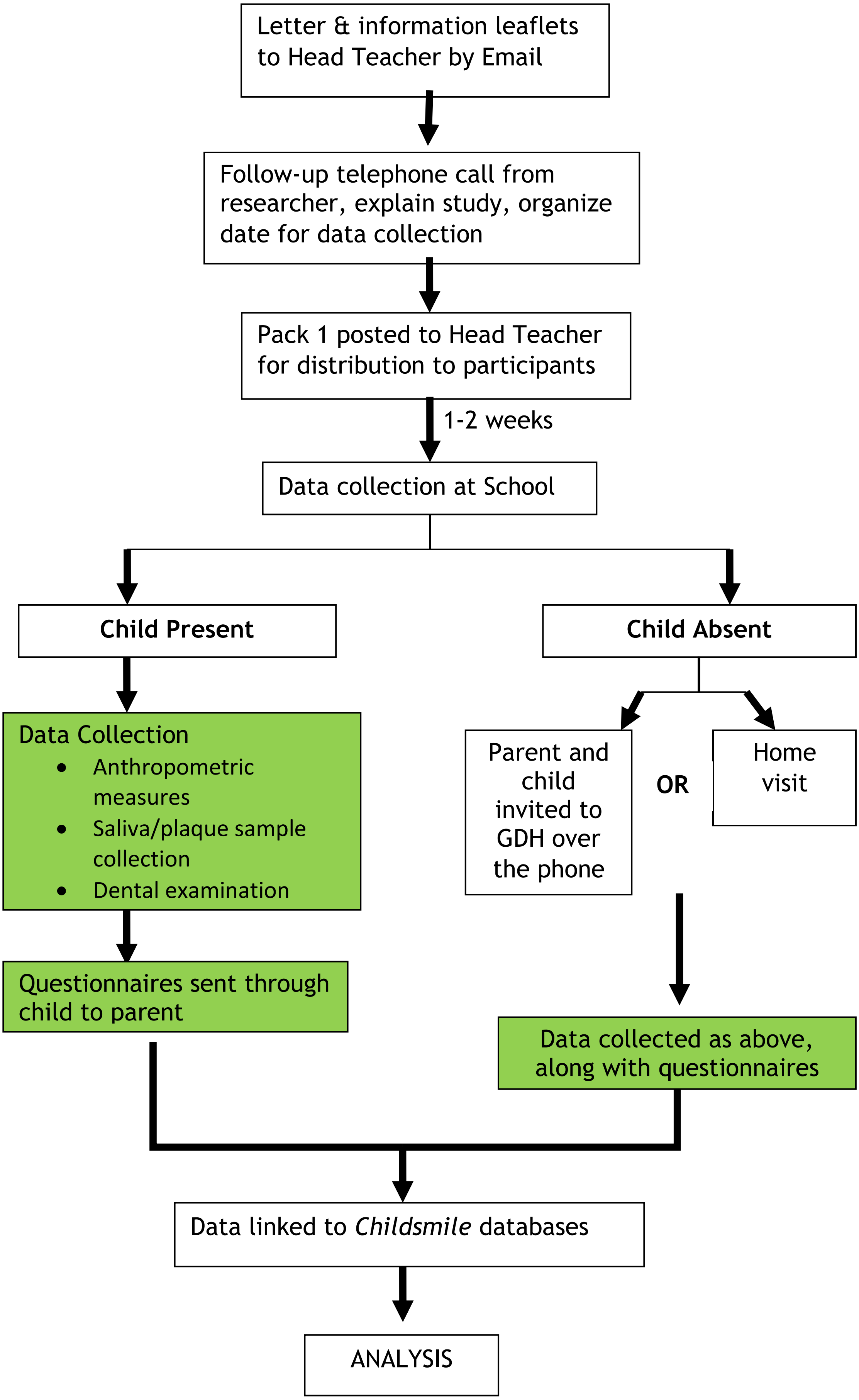


Figure 4-3: Flow diagram of the follow up recruitment and data collection procedures in Sweep 2

4.5 Data Collection

4.5.1 Development of Questionnaires

Two questionnaires, 'About your Study Child' (Appendix E) & 'About your family' (Appendix F) were developed for self-administration by parents/carers of preschool children. Both questionnaires were constructed in consultation with the literature and experts in the various fields and where possible, questions previously used in established population health surveys in Scotland were included (The Scottish Government, 2009a). Questionnaire items comprised a combination of open-ended (e.g. age at first dental visit); and closed questions with pre-defined response categories to which parents/carers were asked to respond by ticking a box. The questionnaires were piloted in December 2010. Following some minor changes to the wordings after the pilot in December 2010, the questionnaires were used in March 2011 (Sweep 1) and again in April 2012 (Sweep 2).

4.5.1.1 About Your Study Child questionnaire

This questionnaire was designed to gather data pertaining to the child taking part in the study. It was comprised of 24 questions, grouped into four domains: oral hygiene practices, early feeding habits; use of child dental services; and parental attitudes and beliefs around oral health (Appendix E).

4.5.1.1.1 Oral hygiene practices

This section of the questionnaire asked seven questions around oral hygiene practices that have previously appeared to show consistent association with caries in preschool children (Harris et al., 2004). The questions included- daily toothbrushing frequency, age at start of regular toothbrushing, toothbrushing just before bedtime and parental assistance/supervision at toothbrushing time. A further three questions were adapted from Conway et al.(2005); a question on rinsing behaviours after toothbrushing and two 'photograph menus' from which the parents were asked to indicate the quantity and brand of toothpaste used. The photographic menu was constructed using nearly all brands of toothpastes that were available for children in Scotland, and common family toothpastes in 2010/2011. This was used to assess if the toothpaste used at home contained the recommended minimum concentration of fluoride for preschool children (1000 parts per million).

4.5.1.1.2 Use of dental services

This section of the questionnaire asked three questions around the child's use of dental services. The questions were adapted from those used in established surveys (Survey of sugar intake among children in Scotland, 2008) including age at first dental visit, the reason for first visit and frequency of routine check up.

4.5.1.1.3 Early feeding habits

This section of the questionnaire aimed to explore early feeding practices as an infant in relation to the child taking part in the study. Six questions around early feeding practices previously known to be related to caries experience were adapted from established surveys (The Avon Longitudinal Study of Parents and Children (ALSPAC), 2007). The questions included if the child was ever breast fed, age at which bottle use was stopped, age at weaning, frequency of night time bottle feeding at two & four years of age and the frequency with which the amount of sugar intake was restricted at home.

4.5.1.1.4 Parental attitudes and beliefs

After reviewing the literature, a measure developed by health psychologists, based upon validated theoretical models of behaviour change, including the Theory of Planned Behaviour, Health belief Model and Health Locus of Control (Pine et al., 2004b), that was tested for validity and reliability in the population (Dundee, Scotland), was identified (Adair et al., 2004). However, the questionnaire in its full form contained 40 items. In a study of this nature investigating multiple risk factors in caries, it was necessary to employ brief items/scales that were reliable and valid, without over-burdening participants.

A cautious yet pragmatic approach was taken to include important attitudinal concepts as reviewed in ***Section 2.2.9.4.1*** and then built scales based on inter-item and item to scale correlations for this sample. Positive scores were given to favourable responses to oral health and negative scores to unfavourable. The sum of these scores formed a quantitative expression of parental attitudes towards oral health, with a higher score indicating favourable attitude. Cronbach's alpha was used to measure internal consistency of the scales.

In collaboration with a Behavioural psychologist (Dr Wendy Gnich), nine items were adapted from Adair et al. (2004). A further item was adapted from Finlayson et al. (2007) to assess parent/carer's belief in dental health fatalism. The parents/carers were asked to indicate their degree of agreement to each of the 10 statements using a 5-point Likert scale (from strongly agree to strongly disagree).

4.5.1.1.5 Other lifestyle factors

A single item was used as a proxy to measure sedentary behaviour. The question asked the average number of hours the child spent watching television or other such screen-based entertainment during school term (The Avon Longitudinal Study of Parents and Children (ALSPAC), 2007).

4.5.1.2 About Your Family questionnaire

This questionnaire was designed to gather data pertaining to the child's familial and socioeconomic circumstances (Appendix F). A questionnaire comprising of 14 closed-ended questions used in a number of other *Childsmile* studies was adapted for the purposes of this study. Additionally, some questions were drawn from those used in the Scottish Health Survey (Scottish Executive, 2009). For the ease of detailing and reporting results, the questions are grouped under domain names- child characteristics, main-carer characteristics and socioeconomic circumstances (not grouped in the original questionnaire that was used).

4.5.1.2.1 Child characteristics

Questions that intended to gather child characteristics included the ethnic background of the participating child- adapted from the Scottish Health Survey (2009); birth rank of child and total number of children in the household – adapted from previous *Childsmile* studies.

4.5.1.2.2 Main carer characteristics

Four questions were selectively chosen to gather information on who was completing the questionnaire, their age, marital status and smoking status; all of which were drawn from previous *Childsmile* studies.

4.5.1.2.3 Socioeconomic circumstances

Socioeconomic position was measured at both individual- and area-level. Individual-level measures included gross annual household income before tax and other deductions, education of the main-carer, receipt of benefits, housing tenure and last/current occupation of the main-carer (only measured at Sweep 2). Two questions were asked to the parent/carer about their education- the number of years of full time education after leaving secondary school, and the highest level of education achieved. In addition, at area-level SEP was assessed using the Scottish Index of Multiple Deprivation (SIMD) (Scottish Executive, 2009b). SIMD 2009 is now superseded by SIMD 2012. However, it is considered appropriate to use the SIMD closest to data collection (The Scottish Government, 2009).

4.5.2 Training and calibration

4.5.2.1 Sweep 1: Cross-sectional

Dental examination: The author/clinical examiner attended a one-day training at the GDHS in April 2010 for the NDIP Basic Inspection criteria. This course prepares dentists to carry out Basic inspections for the NDIP across Scotland. The training involved illustrated lectures and open discussion sessions on how to record the inspections.

Anthropometric measurements: The author/same clinical examiner attended another one-day training in September 2010 at the Yorkhill Hospital. The training involved illustrated lectures and videos, open discussions and practice sessions in collecting heights and weights of children according to Royal College of Paediatrics guidelines (RCPCH, 2013).

Microbiological sampling: The examiner was also trained in collecting plaque samples using a CPITN probe and unstimulated pooled saliva using Salimetrics Children's Swab (SCS) on a pilot sample of six children at the GDHS in March 2011. The SCS is a long absorbent device made of inert polymer designed to collect saliva from children under the age of six years. In addition, appropriate storage measures and methods for recovering saliva and processing were identified.

4.5.2.2 Sweep 2: Longitudinal

Dental examination: The clinical examiner (SS) attended a mandatory two-day annual training course that took place at the GDHS and Gartnavel IT suite in February 2012. This course allows dentists to standardize the conduct of detailed inspections for the NDIP across Scotland. The training involved illustrated lectures, open discussion sessions and IT training on how to record the inspections, based on the criteria set down by the British Association for the Study of Community Dentistry (BASCD) (Pitts et al., 1997). A further session trained SS and a second dentist (NZ) in pairs to accurately communicate codes and scribe examination details.

These were followed by clinical training sessions and calibration assessments involving nursery children from two local authority nursery schools. The results of all participating dentists were compared against an experienced dental examiner to ensure 'substantial agreement' (Landis and Koch, 1977) with respect to inter-examiner reliability of caries diagnosis at the d_3 level. Intra-examiner reliability was not assessed due to practical difficulties while collecting multiple data at multiple nurseries/schools in a limited amount of time during the day.

4.5.3 Main Study

Data were collected through:

- Clinical examinations at nurseries/schools for saliva & plaque samples, dental examination and height & weight measurements.
- A semi-quantitative food frequency questionnaire to explore habitual intake of a range of foods that were of interest to the study.
- Structured questionnaires to capture information on oral hygiene practices, use of dental services, early feeding habits, socio-demographics and parental attitudes &beliefs on oral health.

4.5.3.1 Clinical examinations

Clinical examinations were primarily carried out at nurseries/schools. All necessary infection control steps, including hand-hygiene and the use of disposable examination gloves were taken to ensure compliance with cross infection guidelines.

4.5.3.1.1 Microbiological sampling

It is recommended that the selection of the sampling method is based on the objectives of the study, age of the population and the method used in culturing organisms (Wan et. al., 2001). This study involved collecting various measures from about 200 preschool/primary school children in a nursery/school setting and it was essential that the technique was quick and acceptable to children and staff at the nursery/school. Since this study would archive samples for future use to test multiple analytes, it was decided to collect unstimulated whole saliva that pools in the floor of the mouth. This was also easier and aesthetically more acceptable in a nursery/school setting.

Trained dentist/s (SS and NZ in Sweep 1, SS in Sweep 2) collected saliva and plaque samples from children under nursery/school settings. Unstimulated whole saliva that pools at the floor of the mouth was collected using Salimetrics Children's Swab (SCS). Where possible children were discouraged from supervised *Childsmile* toothbrushing sessions and eating/drinking for at least one hour before the sample collection process. When this was not possible, a record was made of what and when the child had last eaten/drunk. Oral swabs were placed in the mouth for up to 60 seconds, and allowed to absorb saliva. Saliva-saturated swabs were then placed into sterile swab storage tubes (Salimetrics, Europe LTD), transported on ice to the microbiology laboratory.

Supra-gingival plaque was collected from the buccal aspect of the interdental region of primary molars in each quadrant using a CPITN probe and placed in 400 μL of double strength lysis buffer. If the primary molars were missing, supra-gingival plaque was collected from the labial aspect of the primary central incisors or lateral incisors in that order. Both the samples were immediately placed over ice and transported to the laboratory at Glasgow Royal Infirmary.

4.5.3.1.2 Dental examination

Sweep 1: Cross-sectional

It was originally proposed that data obtained from the NDIP for ante-preschool children would be record-linked and used in the cross-sectional analyses. However, it was known at the time of recruitment through the head teachers that there were many new children enrolled in the preschool year.

On further liaison with the NDIP team coordinator, Dr Yvonne Blair, it was apparent that not all of the participants would have undergone a detailed NDIP dental exam in ante-preschool year and will therefore have no outcome data in Sweep 1.

Dental examinations were therefore performed according to the NDIP Basic Inspection criteria using the Active/carious dentinal decay, Missing and Restored (AMR) index. All primary teeth were inspected under torch light using a visual examination with the help of a No. 4 plain mouth mirror and a ball end CPITN probe (ball diameter 0.5mm); and with children sitting. Teeth were classified as

- Sound
- Active/Carious dentinal decay (A)
- Missing due to caries (M)
- Restored due to caries (R)

Probe use was restricted to clearing debris, detection of sealants, restorations and cavitated lesions greater than 0.5 mm. Cotton wool rolls were used to clear teeth of debris and moisture where necessary. Suitable spectacles were used to protect the child's eyes when the torchlight was on. A tooth was considered as carious (A) if there was visible evidence of a cavity that involved the dentine. In addition, restored teeth with recurrent caries were classified as carious. The missing component included only those missing teeth thought to have been lost through caries. Conventions adopted for missing teeth- any missing posterior tooth was considered as extracted due to caries, while an incisor tooth was considered as missing when no successor was obvious or when definite caries was apparent in adjacent anterior teeth. All data collected were entered on to a data record sheet (Appendix G) and a score was calculated after adding up the total number of teeth showing active dentinal decay, missing due to caries and restored due to caries.

The AMR index has only been used for the Scottish NDIP and has never been quality assured. The proposed record-linkage ***(Section 4.6)*** was pursued and used to validate the Sweep 1 (2010-11) caries data collected using the AMR index.

Sweep 2: Longitudinal

Although the AMR index was used to quantify caries at tooth level in Sweep 1, a single dentist who was trained and calibrated according to BASCD criteria examined all children in Sweep 2, which was the primary outcome measure used in this body of work. All children were examined based on the BASCD criteria that record caries, which extends into the dentine (d_3 level- cavitated and visual) standardized for the collection of epidemiological data throughout the UK (Pitts et al., 1997); observing standard infection control protocols.

All examinations were carried out using a No. 4 plain mouth mirror and a ball end CPITN probe (ball diameter 0.5mm); and with children lying on a table, cushioned by a camping mat and an inspection light (Daray X100 with G desk mount) yielding approximately 4000 lux at 1 metre when the brighter of the two settings was used. Probe use was restricted to clearing debris, detection of sealants, restorations and cavitated lesions greater than 0.5 mm. Cotton wool rolls were also used to clear teeth of debris and moisture where necessary. Suitable spectacles were used to protect the child's eyes when the inspection light was on. The examiner was seated behind the child's head. Details of the examination were recorded on paper record sheets used for the NDIP, by the scribe (NZ) in the first instance and then transferred onto a fully encrypted Standard Staff Desktop7 computer running NDIP software version4 by the primary researcher. All data were entered and stored in secure folders on secure network drives in accordance with the Data Protection Protocol for the Dental Public Health Unit, which is aligned to the University of Glasgow Data Protection Protocol.

4.5.3.1.3 Anthropometric measurements

Trained dentist/s (SS and NZ in Sweep 1, SS in Sweep 2) measured heights and weights of all children according to Royal College of Paediatrics guidelines (RCPCH, 2013). Heights and weights were measured to the nearest 0.1cm and 0.1 kg using a stand-alone portable stadiometer (Seca 213) and a Class III Approved electronic flat floor scale (Seca 877) respectively. Heights were measured with children wearing no footwear and weights with children wearing light indoor clothing and footwear removed. Where this was not possible, this was recorded in paper for appropriate adjustments during analyses.

4.5.3.2 Questionnaires

A semi-quantitative Food Frequency Questionnaire and the questionnaires, 'About your study child' and 'About your family' described previously were used to collect data on a range of oral health related behaviours, socio-demographics and parental attitudes and beliefs. The questionnaires were sent home for parental completion through the nursery and returned to the nursery in Sweep 1 and to the GDHS by free-post in Sweep 2.

4.5.3.2.1 Dietary assessment

A semi-quantitative Food Frequency Questionnaire (FFQ) (version C2, Scottish Collaborative Group) previously validated among preschool children was used for collecting dietary data (Survey of sugar intake among children in Scotland, 2008). It is designed to assess habitual diet usually in studies of 100 or more subjects whose diet has remained stable over a period of at least three months and can be administered by interviewer face-to-face, over the telephone or can be self-administered by the participant (main-carer).

The FFQ lists 140 food/drinks with a measure for each item. A photograph was attached to the FFQ to standardize the estimation of the measures. Main-carers were asked to describe their child's diet over the previous three months by giving an estimate of the frequency and amount of each food or drink consumed in a typical week by choosing from one of the nine options, ranging from 'rarely or never' to '7 or more measures per day'. The FFQ was primarily self-administered in this study, except for one parent who requested a face-to-face administration.

4.6 Record linkage

NDIP offers routine dental examination by trained and calibrated dentists based on the BASCD methodology (Pitts et al., 1997). Caries experience is evaluated using the dmft index (decayed, missing and filled deciduous teeth) and dmfs index (decayed, missing and filled deciduous surfaces). The NDIP data for ante-prechool children collected in 2009, hosted within the

Information and Statistics Division (ISD) of the NHS National Services Scotland (NHS NSS), was contacted for access in June 2010. Access was approved by the Caldicott Guardian for NHS NSS, and a Confidentiality Statement was signed by the Principal Supervisor (AS) and Professor Lorna Macpherson as Study Sponsor.

The NDIP data for ante-preschool children collected in 2009-10 was linked to consented participants included in the study through their respective dates of birth and home postcodes. This was used to validate the Sweep 1 (2010-11) caries data collected using the AMR index.

Given the strong evidence that fluoride varnish substantially reduces caries increment (Marinho et al., 2013), all data (clinical and questionnaire data) were planned to be record-linked to *Childsmile* databases that held information regarding fluoride varnish applications at *Childsmile* practice and nurseries/schools. The record linkage was partially successful and data was available for fluoride varnish applications carried out via *Childsmile* practices held within the CERT, Dental Public Health Unit, University of Glasgow. However, there were difficulties with record linking to *Childsmile* nursery/school databases, held with the Information Services Division (ISD), Scotland. The children in this study did not have a Community Health Index (CHI) number, which was the main key for linkage within ISD. The linkage would further need a Privacy Advisory Committee (PAC) approval. It was not possible to have this completed within the limited timeframe prior to analyses. The record-linkage is planned to be pursued for future analyses.

4.7 Measures to increase recruitment and response rates

A decline in response rates to national surveys (Scottish Executive, 2005), research studies and postal questionnaires (Galea and Tracy, 2007) have been observed in recent years. This study used a range of strategies to increase response to the study and questionnaires as noted in the Cochrane Collaboration review by Mapstone et al. (2007) and Edwards et al. (2009).

4.7.1 Incentives

The 13 nurseries that took part in Sweep 1 were offered an equal unconditional incentive for the support the staff offered and any possible disruption the study might have caused. The value of the incentive was calculated at the end of the study as £125 and was issued in the form of a cheque. No such incentives were offered to Primary schools in Sweep 2 as children from the 14 nurseries had spanned out to more than 50 primary schools with each school having on average four children. It was beyond the scope of a pilot study of this nature to offer such incentives to all primary schools.

In addition, names of all children who had complete data (clinical and questionnaire) and returned a prize draw slip with contact details were entered into a prize draw. The prizes were issued as shopping vouchers worth £100 as first prize, £50 as second prize and £25 as third prize. This was done at both Sweep 1 and Sweep 2.

4.7.2 Reminder leaflets

Two reminder leaflets were used in Sweep 1. One of the leaflets was distributed by the primary researcher at a nursery to parent/carers who came to pick children after nursery. This was also an attempt to give parents an opportunity to ask questions about the study. However, this leaflet was discontinued from use after testing in just one of the nurseries, as it did not improve the recruitment rate when compared to a nursery where no such leaflets were used. Nursery-staff distributed the second leaflet 2 weeks after Pack 2 distribution reminding parents/guardians to return consent forms.

Only one reminder leaflet was used in Sweep 2. This was directly posted to the family for questionnaire completion and return, when questionnaires were not received at the GDHS two weeks past the day Pack 3 was sent to parents.

4.7.3 Encouragement from Head-teachers/Staff

The staff at each nursery in Sweep 1 used their discretion to encourage participation and questionnaire completion & return. The various techniques reported to have been used included the head teacher sending text messages using a mobile phone, word of mouth reminder at the nursery entrance and participation encouragement through nursery newsletters. One nursery had a designated Oral Health Team Leader. This individual closely followed-up parents of children on a one-to-one basis.

4.7.4 Follow-up calls, events, home-visits

Parents/guardians who did not return completed questionnaires three weeks past the date for return of questionnaires received up to two follow-up phone calls. Parents were offered support in completing questionnaires over the phone. In the event where the main-carer was unreachable/non-contactable, the phone numbers were tried on repeat occasions up to five times at varying times of the day/week. An attempt was also made to follow-up children who could not be examined at their schools due to sickness/leave (n=20 in Sweep 1, n=7 in Sweep 2) by contacting the parent over the phone to arrange an organized data collection session at the GDHS. For those who were unable to attend the organized session, a home-visit was arranged (n= 6).

4.7.5 Free-post return envelops

A free-post envelope was included in all Pack 2 (questionnaire Packs) in Sweep 2 for the parents/carers to directly post back the questionnaires to GDHS. All follow-up phone calls in both Sweeps were followed by additional questionnaires and free-post envelopes posted to the family.

4.8 Data management & Quality control

Each subject was allocated a code and the microbiological samples, clinical data and questionnaire data allocated a corresponding code. Only the primary researcher and principal supervisor had access to the information linking the participant code to the participant name, which was retained in a secure database, separate from the data for analysis, which was password protected.

4.8.1 Dental caries data

Sweep 1:Cross-sectional

Clinical caries data from paper records were first entered into MS-Excel and then exported into SPSS for Windows (Version 18.0). Caries experience (primary outcome) for each child was evaluated by adding up the total number of teeth that exhibited active dentinal caries (A), missing teeth (M) and restored teeth (R) to yield an AMR score. Caries experience was dichotomised as children with obvious caries experience or no obvious caries experience.

The NDIP data for ante-preschool children collected in 2009-10 was record-linked to consented participants included in the study through their respective dates of birth and home postcodes. This was used to validate the Sweep 1 (2010-11) caries data collected using the AMR index.

Sweep 2: Longitudinal

Data from paper records were entered into NDIP Detailed inspection Access databases at the tooth surface level. The data was exported into MS-Excel format and sent to a Consultant Epidemiologist (Dr Alex McMahon) who has expertise in converting surface level (dmfs) data to tooth level (dmft).

4.8.2 Microbiology

Saliva was recovered from the swabs by centrifugation at 4100 rpm for 10 min at 4℃ (ALC International, PK 120 R, Milan, Italy) within 3-4 hours of collection. The total volume of saliva collected from each participant was recorded and aliquots of 60 μL, 130 μL and 20 μL were stored at -80°C for estimation of cortisol levels, salivary antimicrobial proteins (AMPs) and salivary IgA antibodies respectively. Samples were immediately frozen at -70°C for later quantitation of cariogenic bacteria by TaqMan® real time qPCR. Salivary concentrations of antimicrobial peptides and IgA antibodies were assessed by Enzyme Linked Immunosorbent Assay (ELISA). The plaque samples were vortexed for 10 seconds and stored at –70°C for later analysis of oral microbiogenome content.

4.8.3 About your study child/family questionnaire

Data from the questionnaires were entered directly into SPSS for Windows (Version 18.0). Limits were placed on acceptable values and acceptable data types (numeric, string and scale). Categorical variables with categories containing only a few observations were collapsed based on frequency tabulations.

Educational classification was based on the highest level of qualification obtained and was categorised as Level 0 (no qualification or pre-school leaving qualification), Level 1 (O grade, standard grade, GCSE or equivalent), Level 2 (Higher grade, A level, GSVQ advanced or equivalent), Level 3 (HNC, HND, SVQ Levels 4 or 5 or equivalent) and Level 4 (first degree, higher degree or professional qualification) (Stamatakis et al., 2009).

The current or last occupation of the parent/carer were analysed using the Standard Occupation Classification (SOC) (2010) and subsequently collapsed according to NS-SEC system (2005), with NS-SEC 1 including Higher Managerial, Administrative & Professionals, NS-SEC 2 including intermediate occupations and NS-SEC 3 including routine & manual occupations.

4.8.4 Food Frequency Questionnaire (FFQ)

Data from completed FFQs were entered into an MS Access database. Energy and nutrient intakes were estimated by nutritionists at the University of Aberdeen from a package based on the Food Standards Agency National Diet and Nutrition Survey nutrient database (2004) that estimate NMES as all sugars in fruit juices as well as table sugar, honey; sucrose, glucose and glucose syrups added to foods and added sugars or syrup in preserves. Additionally, sugars naturally present in foods that were canned, stewed, dried or used in preserves were classified as half extrinsic. NMES intake was expressed in grams/day. In addition, NMES was calculated as a percentage of total food energy (assuming the energy content of 16 kJ/g) and categorized into tertiles of intake, ranging from T1 (highest intake) to T3 (lowest intake). Three food groups that were hypothesized to significantly contribute to NMES (Biscuits, Cakes & Pastries, Confectionary and Non-Diet Soft Drinks) were derived. In addition, a food group ‘Crisps & Savory Snacks’ was derived which has previously been shown to be associated with an increased risk of having had treatment for decay (Sheehy et al., 2008).

4.9 Statistical analysis

All data management and analyses were performed in IBM SPSS software, (PASW version 18).

4.9.1 Data cleaning/Quality check

The quality of data entry of a random 10% sample was checked. In addition, an error and inconsistency check was carried out using histograms, frequency distributions and cross-tabulations for all data.

Diet: FFQs with more than ten missing responses were excluded from analysis to avoid possible underestimation. In addition, for quality assurance of complete FFQs, acceptable margins of reported energy intakes were defined as those within -/+ 2 standard deviations of the mean.

Parental attitudes and beliefs: Two scales ‘positive parental attitude scale’ and ‘parental efficacy scale’ were constructed as described in ***Section 4.5.1.1.4.*** Cronbach’s alpha was used to assess the internal reliability.

Anthropometry: Body Mass Index (BMI) was calculated as weight in kilograms divided by height in meters squared. Body mass indices were converted to standard deviation scores relative to the UK reference data in 1990 (Cole et al., 1995) using the Lambda Mu Sigma (LMS) growth programme (Cole and Pan, 2002). The BMI standard deviation scores (z scores) were quality checked by generating a scatter plot with BMI z scores at Sweep 1 against BMI z scores at Sweep 2. For quality assurance, acceptable margins of BMI z scores were defined as those within +/- 4 standard deviations. All children were classified as normal, overweight or obese according to sex and age-specific BMI cut-off points (Cole et al., 1995).

Microbiology: The distributions of data within each microbiological variable were investigated using frequency histograms. Skewed data were $\log_{10}$

transformed to obtain a normal distribution and summary statistics produced as detailed in ***Section 4.7.2***.

The cross-sectional analysis was undertaken as a part of quality control, the results of which are presented in Appendix H.

4.9.2 Descriptive Statistics

Descriptive statistics were produced for each variable. Table 4-1 provides the list of variables by domain names and their factor level of analysis used. Frequency distributions were produced for categorical data and histograms were used to check for distributions of continuous data. Means and standard deviations and 95% confidence intervals (CI) were used to summarize continuous data that were symmetrically distributed and medians, quartiles and range when the data was skewed. Mean AMR/$d_3$mft and the proportion of children with no caries experience were calculated.

4.9.3 Univariable analysis

Outcome variable: Caries experience at Sweep 2

Explanatory variables: Biological, behavioural, psychosocial and socioeconomic factors tabulated in Table 4-1 measured in Sweep 1

All risk factors tabulated in Table 4-1 measured in Sweep 1 were univariably related to caries experience in Sweep 2. Associations between two categorical variables were analysed using cross-tabulations and Chi-squared statistics or Fisher's exact test. Students t-tests or Analysis Of Variance (ANOVA) were used for continuous variables. Binary logistic regression was used to produce unadjusted odds ratios (OR) and 95% confidence intervals (CI) for caries experience.

Table 4-1: Putative risk factors & confounders and their factor level of analysis for caries evaluated in Sweep 1

Variables	Factor level of analysis
Demographics	
Child Characteristics	
Age	Continuous
Gender	Male, Female
Ethnicity	White: Scottish, Others
Number of children in family	1, 2, 3 or more
Birth rank	1st born, 2nd born, 3rd born or later
Main-Carer Characteristics	
Age of main carer	<25 years, 26-35 years, 36 years and over
Relationship between the study child	Father, Mother, Grand parent
Smoking status of main-carer	Current smoker, Not current smoker
Marital status of main-carer	Married/Cohabiting, Single parent
Socioeconomic position	
Individual level	
Gross annual household Income	<£10,000, 10,000 to 19,999, £20,000 and above
Percentage of income from benefits	None to quarter, half to all
Main-Carer's highest level of Education	Level 0: no qualification or pre-school leaving qualification
	Level 1: O grade, standard grade, GCSE or equivalent
	Level 2: Higher grade, A level, GSVQ advanced HNC, HND, SVQ Levels 4 or 5 or equivalent
	Level 3: first degree, higher degree or professional qualification
Main-Carer's Years of Full-time education after leaving secondary school	Not completed secondary school, Further education
Current or last occupation of Main-Carer	Never worked or permanently unemployed due to disability
	NS-SEC 3 - Routine & manual NS-SEC 2 - Intermediate NS-SEC 1 - Higher Managerial, Administrative & Professionals
Housing tenure	Rented: Housing association/Local authority, Rented: privately, Owned/mortgaged
Area level	
Local SIMD	Q1 (most deprived), Q2, Q3, Q4 (lest deprived)
National SIMD	Q1 (most deprived), Q2, Q3, Q4 (lest deprived)
Behavioural risk-factors	
Early feeding habits	
Early feeding	Breast fed, Bottle fed, Breast and bottle fed
Duration of bottle feeding	Never used, under 12 months, Between 1 and 2, Later/Still using a bottle
Night time bottle feeding at two years	Always/Most days, some days, Never
Night time bottle feeding at four years	Always/Most days, some days, Never
Age of commencement of solids	<4 months, 4-6 months, >7 months
Frequency of sugar restricted	Always/Most days, some days, Never

Table 4-1 continued

Variables	Factor level of analysis
Oral hygiene practices	
Daily brushing frequency	Less than twice, More than twice
Brush immediately before Bedtime	Always, Sometimes/Occasionally, Hardly ever/Never
Age when brushing commenced	Under 12 months/First tooth erupted, After 12 months
Adult brushing supervision/assistance	Yes, No
Method for toothpaste removal	Swallow, spit, Rinse
Fluoride concentration in toothpaste	Up to 550 or less than 1000 ppm, >1000ppm
Amount of toothpaste used	Smear, Pea-size, Half a brush, Full brush
Use of dental services	
Age at first dental visit	Never, less than 6 months, 7- 12 months, Later
Reason for first dental visit	Never, Anticipatory, Corrective
Frequency of Routine check up	< 6 months, At least every 12 months, >12 months/Never
Diet – Semi quantitative FFQ	
NMES (% food energy)	Continuous
NMES (% food energy)	<11%, >=11%
NMES (g/day)	T1: lowest intake, T2 and T3: highest intake
Total Sugars (g/day)	T1: lowest intake, T2 and T3: highest intake
Food groups	
Crisps & Savory Snacks (g/day)	T1: lowest intake, T2 and T3: highest intake
Biscuits, Cakes & Pastries (g/day)	T1: lowest intake, T2 and T3: highest intake
Confectionary (g/day)	T1: lowest intake, T2 and T3: highest intake
Non-Diet Soft Drinks (g/day)	T1: lowest intake, T2 and T3: highest intake
Microbiology	
Total salivary *S. mutans* CFU/ml	T1: not detected, T2: 1 to 4.0 x 10^4, T3: 4.0 x 10^4 and above
Parental attitudes & beliefs	
Parental attitudes scale	6 items continuous scale running from negative to positive (worst to best) • My child losing a baby tooth due to tooth decay would be upsetting • It is worthwhile to give my child sweets/biscuits to behave well • Most children eventually develop dental cavities • As a family, we intend controlling how often the child has sugary foods • It is not worth it to battle with my child to brush his/her teeth twice a day • It is often too stressful to say no to my child when they want sweets
Parental efficacy scale	3 items continuous scale running from negative to positive (worst to best) • It is worthwhile to give my child sweets/biscuits to behave well • It is not worth it to battle with my child to brush his/her teeth twice a day • It is often too stressful to say no to my child when they want sweets
Sedentary behaviour	
Daily hours of television viewing during school term	Less than 1 hour, 1-2 hours, 3 or more hours

4.9.4 Multivariable analysis

Figure 4-4 provides a visualization of the modelling strategy. It should be noted that as this was primarily a pilot study, and based on a sample of more deprived children the estimates may be biased and not generalizable to the general population. Due to large number of variables measured, the pilot study will not be adequately powered to detect certain effects and may show wide confidence intervals. Hence, a p-value of 0.20 was set and exact methods of calculating confidence intervals were employed (Burton et al., 1998).

Binary logistic regression was used to produce unadjusted and adjusted odds ratios (OR) and 95% confidence intervals (CI) for caries experience (dichotomised $d_3$mft=0 or >0) according to putative risk factors, both cross-sectionally and longitudinally. In addition, the C-index [95% CI] (Altman and Bland, 1994) was estimated to assess the importance of risk factors in predicting caries.

Following the modelling strategy determined by the hypothetical pathway in Figure 4-1, the selection of independent variables in the multivariable analysis was based on a P value being <0.20 and a lower limit CI of C-index >0.50. Multivariable models were run with putative risk factors for caries experience at Sweep 2 simultaneously within each of the five domains (oral hygiene practices, use of child dental services, early feeding habits, current diet and parental attitudes and beliefs around oral health). Factors with P value <0.20 at the within domain stage were then entered into a final model in which all risk factors were analysed simultaneously. Due to *S. mutans* being more proximal in the hypothetical pathway (Figure 4-1) and therefore expected to show stronger associations with caries than all other risk factors (thus possibly eliminating the relatively more distal factors), two final models were run- one with all the relevant risk factors excluding *S.mutans* counts, and the other with the inclusion of *S.mutans* counts. The final models were further adjusted for the variables, gender and age. Finally, the Receiver Operating Characteristic (ROC) plot was produced and C-index estimated for the overall model each time to assess the overall predictive ability of the multivariable models.

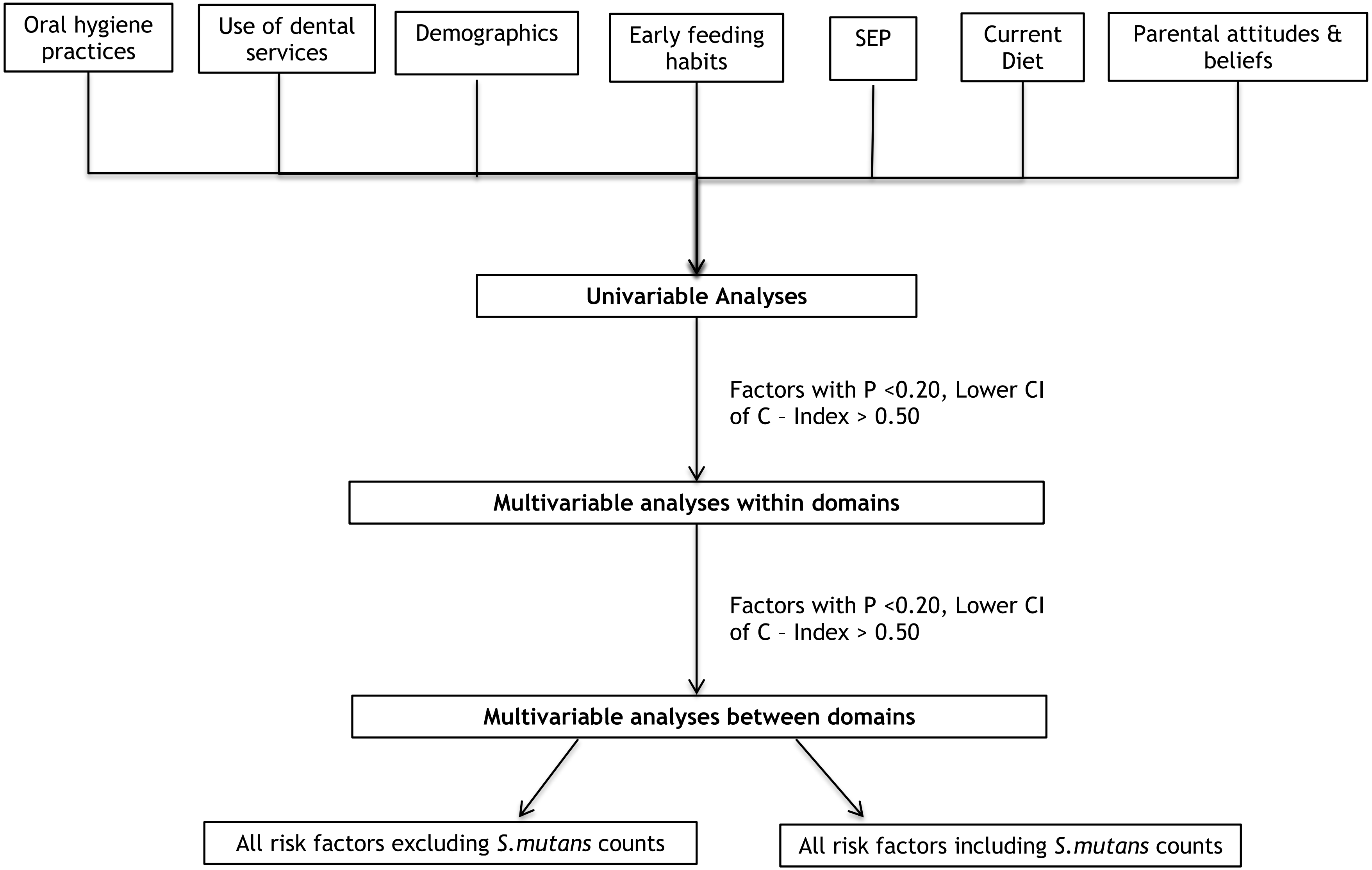


Figure 4-4: Modelling strategy involved in the statistical analyses for exploring associations between various risk factors and dental caries

4.9.5 Socioeconomic gradient analysis

In order to explain the socioeconomic gradient in caries, binary logistic regression was used to produce odds ratios and 95% confidence intervals (CI) for caries experience according to all SEP measures with a p value < 0.20 and lower CI of C-index >0.50 in the univariable analysis. These were adjusted for age, gender and the risk factors that emerged from the final multivariable model- first one domain at a time and then a final model which considered all factors from all domains.

4.9.6 Relative index of inequality

Due to differences in measures of SEP and variations in the percentage of population in each category within a SEP measure, the Relative index of Inequality (RII) was calculated. The RII is a regression-based measure widely used in social inequalities research ***(Section 2.3.7.2)***. It provides a standardized metric to compare the extent of inequality between different measures of SEP or with same measure of SEP over time. It takes into account the size of all the categories across the socioeconomic strata than just the extreme categories (Mackenbach & Kunst, 1997).

RII was derived by ranking the participants on each of the socioeconomic measures from the lowest to the highest (Batty et al., 2006). The rank scores were divided by the sample size to generate a value between 0 and 1.
RII was calculated between each measure of SEP and caries. Then, putative risk factors from the final multivariable model were considered in the model- first one domain at a time and then a final model, which considered all factors from all domains. The level of attenuation was expressed by the percentage reduction in RII using the formula, percent reduction = (Unadjusted RII – Adjusted RII)/ (Unadjusted RII - 1) X 100) (Singh-Manoux et al., 2005).

Chapter 5- Results: Feasibility of building a cohort

This chapter addresses objectives 1(i) to (iv)

5.1 Recruitment of participants

5.1.1 Sweep 1 (Preschool year at nursery)

A detailed breakdown of the participation rate is shown in Figure 5-1. Of the 14 nursery schools invited to take part in the study, 13 nurseries expressed an interest and 1 declined due to staffing issues at the nursery. This gave an eligible sample of 623 children. Letters along with participant information leaflets and consent forms were sent to parents via nursery schools. Two hundred and twenty eight (37% of total eligible) parents responded; two hundred and nineteen parents returned a positive consent and nine returned a negative consent. Of the 219 (35%) children consented for the study, 190 children had clinical data (microbiological samples, dental data and anthropometric data) and 179 had questionnaire (About your study child, About your family and semi-quantitative food frequency questionnaire (FFQ)) data. One hundred and sixty five children had complete data (clinical and questionnaire data).

5.1.2 Sweep 2 (Follow-up one year later at primary school)

Children from the 13 nursery schools in Sweep 1 were dispersed across 53 different primary schools within Glasgow City. One hundred and thirty nine children were clinically examined and sampled across 32 primary schools within Glasgow City, and 36 children at the organized session at Glasgow Dental Hospital & School (GDHS).

Overall, a total of 175 children were clinically examined and sampled in Sweep 2, of which 144 children had complete data (clinical and questionnaire) from Sweep 1.

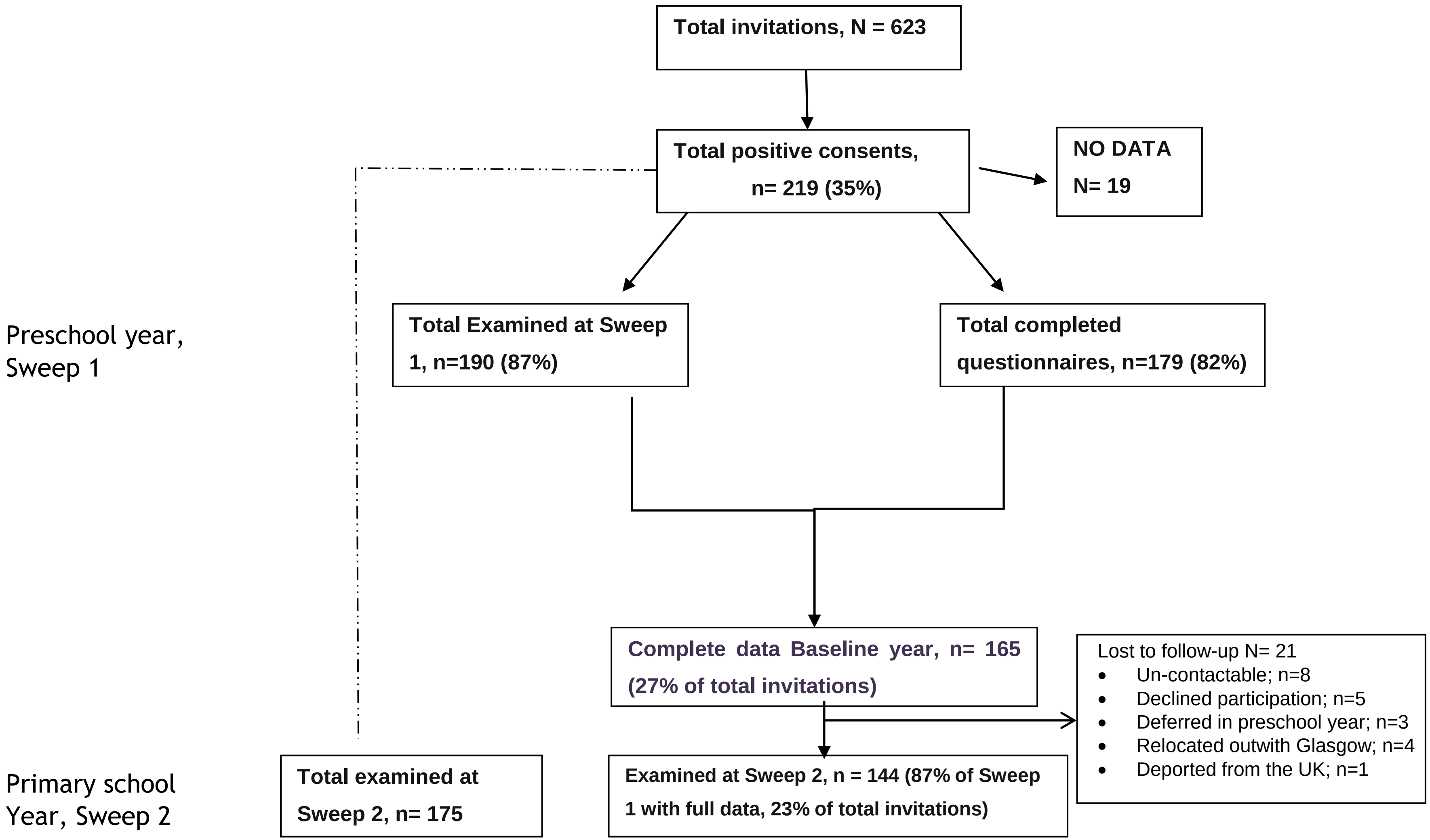


Figure 5-1: Response to Study/Examination/Questionnaire

5.2 Consent rates according to nursery school

Table 5-1 illustrates the consent rates across the 13 nursery schools in Sweep 1. The consent rates ranged from 12% to 84% and there was no obvious pattern by level of deprivation based on nursery postcode. Notably, the nursery (Nursery C) with the highest consent rate (84%) also had the highest proportion of children from the most deprived SIMD quintile (91%).

The researcher or the head teacher speaking to the parents in advance of distributing the information sheets did not improve the consent rate (Nursery B) compared with nurseries where no such methods were used. As expected the consent rate from nursery schools that ran over the summer holidays (highlighted grey in Table 5-1) was erratic.

Table 5-1: Consent rates by nursery

Nursery	SIMD quintile[2]	Total class size (n)	Proportion of children living in SIMD quintile 1[1] (%)	Proportion of children who were consented % [n]
Nursery A	1	63	66	32 [20]
Nursery B	1	61	89	37 [21]
Nursery C	1	32	91	84 [26]
Nursery D	1	42	95	31 [12]
Nursery E	1	53	81	32 [17]
Nursery F	4	70	3	44 [31]
Nursery G	1	45	76	32 [13]
Nursery H	2	90	8	33 [30]
Nursery I	1	50	55	46 [23]
Nursery J	2	32	82	12 [9]
Nursery K	1	44	79	24 [5]
Nursery L	1	22	82	50 [10]
Nursery M	1	19	79	21 [2]

[2] Based on nursery postcode- Scottish Index of Multiple Deprivation 2009 at National level (1- most deprived)

Nurseries running over summer holidays

5.3 Socioeconomic profile of those eligible, those consented and those that did not consent

Due to purposively over-sampling from a deprived area of Scotland (i.e. Glasgow City), there was a higher than expected proportion of children from more deprived areas. The socioeconomic distribution of those who consented was not appreciably different to those who did not consent (p = 0.70) (Table 5-2), suggesting those participating in the study were broadly representative of the population from which they were sampled. At follow-up, the socioeconomic distribution was similar to that at baseline (p = 0.14) (Table 5-3).

Table 5-2: Area-based socioeconomic distribution of those eligible to participate, those consented and those that did not consent in Sweep 1 & 2

Socioeconomic Variable	Total eligible to participate % [n]	Consenters % [n]	Non-consenters % [n]
N total	100 [623]	35.1 [219]	64.9 [404]
SIMD[3] Quintile			
Q1 (most deprived)	54.6 [340]	56.2 [123]	53.7 [217]
Q2	11.7 [73]	11.4 [25]	11.9 [48]
Q3	15.7 [98]	18.7 [41]	14.4 [58]
Q4 & Q5 (least deprived)	11.7 [73]	12.7 [28]	11.1 [45]
Missing	6.3 [38]	0.9 [2]	8.9 [36]
P value		0.702[4]	

[3] Scottish Index of Multiple Deprivation 2009 at National level

[4] Pearson's Chi-square test to assess whether the SIMD distribution of consenters were significantly different from non-consenters

Table 5-3: Socioeconomic profiles of consenters, participants in Sweep 1 & 2

Socioeconomic Variable	Consenters % [n]	Participants[5] Sweep 1 % [n]	Participants Sweep 2 % [n]
N total	35.2 [219]	86.8 [190]	79.5 [175]
SIMD[6] Quintile			
Q1 (most deprived)	56.2 [123]	53.7 [102]	57.5 [100]
Q2	11.4 [25]	13.2 [25]	11.5 [20]
Q3	18.7 [41]	18.4 [35]	17.8 [31]
Q4 & Q5 (least deprived)	12.8 [28]	13.7 [26]	12.2 [21]
Missing	0.9 [2]	1.1 [2]	1.2 [3]
P value		0.137[7]	

5.4 Feasibility of collecting clinical data

5.4.1 Acceptability and Quality of data collected

5.4.1.1 Dental caries data

The methods used to collect caries data were adapted from the National Dental Inspection Programme (NDIP), which has previously been proven acceptable among ante-preschool children and primary-1 children. This study additionally verified acceptability among preschool children.

Clinical examinations were primarily carried out at nurseries/schools and on average lasted for four minutes per child. Dental caries data was successfully collected from all children from whom data collection was attempted.

The NDIP data for ante-preschool children collected in 2009-10 was linked to consented participants included in the study through their respective dates of birth and home postcodes for validating the Sweep 1 (2010-11) caries data collected using the AMR index. Ninety-five children (50% of those with clinical data in Sweep 1) participating in the study were successfully record linked with their ante-preschool NDIP data. There was 'substantial agreement' in

5 Participants are defined as those who have clinical data

6 Scottish Index of Multiple Deprivation 2009 at National level

7 Pearson's Chi-square test to assess whether the SIMD distribution of participants in Sweep 1 were significantly different from participants in Sweep 2

the $d_3$mft at individual level, with 88% of the cases matching, yielding a kappa value of 0.75 (Landis and Koch, 1977; NDIP, 2014). Four children who were scored to have caries experience in the NDIP were classified as having no caries in Sweep 1, implying some underestimation of caries in Sweep 1 or possibly overestimation of caries in the NDIP. Nine children who were scored to have no caries in the NDIP were classified as showing experience of caries in Sweep 1. It is difficult to comment if this was true caries progression or an overestimation.

One participant was excluded from analyses due to inconsistency between caries status in Sweep 1 and Sweep 2. A thorough investigation was carried out to find the cause of the inconsistency by going back to paper records of caries data recorded at three years collected at the NDIP, at Sweep 1 and Sweep 2. One possible explanation for the inconsistency is a transcription error. For example entering a wrong code or illegible writing on the paper record would significantly alter the resultant $d_3$mft value.

In summary, it was feasible to collect clinical data in a nursery and school setting on children of this age group.

5.4.1.2 Microbiological data

The methods used to collect plaque and saliva samples from young children proved acceptable both among nursery-staff and the children themselves. One hundred and ninety children had samples collected at Sweep 1 and 175 at Sweep 2. Twenty-two participants in Sweep 1 and twenty-five participants in Sweep 2 were omitted from analyses due to insufficient volumes of saliva available for all analyses.

The methods used for plaque sample collection in this study was specifically for the analysis of oral microbiogenome content and formed a separate piece of work. The analyses of the plaque samples were pending and could not be used for the quantification of *S.mutans* in this study.

Although the Salimetrics Children's Swab (SCS) was used to absorb saliva from the mouth of every participant for up to 60 seconds, the volume of saliva recovered varied from 60 μL to 1560 μL (Table 5-4). Standard Operation

Procedures were developed for saliva and plaque sampling and are available in Appendix I.

Table 5-4: Volume of saliva (μL) recovered in both Sweeps using Salimetrics Children's Swab for approx. 60 seconds

	Volume of Saliva (μL), N=190	Volume of Saliva (μL), N=159
	Sweep 1	Sweep 2
Mean [SD]	516.2 [358]	447.1 [380]
Minimum, Maximum	60, 1560	60, 1460
Median [Q1, Q3]	350 [260, 690]	380 [250, 610]

Traditional culture techniques (details of the steps are available in Appendix J) were initially used to quantify mutans streptococci from unstimulated saliva samples in Sweep 1. *S. mutans* were not isolated for over 60% of the children. In those children who harboured mutans streptococci, the numbers were very low and not with published norms. Hence, absolute numbers of *S. mutans* were quantified from saliva by TaqMan® QPCR.

The distributions of data for *S.mutans* counts in Sweep 1 and 2, salivary cortisol levels, salivary IgA antibodies specific to *S. mutans* and antimicrobial peptides in Sweep 1 were investigated using histograms. The distribution of salivary *S. mutans* counts in Sweep 1 and 2 were log normally distributed (Figures 5-2).

The concentrations of AMPs, LL37 and calprotectin in saliva in Sweep 1 were assessed. While calprotectin was log normally distributed, LL37 was not detected in all of the children and so the data were negatively skewed (Figure 5-3).

Salivary IgA antibodies specific to *S. mutans* were detected in the saliva of all children assessed and the titres were normally distributed (Figure 5-4a). Salivary cortisol levels were detected in all children assessed which was log normally distributed (Figure 5-4b).

Further analyses of salivary immunological factors and cortisol is ongoing and formed a separate piece of work.

In summary, it was feasible to collect saliva samples from children in the nursery/school setting of sufficient quantity and adequate quality on which key microbiological and immunological markers of interest were measured.

a)

b)

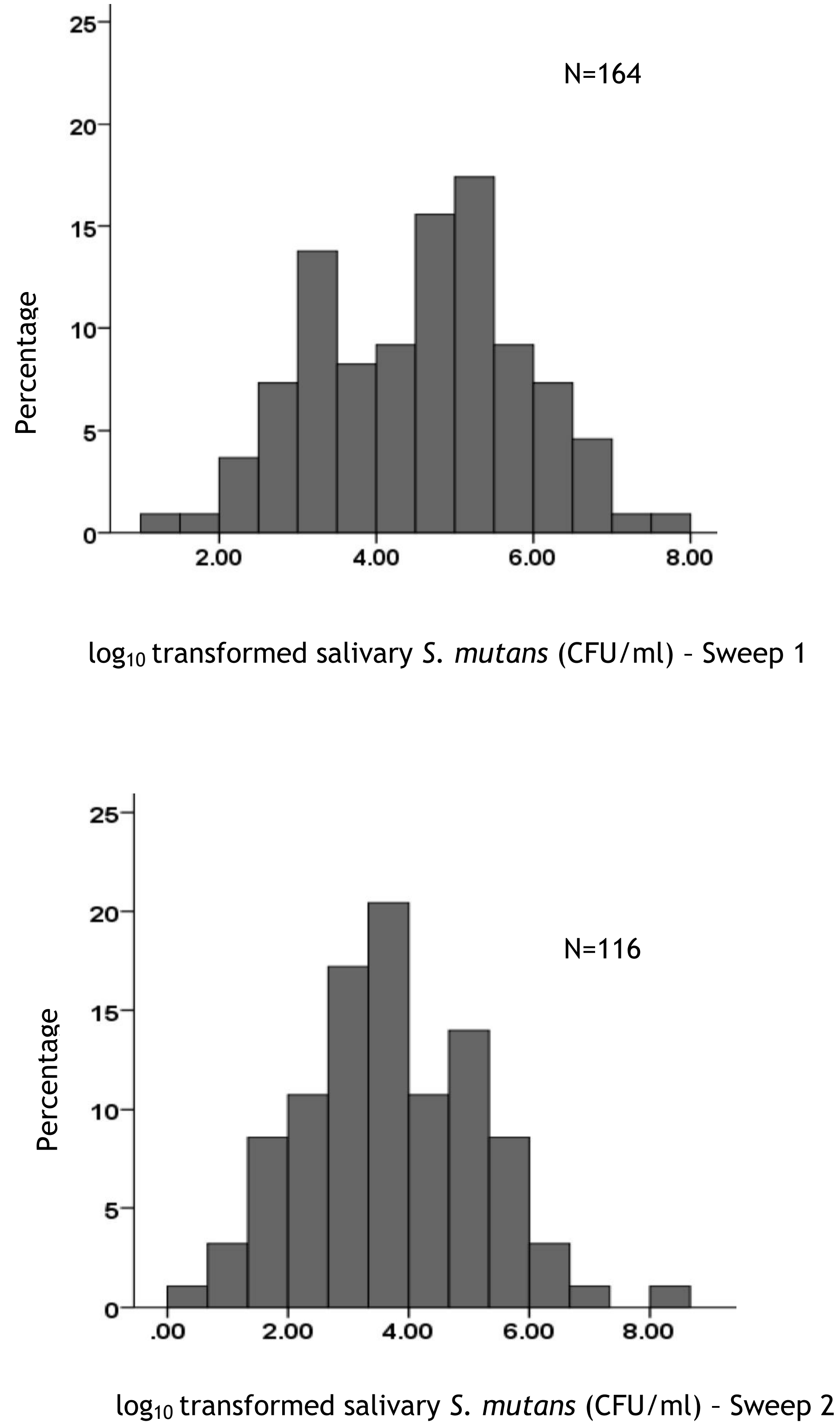


Figure 5-2: Histograms of percentage distribution of $\log_{10}$ transformed salivary *S. mutans* absolute counts in a) four to five-year-olds in Sweep 1 and b) six to seven-year-olds in Sweep 2

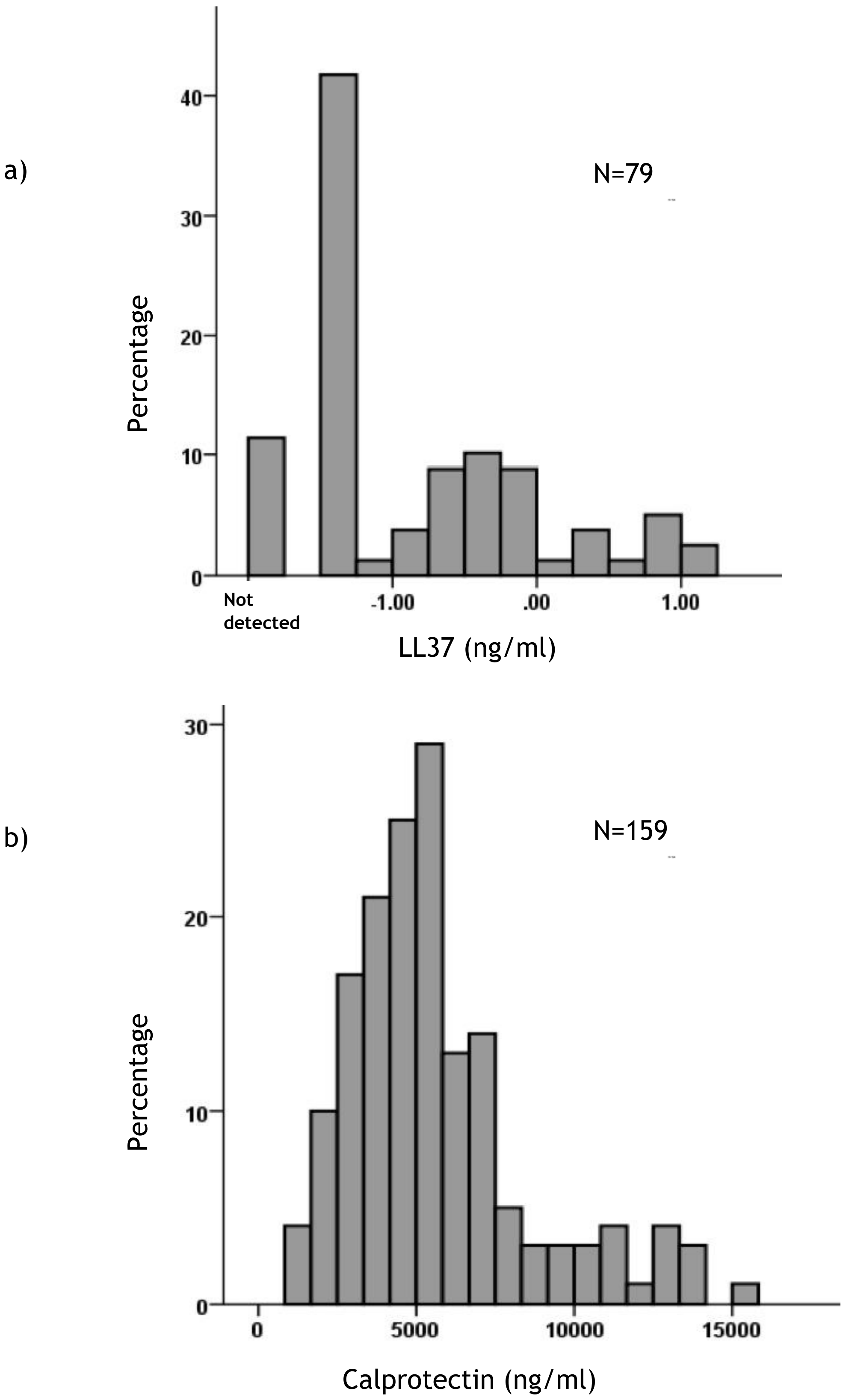


Figure 5-3: Histograms of percentage distribution of log_{10} transformed concentrations of antimicrobial peptides (ng/ml) from the saliva of children in Sweep 1 a) LL37 b) calprotectin

a)

b)

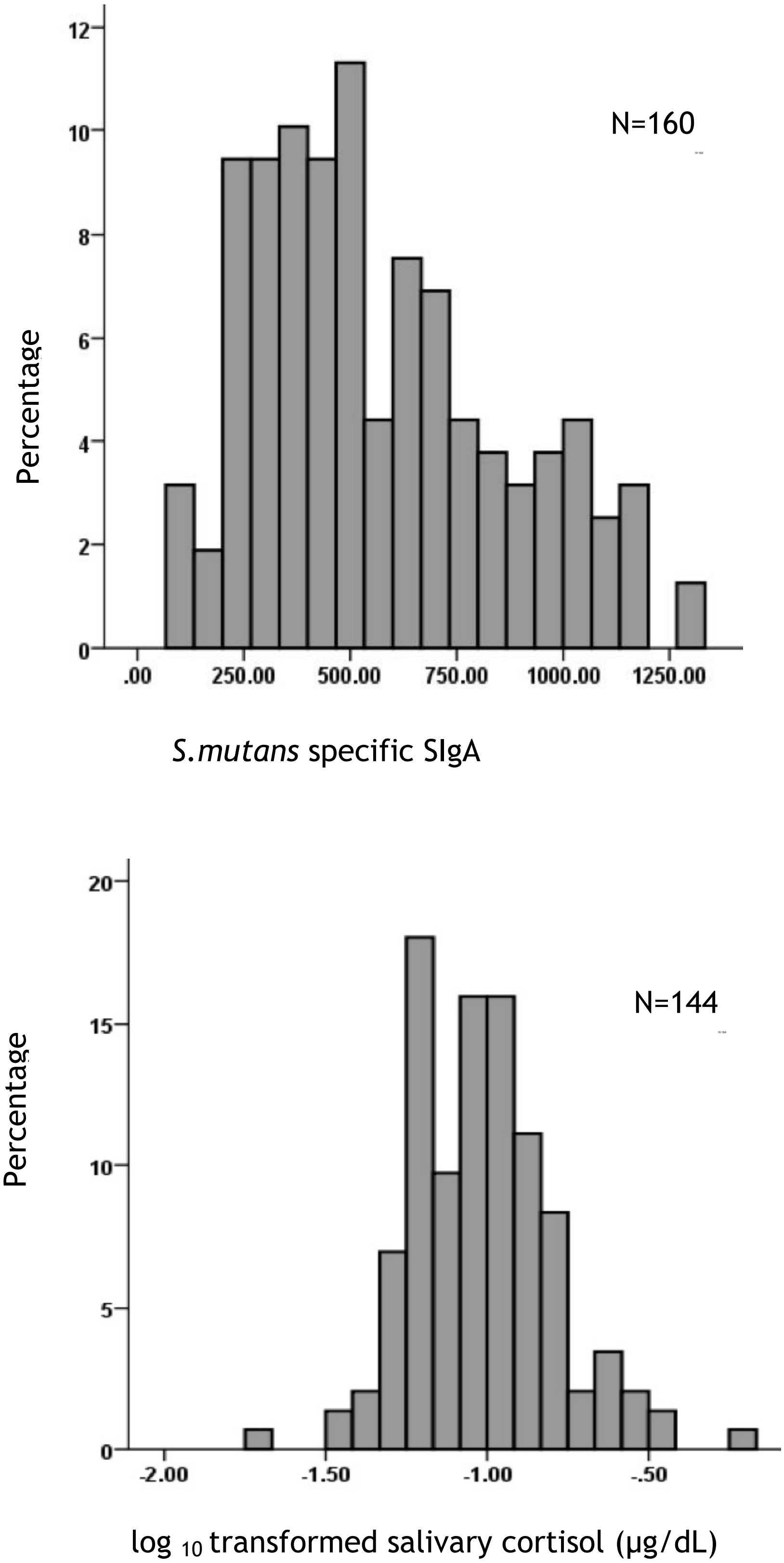


Figure 5-4:Histograms of percentage distribution of a) titres of sIgA antibodies specific for *S. mutans* in Sweep 1 b) $\log_{10}$ transformed salivary cortisol concentrations (µg/dL) in Sweep 1

5.4.1.3 Anthropometric data

The methods used to collect anthropometric data have previously been used in the Child Health Surveillance Programme (CHSP) for both preschool and primary school children in Scotland. Heights and weights were successfully collected from all children for whom data collection was attempted in both Sweeps (N Sweep 1= 190, N Sweep 2= 175). It took approximately one minute per child to have their heights and weights measured. Standard Operation Procedures were produced for collecting heights and weights and are available in Appendix K.

Three participants were excluded from analyses due to large variation in standard deviation scores (>4 and < -4) between Sweep 1 and Sweep 2. This study only intended to collect heights and weights to test feasibility of collecting this data at nursery/school setting, along with other clinical measures potentially as a part of the Child Health Surveillance Programme (CHSP) in the future.

5.4.1.3.1 Body Mass Index (BMI)

The BMI z scores, relative to 1990 references (Cole et al., 1995) were approximately normally distributed (Figure 5-6). The mean [SD] BMI z scores at Sweep 1 and 2 were 0.41 [1.01] and 0.37 [1.06] respectively. The BMI distribution of children classified as overweight and obesity combined was 25% in Sweep 1 and 22% in Sweep 2 (Figure 5-5). This is comparable with the most recent figures (21.9%) for P1 children in Scotland recorded on the CHSP School system (ISD Scotland, 2013a).

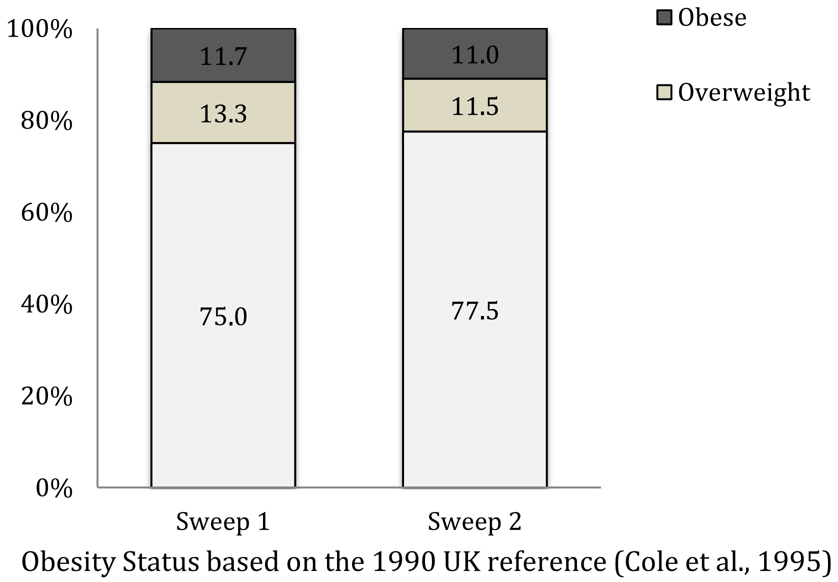


Figure 5-5: Obesity status of children in Sweep 1 and Sweep 2

In summary, it was feasible to measure heights and weights along with conducting a clinical examination in a nursery/school setting on preschool and primary-1 age group.

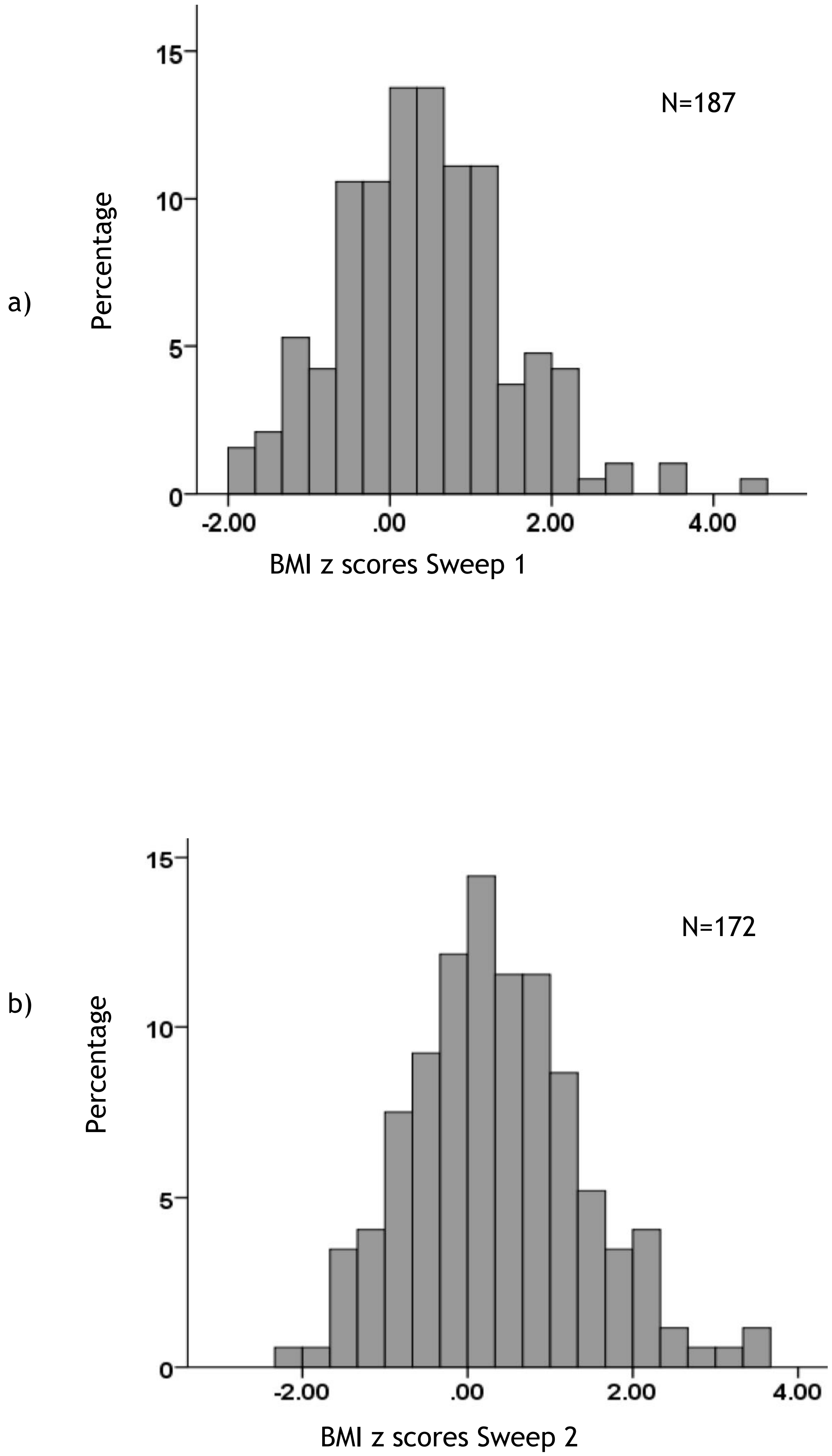


Figure 5-6: Histograms showing the distribution of BMI z scores a) four to five-year-olds in Sweep 1 and b) six to seven-year-olds in Sweep 2

5.5 Feasibility of collecting questionnaire data

As suggested in Figure 4-1, the biological, behavioural, psychosocial and socioeconomic factors of interest were to be collected in Sweep 1 and the main outcome measure was caries experience at Sweep 2. Hence, a decision was made to prioritize the collection of clinical data at the expense of questionnaires in Sweep 2. Strategies to increase response rates were primarily targeted at obtaining clinical data in Sweep 2.

The 'About your study child' and 'About your family' questionnaires were completed to a high standard with necessary detail. Of all the completed questionnaires that were received in Sweep 1, 92% (n=164) of the questionnaires were 100% complete. Unsurprisingly, questions on gross household income and receipt of benefits were sensitive and these were the questions that were most commonly left unanswered (n=14).

Of the 171 completed FFQs received, eight Food Frequency Questionnaires (FFQs) with more than 10 blank lines were classed as incomplete and not analysed in Sweep 1. Thus, completed FFQs were available for 95.5% of those who returned. Seven FFQs classed as over/underreported FFQs were further excluded from analyses.

5.6 Time and cost implications

Time spent on recruitment involved selecting eligible participants, emailing/posting letters and meeting with head teachers, obtaining lists detailing the names, dates of birth and home postcodes of all preschool children; and sending out invitation letters, participant information leaflets and consent forms to over 600 families and collecting consent forms from parents (via nursery). Those parents who consented for their children were sent three sets of questionnaires that were manually labelled for anonymity. This was an onerous process as the aforementioned tasks were carried out in multiple sweeps over a short period of three months chiefly by the primary researcher with occasional help from two other personnel.

Time spent on clinical data collection involved the time of the primary researcher and a second dentist (NZ). Assembling equipment for data collection at the nursery/school detailed in ***Section 4.5.3.1*** lasted eight to ten minutes. A further three to five minutes were involved in collecting caries clinical data per child. In this way, a full working day (4 hours) was required to examine and collect data in spite of the low recruitment rates at each nursery. Face-to-face questionnaire completion for the only one parent in Sweep 1 who requested this mode lasted 65 minutes mainly due to language barriers. Follow-up phone calls as reminders for questionnaire completion and return took approximately seven minutes per parent. Although most parents were only available to talk after 1800 hours, this proved to be a useful method. Thirty-one and thirty-four questionnaires were obtained as a result of these follow-up calls in Sweep 1 and Sweep 2 respectively. These telephone calls were made from a standard staff desk phone at GDHS incurring no/negligible costs.

Table 5-5 outlines the costs involved for the study in Sweep 1. Notably, high costs incurred in Sweep 1 were due to travelling to varying locations of the nurseries around Glasgow for pack delivery, consent form and questionnaire collection, data collection and transporting samples to the laboratory within 2-3 hours of collection. The high cost incurred in Sweep 1 due to transportation was taken into consideration and where possible all contacts in Sweep 2 were made over phone rather than personal visits. Packs were posted to the schools and completed questionnaires requested for return to the GDHS using a postage paid envelope enclosed within every questionnaire pack. The cost of transportation was thus reduced by a third in Sweep 2 (£358.45).

Table 5-5: Costs involved in the study for Sweep 1

Item	Unit	Sub-Units	Numbers required	No. of Sub-units required	No. of units Ordered	Price per Unit	Total Price in Sweep 1
CLINICAL							
Oral Swab	1	50	219	200	4	£42.50	£170.00
Swab storage tube	1	50	219	200	4	£30.00	£120.00
4" Swab storage tube boxes	1	0			2	£9.00	£18.00
Plaque eppendorfs			219		8	£3.50	£28.00
Bar coded labels	1	200	219	876	4.38	£18.00	£78.84
Mouth Mirror	1	12	24	24	2	£5.00	£10.00
CPITN Probe	1	40	40	40	1	£95.00	£95.00
Examination light					0		Borrowed
Insulated Storage bag for Saliva swabs	1				0		
ANTHROPOMETRICS							
Weighing Scale (Seca 877)	1				1	£185.00	£185.00
Stadiometer (Tanita H 001/portable)	1				1	£60.00	£60.00
DIETARY ASSESSMENT							
FFQ supply	1	1	219	219	219	£1.00	£219.00
Nutrient Analysis	1	1	219	219	219	£3.00	£657.00
Additional Analysis (NMES)	1	1	219	219	400	£1.00	£400.00
QUESTIONNAIRES							
About your study child			219		219	£1.50	£328.50
About your family			219		219	£1.50	£328.50
MISCELLANEOUS							
Teddy Bear- Prop	1				0		Borrowed
Stickers for children	1	15	219	200	14		£28.00
Carry Bag	1				0		Borrowed
Sunglasses					0		Borrowed
Transportation Expenses							£1,492.00
Medical waste bags					0		Borrowed
Latex gloves	1	100	400	400	6	£7.00	£42.00
Nursery incentives	13				13	£125.00	£1,625.00
Parents Prize Draw	1				1	£175.00	£175.00
TOTAL							**£ 6059.84**

Chapter 6– Results: Statistical modelling

This chapter addresses objectives 3 (i) and (ii)

6.1 Characteristics of the study sample

Table 6-1 to 6-9 presents the characteristics of the study sample. The tables 6-1 to 6-7 include missing data, to demonstrate the completeness of each question in the questionnaires. For reasons explained previously, the response to questionnaires at Sweep 2 was relatively low (n=100) compared to Sweep 1 (n=179). Hence, direct comparisons are not made in the following sub-sections except when sufficient data are available. However, the frequency distribution of characteristics for all children who had a complete questionnaire returned in Sweep 2 has been tabulated in Table 6-1 to 6-9 for reference.

The majority of the questionnaires in Sweep 1 were completed by the mother (91%) and a very small proportion by the father (seven percent) or grandparent (two percent).

6.1.1 Child characteristics

Table 6-1 shows the ‘child characteristics’ at both Sweeps. The age of the participating children ranged from 3.8 to 5.6 years in Sweep 1 and 4.9 to 6.5 years in Sweep 2. There were slightly more boys (53% - Sweep 1, 54%- Sweep 2) than girls (47% in Sweep 1 and 46% in Sweep 2) in both Sweeps. Forty five percent of the children were first born. The number of other children in the family of the study child varied from one to five or more, with 2 being the most common (44%). The majority of the children were Scottish-White (75%). Other ethnic groups in the study, although in very small numbers (less than seven percent each), included Pakistanis, Chinese, Black Africans, Indians, those of mixed backgrounds, Asians other than Pakistani, Indian and Chinese, and other British.

Table 6-1: Characteristics of Study Sample- Child Characteristics

	Sweep 1 % [n]		Sweep 2 % [n]	
Total children consented	**219**			
	Responders		Responders	
	Including missing	Excluding missing	Including missing	Excluding missing
Gender				
Male	45.7 [100]	52.6	42.9 [94]	54.0
Female	41.1 [90]	47.4	36.5 [80]	46.0
Missing	13.2 [29]		20.6 [45]	
Age of child (years)				
Mean [SD]		4.8 [0.4]		5.7 [0.3]
Minimum, Maximum		3.8, 5.6		4.9, 6.5
Median [Q_1, Q_3]		4.7 [4.5, 5.1]		5.7 [5.4, 6.0]
Ethnicity				
White Scottish	58.9 [129]	75.0	34.7 [76]	75.2
Other White British	1.8 [4]	2.3	1.8 [4]	4.0
White: Any other background	0.5 [1]	0.6	0.5 [1]	1.0
Asian/Asian Scottish or British: Pakistani	5.0 [11]	6.4	1.4 [3]	3.0
Asian/Asian Scottish or British: Indian	0.9 [2]	1.2	0.9 [2]	2.0
Asian/Asian Scottish or British: Chinese	2.3 [5]	2.9	0.9 [2]	2.0
Asian/Asian Scottish or British: Others	3.2 [7]	4.1	0.9 [2]	2.0
Mixed background	3.7 [8]	4.7	3.7 [8]	7.9
Black, Black Scottish or British: African	2.3 [5]	2.9	1.4 [3]	3.0
Missing	21.5 [47]		53.9 [118]	
Number of children in family				
1	17.4 [38]	22.4	14.2 [31]	21.8
2	33.8 [74]	43.5	29.2 [64]	45.1
3	20.5 [45]	26.5	16.4 [36]	25.4
4	3.2 [7]	4.0	2.7 [6]	4.2
5 or more	2.7 [6]	3.6	2.2 [5]	3.5
Missing	22.4 [49]		35.2 [77]	
Position of child in family				
First born	34.2 [75]	44.7	28.8 [63]	44.4
Second born	27.9 [61]	36.3	23.7 [52]	36.6
Third born	11.9 [26]	15.5	10.5 [23]	16.2
Fourth or later	2.7 [6]	3.6	1.8 [4]	2.8
Missing	23.3 [51]		35.2 [77]	

6.1.2 Main-carer characteristics

Characteristics of the main carer are presented in **Error! Reference source not found.**. Half of the participating main-carers (mothers) of the children were aged between 25 and 35 years (49%) and only 11% were young mothers who were below 25 years of age. The large majority (70%) of children participating in the study lived in a household with two parent figures and nearly a third (29%) lived in a household where the main carer smoked.

Table 6-2: Characteristics of Study Sample- Main-Carer Characteristics

	Sweep 1 % [n]		**Sweep 2 % [n]**	
	Responders		Responders	
	Including missing	Excluding missing	Including missing	Excluding missing
Relationship with Study Child				
Mother	72.6 [159]	91.4	41.6 [91]	89.2
Father	5.5 [12]	6.9	4.6 [10]	9.8
Grandparent	1.4 [3]	1.7	0.5 [1]	1.0
Missing	20.5 [45]		53.4 [117]	
Age of main carer				
<25 years	8.7 [19]	11.0	25.1 [55]	5.0
25 -35 years	38.8 [85]	49.1	18.3 [40]	40.0
36 years and over	31.5 [69]	39.9	2.3 [5]	55.0
Missing	21.0 [46]		54.3 [119]	
Main Carer marital status				
Married	37.4 [82]	47.4	25.6 [56]	54.9
Cohabiting	17.8 [39]	22.5	6.8 [15]	14.7
Single	20.5 [45]	26.0	11.9 [26]	25.5
Widowed/separated/Divorced	3.2 [7]	4.0	2.3 [5]	4.9
Missing	21.0 [46]		53.4 [117]	
Main-carer smoking status				
Current Smoker	23.3 [51]	29.3	10.5 [23]	22.5
Never smoked/Smoked in past	56.2 [123]	70.7	36.1 [79]	77.5
Missing	20.5 [45]		53.4 [117]	

6.1.3 Socioeconomic circumstances

Socioeconomic circumstances of the participants were measured both at an area-level and individual-level using a variety of measures as discussed in ***Section 4.5.1.2.3*** and the distribution of the participants according to these measures tabulated in Table 6-3.

6.1.3.1 Scottish Index of Multiple deprivation

The distribution of the participants according to SIMD when classified using local SIMD 2009 was relatively evenly spread across Q2, Q3 and Q4 quintiles at both Sweeps. Children from Q1 (the most deprived quintile) were over-represented accounting for 38% of the sample in Sweep 1 and 39% in Sweep 2. Only seven (five in Sweep 2) children belonged to Q5 (least deprived quintile).

Similar patterns were seen when the participants were classified according to the national SIMD in Sweep 1 and Sweep 2 with the majority falling into Q1.

6.1.3.2 Gross annual household income

There was almost an equal distribution of parents belonging to the different income groups, with approximately 30% each in the bottom two income categories (< £10,000 and £10,000 to £19,999). Thirty-four percent (34%) of the parents reported earning between £20,000 – £49,999 and seven percent above £50,000 in Sweep 1. A very similar distribution was evident in Sweep 2.

6.1.3.3 Receipt of benefits

While 41% of the parents did not receive any form of Governmental benefits (except child benefit), 28% reported receiving all of their household income through benefits schemes.

6.1.3.4 Current or last occupation of the main-carer

The current or last occupation of the main-carer was measured in Sweep 2 only and was categorised using the Standard Occupation classification (SOC) (2010) as described in ***Section 4.6.2.3***. The numbers under each category were relatively small due to large number of questionnaires (53%) that were not returned in Sweep 2. However, 93% of the parents/carers who returned the questionnaires reported having not changed their occupation over the last year and hence the following results may be representative of their occupational status in Sweep 1. Overall, 5% of the main carers never worked or were permanently unemployed due to a disability. The largest number of main-carers belonged to the ‘Professionals’ group (8%).

6.1.3.5 Highest educational qualification of the main-carer

Over two-thirds (67%) of the parents/carers had at least one year of full time education after leaving secondary school. While 23% of the parents had reported to have a First/higher degree/professional qualification, 15% had no educational qualification other than a pre-school leaving certificate. A third of the parents (33%) reported having an intermediate degree (Higher grade, standard grade, GSVQ advanced, HNC, HND, SVQ level 4 or 5 or equivalent).

6.1.3.6 Housing tenure

There were almost an equal proportion of parents living in a rented house from Local Authority/Council/Housing Association, and living in an owned/being mortgaged house. A small proportion (15%) stated living in a privately rented apartment/house.

Table 6-3: Characteristics of Study Sample- Socio-economic circumstances

	Sweep 1 % [n]		Sweep 2 % [n]	
	Responders		Responders	
	Including missing	Excluding missing	Including missing	Excluding missing
Local SIMD[8] Quintile				
Q1 (most deprived)	37.4 [82]	38.0	30.6 [67]	39.2
Q2	19.6 [43]	19.9	16.0 [35]	20.5
Q3	18.7 [41]	19.0	15.1 [33]	19.3
Q4	19.6 [43]	19.9	14.2 [31]	18.1
Q5 (least deprived)	3.2 [7]	3.2	2.3 [5]	2.9
Missing	1.4 [3]		21.9 [48]	
National SIMD[9] Quintile				
Q1 (most deprived)	56.2 [123]	56.7	45.7 [100]	58.1
Q2	11.4 [25]	11.5	9.1 [20]	11.6
Q3	18.7 [41]	18.9	14.2 [31]	18.1
Q4	10.0 [22]	10.1	7.7 [17]	9.9
Q5 (least deprived)	2.7 [6]	2.8	1.8 [4]	2.3
Missing	0.9 [2]		21.5 [47]	
Gross annual household Income				
< £10,000	21.5 [47]	29.2	10.0 [22]	24.2
£10,000 – £19,999	21.9 [48]	29.8	10.5 [23]	25.3
£20,000 – £49,999	25.1 [55]	34.2	16.0 [35]	38.5
£50,000 & more	5.0 [11]	6.8	5.0 [11]	12.1
Missing	26.5 [58]		53.0 [116]	
Percentage of income from benefits				
None	31.1 [68]	41.4	21.0 [46]	47.4
About a quarter	12.3 [27]	16.5	5.5 [12]	12.4
About a half	7.8 [17]	10.5	5.5 [12]	12.4
About three quarters	2.7 [6]	3.6	0.9 [2]	2.1
All	21.0 [46]	28.0	11.4 [25]	25.8
Missing	25.2 [55]		55.7 [122]	
Current or Last occupation of Main-Carer				
Never worked or permanently unemployed due to disability			4.6 [10]	9.8
Manager & Senior officials			2.7 [6]	5.9
Professionals			7.8 [17]	16.7
Associate professional/Technical	***Not measured in Sweep 1***		5.0 [11]	10.8
Admin & Secretarial			6.8 [15]	14.7
Skilled trades			1.4 [3]	2.9
Caring, leisure & services			6.8 [15]	14.7
Sales & customer services			5.0 [11]	10.8
Process, plant machinery			1.4 [3]	2.9
Elementary occupation			5.0 [11]	10.8
Missing			53.4 [117]	

[8] Scottish Index of Multiple Deprivation 2009 at Local Health Board level (NHS Greater Glasgow & Clyde)

[9] Scottish Index of Multiple Deprivation 2009 at National level

Table 6-3 **continued**

	Sweep 1 % [n]		Sweep 2 % [n]	
	Responders		Responders	
	Including missing	Excluding missing	Including missing	Excluding missing
Main-Carer's years of Full-time Education				
Not finished secondary school	7.3 [16]	9.6	1.4 [3]	3.1
Finished, but no further education	17.8 [39]	23.5	12.8 [28]	28.9
One to two Years	18.7 [41]	24.7	7.3 [16]	16.5
Three to four Years	16.0 [35]	21.1	11.9 [26]	26.8
Five or more	16.0 [35]	21.1	11.0 [24]	24.7
Missing	24.2 [53]		55.7 [122]	
Main-Carer's highest level of Education				
Level 0[10]	11.9 [26]	15.1	6.8 [15]	14.9
Level 1[11]	22.4 [49]	28.5	7.8 [17]	16.8
Level 2[12]	26.0 [57]	33.1	17.4 [38]	37.6
Level 3[13]	18.3 [40]	23.3	14.2 [31]	30.7
Missing	21.5 [47]		53.9 [118]	
Housing Tenure				
Rented: local Authority/Council	8.2 [18]	10.4	5.9 [13]	12.9
Rented: Housing Association	24.2 [53]	30.6	11.9 [26]	25.7
Rented: Privately unfurnished	8.7 [19]	11.0	4.1 [9]	8.9
Rented: Privately furnished	2.7 [6]	3.5	1.4 [3]	3.0
Being bought/mortgaged	33.3 [73]	42.2	21.5 [47]	46.5
Owned with no mortgage	1.8 [4]	2.3	0.9 [2]	2.0
Missing	21.0 [46]		53.9 [118]	

6.1.4 Oral hygiene practices

Table 6-4 illustrates the distribution of the participants according to oral hygiene practices. Seventy eight percent (78%) of the parents reported commencing their child's toothbrushing before 12 months of age or when the first tooth erupted, and the majority (76%) brushed twice or more everyday (Table 6-4). Over half of the children were reported to be always brushing before bedtime (53%). All parents reported using a toothpaste containing at least 1000 ppm fluoride except one. The vast majority also reported that they supervised/helped their child while toothbrushing (85%) and used small

[10] Level 0 - no qualification or pre-school leaving qualification

[11] Level 1 - O grade, standard grade, GCSE or equivalent

[12] Level 2 - Higher grade, A level, GSVQ advanced HNC, HND, SVQ Levels 4 or 5 or equivalent

[13] Level 3 - first degree, higher degree or professional qualification

amounts of toothpaste (70%), such as a ‘pea-size’ or ‘smear’. A majority of children (62%) reportedly spit after toothbrushing while 25% rinsed out.

Table 6-4: Characteristics of Study Sample- Oral hygiene Practices

	Sweep 1 % [n]		Sweep 2 % [n]	
	Responders		Responders	
	Including missing	Excluding missing	Including missing	Excluding missing
Age when toothbrushing started				
Under 12 months/First tooth	61.2 [134]	78.4	***Not measured in***	
After 12 months	16.9 [37]	21.6	***Sweep 2***	
Missing	21.9 [48]			
Daily toothbrushing frequency				
Not Every day	2.7 [6]	3.5	0.0 [0]	0.0
Once a day	16.0 [35]	20.2	6.4 [14]	14.1
Twice a day	50.7 [111]	64.2	35.2 [77]	77.8
More than twice a day	9.6 [21]	12.1	3.7 [8]	8.1
Missing	21 [46]		54.8 [120]	
Brush before bedtime				
Always	42.5 [93]	53.1	29.7 [65]	63.7
Sometimes	28.3 [62]	35.4	11.4 [25]	24.5
Occasionally	5.5 [12]	6.9	4.1 [9]	8.8
Hardly ever	2.7 [6]	3.4	0.9 [2]	2.0
Never	0.9 [2]	1.1	0.5 [1]	1.0
Missing	20.1 [44]		53.4 [117]	
Fluoride concentration in toothpaste				
Up to 550 or less than 1000	0.5 [1]	0.6	***Not measured in***	
1000/1100 ppm	53.4 [117]	67.2	***Sweep 2***	
> 1100 ppm	25.6 [56]	32.2		
Missing	20.5 [45]			
Adult supervised tooth-brushing				
Yes	67.6 [148]	84.6	33.8 [74]	74.7
No	12.3 [27]	15.4	11.4 [25]	25.3
Missing	20.1 [44]		54.8 [120]	
Amount of toothpaste used				
Smear	2.3 [5]	2.9	0.9 [2]	2.0
Pea-size	53.9 [118]	67.4	31.5 [69]	67.6
Half a brush	21.0 [46]	26.3	11.9 [26]	25.5
Full brush	2.7 [6]	3.4	2.3 [5]	4.9
Missing	20.1 [44]		53.4 [117]	
Method for toothpaste removal				
Swallow	11.0 [24]	13.7	5.0 [11]	11.1
Spit	49.3 [108]	61.7	27.4 [60]	60.6
Rinse out	19.6 [43]	24.6	12.8 [28]	28.3
Missing	20.1 [44]		54.8 [120]	

6.1.5 Use of Dental services

Table 6-5 illustrates the distribution of participants according to use of dental services. While parents/carers of 38% of the children reported having taken their child for their first dental visit before their first birthday, 13% reported never taking their child for a dental visit. The common reasons for the first visit were anticipatory such as 'just went for getting used to' or 'check up'. More than half of the parents (68%) reported taking their child for a routine check-up every six months while 15% never did.

Table 6-5: Characteristics of Study Sample- Use of dental services

	Sweep 1 % [n]		Sweep 2 % [n]	
	Responders		Responders	
	Including missing	Excluding missing	Including missing	Excluding missing
Age at first dental visit				
Never visited dentist	10.0 [22]	12.7		
Less than or at 6 months	9.6 [21]	12.1	***Not measured in***	
7- 12 months	20.5 [45]	26.0	***Sweep 2***	
13 months and over	38.8 [85]	49.1		
Missing	21.0 [46]			
Reason for first dental visit				
Trouble with teeth	3.2 [7]	4.0		
Note from nursery dentist	1.8 [4]	2.3		
Check up	22.4 [49]	28.0	***Not measured in***	
Just went for getting used to	35.2 [77]	44.0	***Sweep 2***	
Organized *Childsmile* visit	6.8 [15]	8.6		
Never been	10.0 [22]	12.6		
Trauma	0.5 [1]	0.6		
Missing	20.1 [44]			
Frequency of routine check				
<= 6 months	53.8 [118]	67.8	35.6 [78]	75.7
Between 6 and 12 months	11 [24]	13.8	6.8 [15]	14.6
>12 months	2.7 [6]	3.4	2.7 [6]	5.8
Never	11.9 [26]	14.9	1.8 [4]	3.9
Missing	20.6 [45]		53.0 [116]	

6.1.6 Early feeding habits

Forty two percent (42%) of the parents/carers reported initiating breastfeeding of which 22% stated exclusively breastfeeding their child as infants and others (20%) combined/continued with a bottle. More than half (58%) of the parents/carers reported exclusively bottle feeding their child as an infant (Table 6-6).

About half (53%) of the parents reported commencing solids at four to six months of age. More than a quarter of the children (26%) were reported to have stopped using a baby bottle with a teat before their first birthday. While 37% were reported as not taking a bottle to bed with a drink (other than water) at two years of age, 26% were taking it every night. Furthermore, 13% continued this practice at four years. Only a third (30%) of the parents restricted the amount of sugars their child had on a daily basis.

Table 6-6: Characteristics of Study Sample- Early feeding habits

	Sweep 1 % [n]		Sweep 2 % [n]	
	Responders		Responders	
	Including missing	Excluding missing	Including missing	Excluding missing
Breast or bottle feeding				
Breast fed only	17.4 [38]	21.7	***Not measured in***	
Bottle fed only	46.1 [101]	57.7	***Sweep 2***	
Breast and bottle fed	16.4 [36]	20.6		
Missing	20.1 [44]			
Age bottle feeding completely stopped				
Never used a baby bottle	5.5 [12]	6.9	***Not measured in***	
Under 12 months	15.5 [34]	19.4	***Sweep 2***	
Between 1 year and 2 years	36.1 [79]	45.1		
Between 2 and 3 years	12.3 [27]	15.4		
Between 3 and 4 years	6.4 [14]	8.0		
Still using baby bottle	4.1 [9]	5.1		
Missing	20.1 [44]			
Night time bottle feeding (other than water) up to 2 years				
Every day	20.5 [45]	25.9		
Most days	8.7 [19]	10.9		
Some days	12.3 [27]	15.5	***Not measured in***	
Hardly ever	8.2 [18]	10.3	***Sweep 2***	
Never	29.7 [65]	37.4		
Missing	20.5 [45]			
Age of commencement of solids				
2-3 months	7.3 [16]	9.2	***Not measured in***	
4-6months	52.5 [115]	66.1	***Sweep 2***	
7-12 months	16 [35]	20.1		
Over 12 months	3.7 [8]	4.6		
Missing	20.5 [45]			
Night time bottle feeding (other than water) up to 4 years				
Every day	10.0 [22]	12.7	4.6 [10]	9.9
Most days	3.7 [8]	4.6	2.3 [5]	5.0
Some days	10.0 [22]	12.7	4.6 [10]	9.9
Hardly ever	11.9 [26]	15.0	5.0 [11]	10.9
Never	43.4 [95]	54.9	29.7 [65]	64.4
Missing	21.0 [46]		46.1 [101]	
Frequency sugar restricted				
Every day	24.2 [53]	30.3	11.0 [24]	24.5
Most days	32.9 [72]	41.1	19.2 [42]	42.9
Some days	16.4 [36]	20.6	14.2 [31]	31.6
Hardly ever	4.6 [10]	5.7	0.5 [1]	1.0
Never	1.8 [4]	2.3	0.0 [0]	0.0
Missing	20.1 [44]		55.3 [121]	

6.1.7 Current Diet

A minimum of 100 complete FFQs are required for robust nutritional analysis under the guidelines drawn up by the Scottish Collaborative Group (The survey of sugar intake among children in Scotland, 2008). Since less than 100 complete FFQs were returned in Sweep 2, the results of the analyses were found to be unreliable and misleading and hence not presented.

Table 6-7 illustrates the mean intakes of various macronutrients, saturated fat, total sugars, Non-Milk Extrinsic Sugars (NMES) and selected food groups in Sweep 1 as discussed previously in ***Section 4.6.2.4***. Sixty three percent (63%) of the children exceeded the recommended NMES intake of no greater than 11% of food energy. Large variations with respect to the daily consumption of total sugars (42.2 to 670.2 g/day) and NMES (18.7 to 471.8 g/day) was seen. The mean intake of NMES was 87.6 g/day, which on average comprised 16.3% of total food energy. There was considerable variation in the intake of macronutrients and daily consumption of selected food-groups, the greatest variation seen with respect to daily consumption of non-diet soft drinks (including acidic fruit juices and fizzy drinks) ranging from 0 to 2400 g/day (equivalent of six cans of drinks).

Table 6-7: Characteristics of Study Sample- Current diet

	Sweep 1
Energy (Kcal/ day)	
Mean [SD]	1941.3 [1098.4]
Minimum, Maximum	756.0, 9167.0
Median [Q1, Q3]	1658.0 [1335.0, 2110.0]
Missing % [n]	21.9 [48]
Carbohydrate (g/day)	
Mean [SD]	274.2 [159.3]
Minimum, Maximum	102.2, 1113.0
Median [Q1, Q3]	227.9 [185.7, 313.4]
Missing % [n]	21.9 [48]
Protein (g/day)	
Mean [SD]	64.3 [33.6]
Minimum, Maximum	25.3, 282.3
Median [Q1, Q3]	57.9 [43.4, 74.2]
Missing % [n]	21.9 [48]
Fat (g/day)	
Mean [SD]	72.3 [45.2]
Minimum, Maximum	26.1, 428.5
Median [Q1, Q3]	61.7 [48.3, 83.3]
Missing % [n]	21.9 [48]
Saturated fat (g/day)	
Mean [SD]	30.3 [17.6]
Minimum, Maximum	10.5, 156.8
Median [Q1, Q3]	25.7 [19.6, 35.3]
Missing % [n]	21.9 [48]
Total sugars (g/day)	
Mean [SD]	155.9 [105.3]
Minimum, Maximum	42.2, 670.2
Median [Q1, Q3]	118.7 [93.4, 175.1]
Missing % [n]	21.9 [48]
Total sugars (% food energy)	
Mean [SD]	29.7 [7.7]
Minimum, Maximum	13.4, 63.1
Median [Q1, Q3]	28.7 [25.1, 32.5]
Missing % [n]	21.9 [48]
NMES intake (g/day)	
Minimum, Maximum	18.7, 471.8
Median [Q1, Q3]	62.6 [46.6, 99.6]
Missing % [n]	21.9 [48]

Table 6-7 **continued**

	Sweep 1
NMES (% food energy)	
Mean [SD]	16.3 [7.0]
Minimum, Maximum	5.3, 54.1
Median [Q1, Q3]	14.7 [12.3, 18.8]
Missing % [n]	21.9 [48]
NMES (% food energy)	
< 11% food energy % [n]	15.1 [33]
>= 11% food energy % [n]	63.0 [138]
Missing % [n]	21.9 [48]
Intrinsic Milk Sugars	
Mean [SD]	68.2 [42.9]
Minimum, Maximum	16.5, 343.0
Median [Q1, Q3]	60.1 [40.1, 78.9]
Missing % [n]	21.9 [48]
Crisps & Savory Snacks (g/day)	
Mean [SD]	19.41 [20.6]
Minimum, Maximum	0.0, 130.0
Median [Q1, Q3]	13.1 [6.4, 24.7]
Missing % [n]	24.7 [54]
Biscuits, Cakes & Pastries (g/day)	
Mean [SD]	41.1 [52.0]
Minimum, Maximum	0.0, 379.29
Median [Q1, Q3]	26.5 [16.6, 45.4]
Missing % [n]	26.0 [57]
Confectionary (g/day)	
Mean [SD]	23.7 [28.7]
Minimum, Maximum	0.0, 287.3
Median [Q1, Q3]	17.1 [8.8, 28.9]
Missing % [n]	23.3 [51]
Non-Diet Soft Drinks (g/day)	
Mean [SD]	217.3 [358.8]
Minimum, Maximum	0.0, 2400.0
Median [Q1, Q3]	103.6 [34.0, 200.0]
Missing % [n]	29.2 [64]

6.1.8 Parental attitudes and beliefs

The majority of the parents/carers appeared to have positive attitudes around oral health, with most reporting to agree with positive traits (Figure

6-1 a & b). Items that showed notable variation in responses included an item that measured dental fatalism belief, ***'Most children eventually develop dental cavities'*** and another item that was intended to measure parental efficacy in caries prevention, ***'If my child uses fluoride toothpaste, it will prevent tooth decay'***. A quarter (23%) of the parents 'agreed/strongly agreed' and almost another quarter (28%) were 'Unsure' with the former statement measuring fatalism. While thirty-one percent (31%) of the parents were 'Unsure' about the latter statement and 13% 'disagreed/Strongly disagreed'.

Two scales 'positive parental attitude scale' and 'parental efficacy scale' were constructed as described in ***Section 4.5.1.1.4*** in Sweep 1. The 'positive parental attitude' scale was constructed using eight items and the 'parental efficacy scale' using three items as shown in Table 6-8. Item analysis for the two scales showed good internal reliability coefficients: Cronbach's alpha of 0·74 and 0·70 for the former and latter scale respectively. The distributions of scores for each scale were investigated using frequency histograms (Figure 6-3 a & b). The percentage distribution of scores for both the scales were negatively skewed due to more parents showing a higher score indicating favourable attitudes and self-efficacy towards oral health.

Table 6-8: Statements used to construct each of the scales and their respective Cronbach's alpha

Parental efficacy scale- Cronbach's alpha of 0·70
-It is worthwhile to give my child sweets/biscuits to behave well -It is not worth it to battle with my child to brush his/her teeth twice a day -It is often too stressful to say no to to my child when they want sweets
Positive parental attitude scale- Cronbach's alpha of 0·74
-My child losing a baby tooth due to tooth decay would be upsetting -It is worthwhile to give my child sweets/biscuits to behave well -Most children eventually develop dental cavities -As a family, we intend controlling how often the child has sugary foods or drinks -It is not worth it to battle with my child to brush his/her teeth twice a day -It is often too stressful to say no to my child when they want sweets -As a parent, it is my responsibility to prevent my child getting tooth decay -It is the responsibility of the dentist to prevent my child getting tooth decay

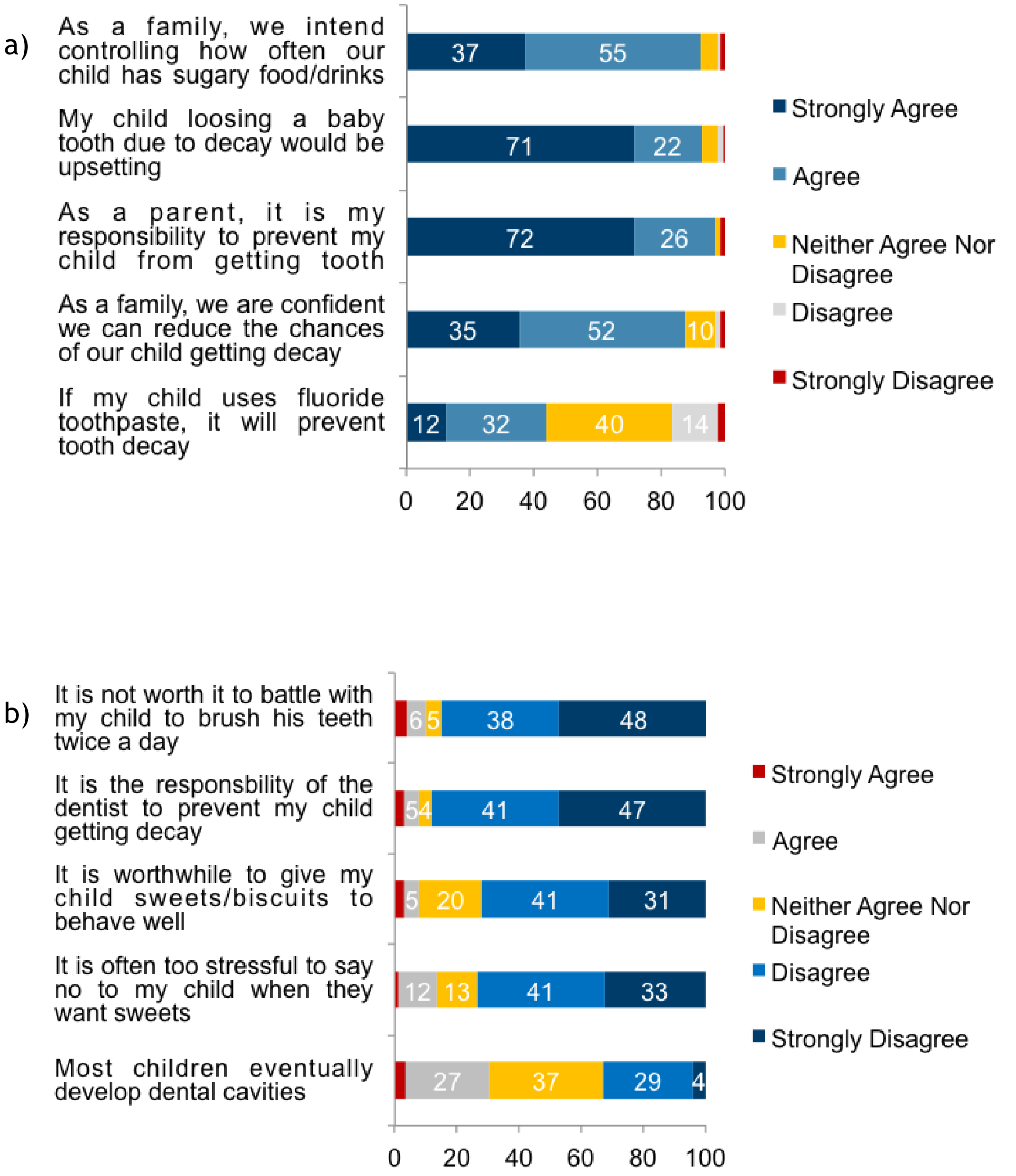


Figure 6-1: Bar chart showing distribution of responses in Sweep 1 for a) positive statements, b) negative statements

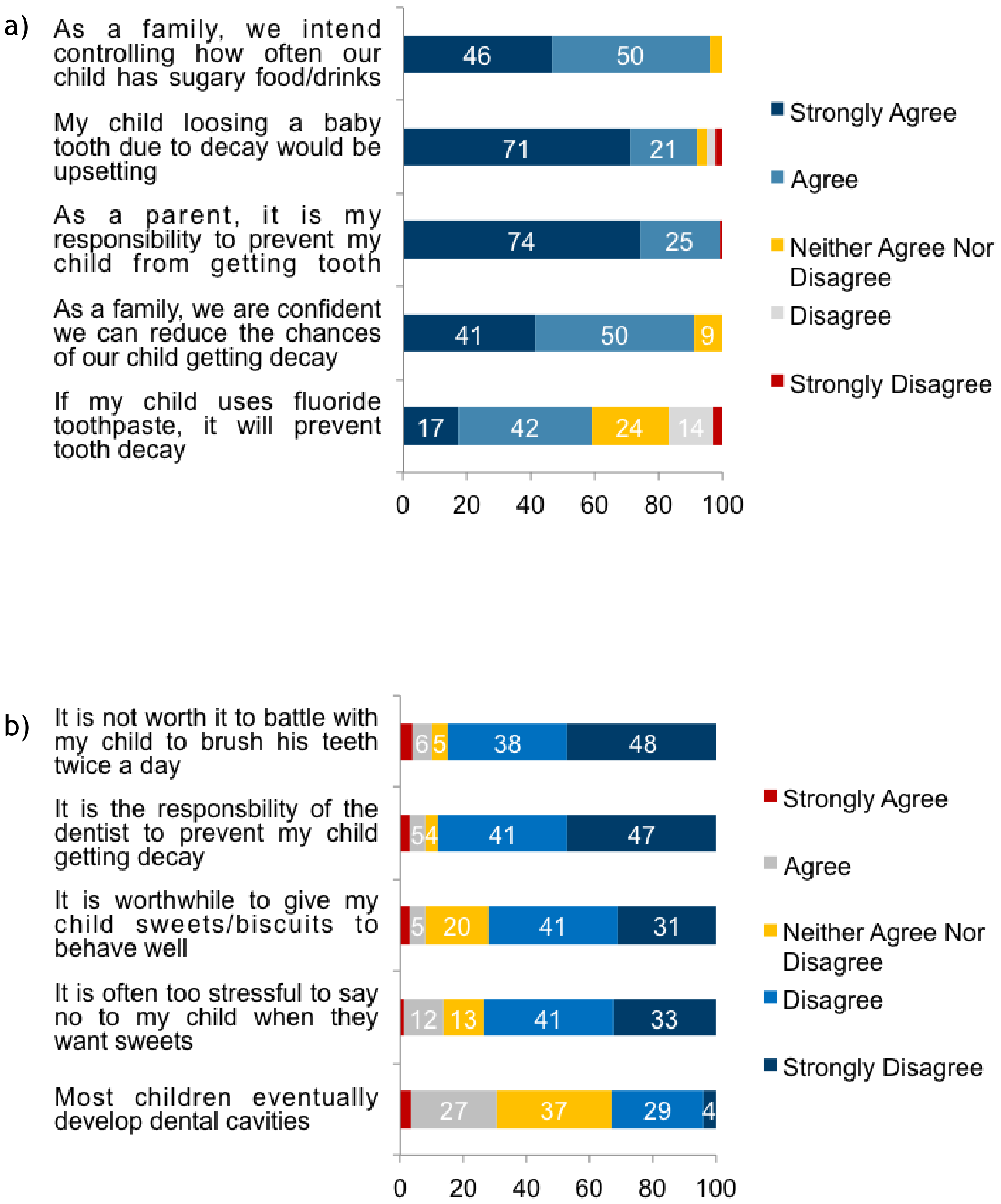
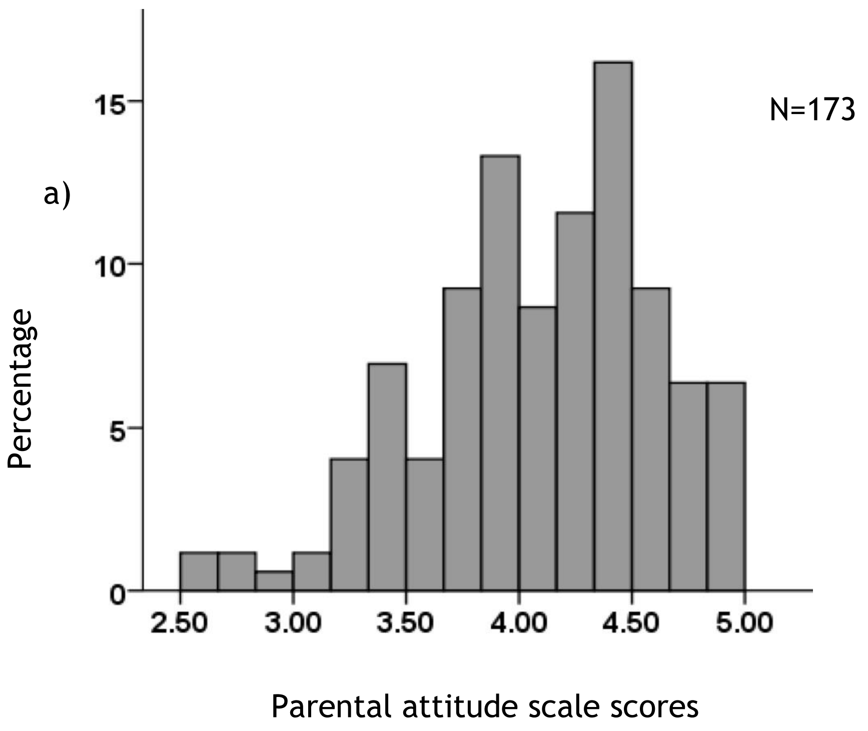


Figure 6-2: Bar chart showing distribution of responses in Sweep 2 for a) positive statements, b) negative statements

a)



b)

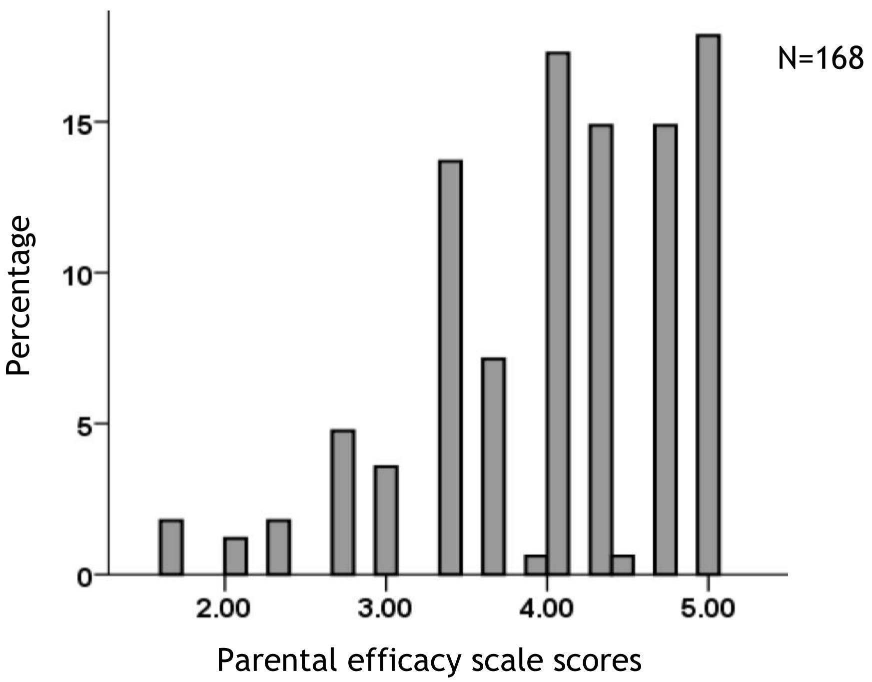


Figure 6-3: Histograms of percentage distribution of a) positive parental attitudes scale in Sweep 1and b) parental efficacy scale in Sweep 1.
A higher score indicates a more favourable attitude towards oral health.

6.1.9 Streptococcus mutans

Absolute counts of *S. mutans* were quantified from saliva using TaqMan® QPCR. The count of *S. mutans* in saliva of children ranged from 2.6 x 10^1 to 4.5 x 10^7 CFU/ml with a geometric mean of 3.5 x 10^4 CFU/ml in Sweep 1 (Table 6-9).

Table 6-9: Characteristics of Study Sample- Microbiology

	Sweep 1	
	Responders	
	Including missing	Excluding missing
N= 219		
***S. mutans* (CFU/ml)**		
Not Detected % [n]	25.1 [55]	34%
Detected % [n]	49.8 [109]	67%
Missing % [n]	25.1 [55]	
*Mean [SD]		3.5 x10^4 [2.0 x10^1]
*Minimum, Maximum		2.6 x10^1, 4.5x10^7
*Median [Q1, Q3]		4.0 x10^4 [3.0x10^3, 3.0x10^5]

*Data for children who had detectable levels of *S.mutans* in Sweep 1. Geometric data generated from back transformations of $\log_{10}$ transformed data are shown.

6.1.10 Summary

In summary, the sample comprised mainly of deprived families. The completeness of the questionnaires were good in both Sweeps. The lower response to questionnaires in Sweep 2 may reflect the importance of using various strategies to increase response rate, particularly when the sample primarily comprises of the more deprived groups. However, there was no differential participation in Sweep 2 based on behavioural and social factors.

It is feasible to look at changes in diet and parental attitudes & beliefs over time, but is outwith the scope of this thesis.

6.2 Obvious caries experience at Sweep 1 and Sweep 2

The proportion of children with obvious caries experience was 35.8% in Sweep 1 and 47.7 % in Sweep 2 (Table 6-10). The mean number of teeth with caries experience changed from 1.3 (AMR) in Sweep 1 to 1.9 ($d_3$mft) in Sweep 2.

The burden of the disease in children affected by caries was high, with the mean number of teeth affected [SD] being 3.7 [2.6] in Sweep 1 and 3.9 [3.0] in Sweep 2 (Table 6-11).

Table 6-10: Distribution and summary statistics of caries experience in baseline year (Sweep 1) and follow-up year (Sweep 2)

Total number of children who consented in the study	N= 219				
	Sweep 1 (AMR)	Sweep 2[14] ($d_3$mft)	Sweep 2[15] ($d_3$mft)	NDIP 2009/10[16]	NDIP 2013/14[17]
Total number of children examined % [n]	86.8 [190]	79.5 [175]	65.8 [144]	19 [2583]	[1188][18]
Mean age [SD]	4.8 [0.4]	5.7 [0.3]	5.7 [0.3]	3.7 [0.3]	5.5 [0.3]
Proportion with obvious caries in primary teeth % [n]	35.8 [68]	47.7 [83]	47.2 [68]	19 [483]	41.2 [490]
Mean AMR/$d_3$mft [SD]	1.3 [2.6]	1.9 [3.0]	1.9 [3.0]	0.4 [1.1]	1.8
Minimum, Maximum	0, 18	0, 18	0, 18	*No data available*	*No data available*
Median [Q_1, Q_3]	0.0 [0, 2]	0.0 [0, 3]	0.0 [0, 3]	*No data available*	*No data available*

Table 6-11: Burden of disease in children with obvious caries experience at Sweep 1 and Sweep 2

Total number of children in the study	N= 219	
Total examined	Sweep 1 (AMR) n= 190	Sweep 2 n= 175
Proportion with obvious caries in primary teeth % [n]	35.8 [68]	47.7 [83]
Mean AMR/$d_3$mft [SD]	3.7 [3.1]	3.9 [3.2]
Minimum, Maximum	1, 18	1, 18
Median [Q_1, Q_3]	3.0 [1, 5]	3.0 [2, 5]

[14] For all children clinically examined in Sweep 2

[15] For those children who had complete data in Sweep 1 and clinical dental data in Sweep 2

[16] NHS Greater Glasgow & Clyde; McMahon et al., 2011

[17] Glasgow city CHP; NDIP, 2014,

[18] Representative sample

6.3 Longitudinal changes in caries experience

Table 6-12 shows the longitudinal caries experience status for the 159 children who had a clinical dental examination completed at both time points. The proportion of children with no obvious caries experience at both Sweep 1 and Sweep 2 was 51.6%. Fifteen percent of the children with no caries experience in Sweep 1 were found to have caries experience in Sweep 2.

Table 6-12: Caries progression in children who had clinical data at baseline year (Sweep 1) and follow-up year (Sweep 2)

Total number of children who had clinical data at both time points		N= 159
Caries experience		**% [n]**
Sweep 1	**Sweep 2**	
No	No	51.6 [82]
No	Yes	15.1 [24]
Yes	Yes	33.3 [53]
Yes	No	0

6.4 Developing a disease model

The following section addresses objective 3 (i). It reports the results of the longitudinal analyses, which utilises the hypothetical pathway in Figure 4.3 to investigate which of the risk factors measured in Sweep 1 are associated with caries experience in Sweep 2 (outcome).

6.4.1 Univariable Analysis

All the variables detailed in Table 4-1, collected in Sweep 1 were univariably analysed to produce unadjusted Odds Ratios (OR) and 95% confidence intervals (CI) for caries experience (dichotomised $d_3$mft>0) in Sweep 2 following the methods described in ***Section 4.7.3***. The significance level was based on p value being <0.20. This conservative level was set due to the relatively small sample size in this pilot study, which might result in wide confidence intervals that might nonetheless indicate potentially important effects. Variables that had p >0.20 and those that showed a low predictability (Lower CI of C-index <0.50) were excluded from the next stage of analysis.

6.4.1.1 Demographics domain

Children from single parent families (compared to two parent households) were more likely to have experience of caries (OR [95%CI], 1.68 [0.81 to 3.47], p= 0.17) as were those that belonged to homes with a smoker (compared to homes with no smokers) (Table 6-13). For a child whose main-carer was reported as a 'current smoker' the unadjusted OR [95% CI] for caries experience at Sweep 2 was 1.77 [0.85 to 3.68] (p= 0.13).

Table 6-13: Unadjusted Odds Ratios & 95% Confidence Intervals for caries experience (Sweep 2) and C-index according to demographic domain (Sweep 1)

Explanatory Variables	Caries experience N	%	Unadjusted OR [95% CI]
Child characteristics			
Gender			
Male	[46/94]	48.9	1 [ref]
Female	[38/80]	47.5	0.94 [0.52 to 1.72]
P value			0.850
C-index [95% CI]			0.51 [0.42 to 0.60]
Age of child (years)			0.71 [0.30 to 1.70]
P value			0.439
C-index [95% CI]			0.54 [0.45 to 0.62]
Ethnicity			
White: Scottish	[53/113]	46.9	0.94 [0.43 to 2.09]
Others[19]	[15/31]	48.4	1 [ref]
P value			0.883
C-index [95%CI]			0.51 [0.41 to 0.60]
Number of children in family			
1	[13/31]	41.9	1 [ref]
2	[32/64]	50.0	1.39 [0.59 to 3.29]
3 or more	[21/47]	44.7	1.12 [0.45 to 2.80]
P value			0.728
C-index [95% CI]			0.54 [0.44 to 0.63]
Position of child in family			
First born	[28/63]	44.4	1 [ref]
Second born	[28/52]	53.9	1.46 [0.70 to 3.05]
Third born or later	[10/27]	37.0	0.74 [0.29 to 1.86]
P value			0.335
C-index [95% CI]			0.57 [0.47 to 0.66]
Parent/carer characteristics			
Age of main carer			
<25 years	[8/15]	53.3	1.65 [0.53 to 5.12]
26 -35 years	[35/69]	50.7	1.48 [0.74 to 2.97]
36 years and over	[25/61]	41.0	1 [ref]
P value			0.471
C-index [95% CI]			0.55 [0.46 to 0.65]
Main carer marital Status			
Married/Cohabiting	[45/104]	43.3	1[ref]
Single parent	[23/41]	56.1	1.68 [0.81 to 3.47]
P value			0.165
C-index [95% CI]			0.55 [0.46 to 0.65]
Main-Carer smoking status			
Current Smoker	[23/40]	57.5	1.77 [0.85 to 3.68]
Never smoked/smoked in past	[46/106]	43.4	1 [ref]
P value			0.130
C-index [95% CI]			0.56 [0.46 to 0.65]

[19] Other Whites, Asians, Blacks and Mixed

6.4.1.2 Socioeconomic circumstances

A strong socioeconomic gradient for caries experience was evident, with those considered most deprived experiencing higher levels of disease than those least deprived. All SEP measures in Sweep 1 except one (years of main-carer's full time education) showed an association with caries at Sweep 2, with children from lower SEP consistently showing higher odds of caries (Table 6-14). The strongest predictor of caries at Sweep 2 was the highest level of education of the main-carer. Children from households with the main-carer having no formal qualification or only a pre-school leaving certificate were 8 times (95% CI - 2.36 to 29.90, p= 0.009) more likely to have experience of caries at Sweep 2 compared to those with main-carers having a first degree or professional qualification. The lower limit of c- index was higher than 0.50 for all measures except one (main-carer's years of full time education), implying that SEP is a good predictor of caries.

Table 6-14: Unadjusted Odds Ratios & 95% Confidence Intervals for caries experience (Sweep 2) and C-index according to socioeconomic circumstances (Sweep 1)

Explanatory Variables	Caries experience N	%	Unadjusted OR [95% CI]
Local SIMD[20] Quintile			
Q1 (most deprived)	[38/67]	56.7	2.32 [1.01 to 5.34]
Q2	[22/35]	62.9	2.99 [1.14 to 7.87]
Q3	[8/33]	24.2	0.57 [0.20 to 1.61]
Q4 & Q5 (least deprived)	[13/36]	36.1	1 [ref]
P value			0.003
C-index [95% CI]			0.66 [0.58 to 0.74]
National SIMD[21] Quintile			
Q1 (most deprived)	[58/100]	58.0	3.45 [1.24 to 9.64]
Q2	[6/20]	30.0	1.07 [0.28 to 4.12]
Q3	[12/31]	38.7	1.58 [0.48 to 5.20]
Q4 & Q5 (least deprived)	[6/21]	28.6	1 [ref]
P value			0.015
C-index [95% CI]			0.63 [0.55 to 0.71]
Gross annual household Income			
< £10,000	[22/34]	64.7	4.07 [1.66 to 9.99]
£10,000 – £19,999	[23/43]	53.5	2.56 [1.13 to 5.79]
£20,000 & more	[18/58]	31.0	1 [ref]
P value			0.005
C-index [95% CI]			0.65 [0.56 to 0.74]
Main-Carer's years of Full-time Education			
Not completed secondary school	[22/45]	48.9	1.12 [0.55 to 2.26]
Further education	[44/95]	46.3	1 [ref]
P value			0.776
C-index [95% CI]			0.51 [0.42 to 0.61]

[20] Scottish Index of Multiple Deprivation 2009 at Local Health Board level (NHS Greater Glasgow & Clyde)

[21] Scottish Index of Multiple Deprivation 2009 at National level

Table 6-14 continued

Main-Carer's highest Level of Education			
Level 0[22]	[14/19]	73.7	8.40 [2.36 to 29.90]
Level 1[23]	[21/42]	50.0	3.00 [1.14 to 7.90]
Level 2[24]	[24/48]	50.0	3.00 [1.17 to 7.70]
Level 3[25]	[9/36]	25.0	1 [ref]
P value			0.009
C-index [95% CI]			0.65 [0.56 to 0.74]
Percentage of income from benefits			
Half or more	[38/59]	64.4	3.98 [1.95 to 8.12]
None or About a quarter	[25/80]	31.3	1 [ref]
P value			<0.001
C-index [95% CI]			0.66 [0.57 to 0.76]
Current or last occupation of Main-Carer			
Never worked or permanently unemployed due to disability	[5/9]	55.6	2.86 [0.59 to 13.96]
NS-SEC 3[26]	[27/43]	62.8	3.86 [1.31 to 11.38]
NS-SEC 2[27]	[7/26]	26.9	0.84 [0.24 to 2.91]
NS-SEC 1[28]	[7/23]	30.4	1 [ref]
P value			0.013
C-index [95% CI]			0.68 [0.57 to 0.78]
Housing Tenure			
Rented- not private[29]	[40/60]	66.7	3.91 [1.88 to 8.16]
Rented- privately	[5/17]	29.4	0.82 [0.26 to 2.60]
Owned/mortgaged	[23/68]	33.8	1 [ref]
P value			<0.001
C-index [95% CI]			0.67 [0.58 to 0.76]

6.4.1.3 Oral hygiene practices

Daily toothbrushing frequency, brushing before bedtime, the method used for toothpaste removal and age of commencement of regular toothbrushing were associated with caries experience at Sweep 2. The c-index somewhat reflected the same, with the former three variables predicting caries (c-index >0.50). Children who brushed less than twice daily were more likely to have experience of caries at Sweep 2 (OR 3.50, 95% CI 1.53 to 8.00, p= 0.003) than children who brushed twice or more daily. Children who commenced

[22] Level 0 - no qualification or pre-school leaving qualification

[23] Level 1 - O grade, standard grade, GCSE or equivalent

[24] Level 2 - Higher grade, A level, GSVQ advanced HNC, HND, SVQ Levels 4 or 5 or equivalent

[25] Level 3 - first degree, higher degree or professional qualification

[26] NS-SEC 3 - Routine & manual

[27] NS-SEC 2 - Intermediate

[28] NS-SEC 1 - Higher Managerial, Administrative & Professionals

[29] Rented from local Authority/Council/Housing association

toothbrushing after their first birthday and those who irregularly brushed before bedtime were also at an increased risk of caries at Sweep 2 compared to those who brushed before their first birthday and those who always brushed before bedtime (ORs [95%CI] of 2.69 [1.16 to 5.74] and 3.01 [1.53 to 5.94] respectively). In addition, there was an increased odds of caries experience at Sweep 2 in those rinsing out toothpaste compared to those who spat (OR [95% CI]: 3.54 [1.57 to 7.96], p= 0.006).

Table 6-15: Unadjusted Odds Ratios & 95% Confidence Intervals for caries experience (Sweep 2) and C-index according to Oral hygiene practices (Sweep 1)

Explanatory Variables	Caries experience N	%	Unadjusted OR [95% CI]
Daily toothrushing frequency			
Less than twice	[24/34]	70.6	3.50 [1.53 to 8.00]
Twice or more than twice	[46/113]	40.7	1[ref]
P value			0.003
C-index [95% CI]			0.61 [0.51 to 0.70]
Brush before bedtime			
Always	[28/80]	35.0	1 [ref]
Sometimes / Occasionally	[39/61]	63.9	3.29 [1.64 to 6.60]
Hardly ever/ Never	[3/6]	50.0	1.86 [0.35 to 9.82]
P value			0.004
C-index [95% CI]			0.64 [0.55 to 0.73]
Age when toothbrushing started			
Under 12 months/First tooth erupted	[51/116]	44.0	1 [ref]
After 12 months	[19/30]	63.3	2.20 [0.96 to 5.04]
P value			0.062
C-index [95% CI]			0.56 [0.47 to 0.66]
Adult supervised tooth-brushing			
Yes	[62/125]	49.6	1 [ref]
No	[8/22]	36.4	0.58 [0.23 to 1.48]
P value			0.255
C-index [95% CI]			0.53 [0.44 to 0.62]
Method for toothpaste removal			
Swallow	[12/21]	57.1	2.26 [0.86 to 5.94]
Spit	[33/89]	37.1	1 [ref]
Rinse out	[25/37]	67.6	3.54 [1.57 to 7.96]
P value			0.006
C-index [95% CI]			0.64 [0.55 to 0.73]
Amount of toothpaste used			
Recommended: Pea-size	[46/100]	46.0	1 [ref]
Others: Smear, Half a brush, Full brush	[24/47]	51.1	1.23 [0.61 to 2.45]
P value			0.567
C-index [95% CI]			0.52 [0.43 to 0.62]

6.4.1.4 Use of dental services

Older age at first dental visit and infrequent routine dental checks increased the odds of caries among children at Sweep 2 (Table 6-16).

As expected, when the reason for the first dental visit was for other than anticipatory care, the odds of caries among children at Sweep 2 were very high (OR [95%CI] 8.85 [1.06 to 74.17], p value= 0.11).

Table 6-16: Unadjusted and adjusted Odds Ratios & 95% Confidence Intervals for caries experience (Sweep 2) and C-index according to Use of dental services (Sweep 1)

Explanatory Variables	Caries experience		Unadjusted OR [95% CI]
	N	%	
Age at first dental visit			
Never been to dentist	[10/19]	52.6	1.96 [0.69 to 5.58]
Within first year after birth	[21/58]	36.2	1 [ref]
After first year of birth	[39/68]	57.4	2.37 [1.15 to 4.87]
P value			0.059
C-index [95% CI]			0.60 [0.51 to 0.70]
Reason for first dental visit			
Corrective	[7/8]	87.5	8.85 [1.06 to 74.17]
Anticipatory	[53/120]	44.2	1 [ref]
Never visited a dentist	[10/19]	52.6	1.41 [0.53 to 3.71]
P value			0.113
C-index [95% CI]			0.56 [0.47 to 0.66]
Frequency of Routine check up			
<= 6 months	[45/100]	45.0	1 [ref]
At least every 12 months	[9/20]	45.0	1.00 [0.38 to 2.63]
>12 months	[4/5]	80.0	4.89 [0.53 to 45.30]
Never	[12/22]	54.6	1.47 [0.58 to 3.71]
P value			0.475
C-index [95% CI]			0.55 [0.45 to 0.64]

6.4.1.5 Early feeding habits

All variables used to measure early feeding habits were associated with caries experience at Sweep 2 in the univariable analysis (Table 6-17). The c-index mirrored these results, with all variables under this domain except one (night time bottle feeding other than water up to four years) predicting caries at Sweep 2. The OR for caries experience at Sweep 2 increased when sugar intake restrictions enforced at home were relaxed, with OR [95% CI] for children reported to be 'hardly/never restricted' being 2.94 [1.25 to 6.94] (p= 0.005). Compared to those children who were weaned from bottle between one and two years, the odds of those who were weaned before 12 months showing caries experience at Sweep 2 was 3.52 [1.35 to 9.19] (p= 0.038). Children who were weaned on to solids later than 6 months also showed high odds for caries at Sweep 2 (OR [95%CI]: 2.55 [1.13 to 5.73], p= 0.076). Compared to those children who were never bottle fed during the night time at two years, the odds of those who were 'sometimes' bottle fed was 2.08 [1.01 to 4.26] (p= 0.034).

Table 6-17: Unadjusted Odds Ratios & 95% Confidence Intervals for caries experience (Sweep 2) and C-index according to Early feeding habits (Sweep 1)

Explanatory Variables	Caries experience N	%	Unadjusted OR [95% CI]
Breast or bottle feeding			
Breast fed only	[13/32]	40.6	1 [ref]
Bottle fed only	[46/82]	56.1	1.87 [0.82 to 4.28]
Breast and bottle fed	[11/33]	33.3	0.73 [0.27 to 2.01]
P value			0.061
C-index [95% CI]			0.60 [0.51 to 0.69]
Age bottle feeding completely stopped			
Never used a baby bottle	[3/11]	27.3	0.56 [0.14 to 2.28]
Under 12 months	[19/27]	70.4	3.52 [1.35 to 9.19]
Between 1 yr and 2 yrs	[27/67]	40.3	1 [ref]
Over two years or Still using baby bottle	[21/42]	50.0	1.48 [0.68 to 3.22]
P value			0.038
C-index [95% CI]			0.63 [0.54 to 0.72]
Night time bottle feeding (other than water) up to two years			
Every/ Most days	[30/55]	54.6	2.08 [1.01 to 4.26]
Some days	[13/20]	65.0	3.21 [1.14 to 9.08]
Hardly ever/ Never	[26/71]	36.6	1 [ref]
P value			0.034
C-index [95% CI]			0.62 [0.52 to 0.71]
Frequency sugar restricted			
Every/ Most days	[42/107]	39.3	1 [ref]
Some days	[19/29]	65.5	6.96 [1.43 to 33.83]
Hardly ever/ Never	[9/11]	81.8	2.94 [1.25 to 6.94]
P value			0.005
C-index [95% CI]			0.63 [0.54 to 0.72]
Night time bottle feeding (other than water) up to four years			
Every/ Most days	[14/22]	63.6	2.31 [0.89 to 5.98]
Some days	[12/21]	57.1	1.76 [0.68 to 4.54]
Hardly ever/ Never	[44/102]	43.1	1 [ref]
P value			0.154
C-index [95% CI]			0.58 [0.48 to 0.67]
Age of commencement of solids			
< 4 months	[7/14]	50.0	1.39 [0.45 to 4.27]
4-6 months	[41/98]	41.8	1 [ref]
7 months and over	[22/34]	64.7	2.55 [1.13 to 5.73]
P value			0.076
C-index [95% CI]			0.59 [0.50 to 0.68]

6.4.1.6 Current Diet

NMES as a percentage of food energy collected in Sweep 1 was associated with caries at Sweep 2 in the univariable analysis (Table 6-18). For every additional one percent of NMES as a percentage of food energy at Sweep 1, the odds of a child showing caries at Sweep 2 by nine percent (OR 1.09, 95% CI [1.02 to 1.16], p= 0.007). In addition, children whose NMES intake was above the recommended 11% of food energy showed increased odds of showing caries at Sweep 2 (OR 1.83, 95%CI [0.78 to 4.30] (p= 0.165). Of the food groups that were analysed, children with the medium and highest intake of 'crisps and savoury snacks' were at increased odds of caries. Children in the highest tertile of 'crisps and savoury snacks' showed the highest risk compared with those in the lowest tertile of intake (OR 2.18, 95%CI [0.98 to 4.87] (p= 0.106). Although children with the medium and highest intake of 'non- diet soft drinks' were at increased odds of caries compared with those in the lowest intake tertile, the p value was greater than the threshold of 0.20.

Table 6-18: Unadjusted Odds Ratios & 95% Confidence Intervals for caries experience (Sweep 2) and C-index according to Diet (Sweep 1)

Explanatory Variables	Caries experience N	%	Unadjusted OR [95% CI]
NMES<11% food energy	[10/28]	35.7	1 [ref]
NMES>=11% food energy	[59/117]	50.4	1.83 [0.78 to 4.30]
P value			0.165
C-index [95% CI]			0.55 [0.45 to 0.64]
NMES (% food energy)			1.09 [1.02 to 1.16]
P value			0.007
C-index [95% CI]			0.60 [0.51 to 0.69]
NMES (g/day)			
T1: <= 51.9	[20/49]	40.8	1 [ref]
T2: 52.0 to 81.1	[25/51]	49.0	1.39 [0.63 to 3.08]
T3: 81.2 & above	[24/45]	53.3	1.66 [0.73 to 3.75]
P value			0.465
C-index [95% CI]			0.56 [0.46 to 0.65]
Total Sugars (g/day)			
T1: <=103.2	[20/49]	40.8	1 [ref]
T2: 103.3 to 149.0	[24/49]	49.0	1.39 [0.63 to 3.10]
T3: 149.1 & above	[25/47]	53.2	1.65 [0.73 to 3.70]
P value			0.467
C-index [95% CI]			0.56 [0.46 to 0.65]
Crisps & Savoury Snacks (g/day)			
T1: <=9.63	[21/51]	41.2	1 [ref]
T2: 9.64 to 18.38	[18/43]	41.9	1.03 [0.45 to 2.34]
T3: 18.39 & above	[29/48]	60.4	2.18 [0.98 to 4.87]
P value			0.106
C-index [95% CI]			0.59 [0.49 to 0.68]
Biscuits, Cakes & Pastries (g/day)			
T1: <=20.84	[22/46]	47.8	1 [ref]
T2: 20.85 to 36.93	[23/46]	50.0	1.09 [0.48 to 2.47]
T3: 36.94 & above	[20/45]	44.4	0.87 [0.38 to 1.99]
P value			0.867
C-index [95% CI]			0.53 [0.43 to 0.62]
Confectionary (g/day)			
T1: <= 10.69	[19/42]	45.2	1 (ref]
T2: 10.70 to 23.81	[25/54]	46.3	1.04 [0.46 to 2.35]
T3: 23.82 & above	[25/47]	53.2	1.38 [0.60 to 3.17]
P value			0.707
C-index [95% CI]			0.54 [0.44 to 0.63]
Non-Diet Soft Drinks (g/day)			
T1: <= 70.0	[21/51]	41.2	1 [ref]
T2: 70.01 to 161.35	[20/38]	52.6	1.59 [0.68 to 3.70]
T3: 161.36 & above	[24/42]	57.1	1.91 [0.83 to 4.36]
P value			0.283
C-index [95% CI]			0.57 [0.48 to 0.67]

6.4.1.7 Parental attitudes and beliefs

The parental attitudes scale was associated with caries at Sweep 2 in the univariable analysis, with more positive parental attitudes towards oral health showing lower odds of caries among children (Table 6-19). Similarly, higher parental efficacy score showed a slightly reduced odds of caries at Sweep 2. The lower limit of the c-index for the former scale was higher than 0.50, implying it to predict caries at Sweep 2.

Table 6-19: Unadjusted Odds Ratios & 95% Confidence Intervals for caries experience (Sweep 2) and C-index according to Parental Attitudes and Beliefs (Sweep 1)

Explanatory Variables	Unadjusted OR [95% CI]
Positive Parental attitudes Scale	0.54 [0.28 to 1.06]
P value	0.072
C-index	0.60 [0.51 to 0.69]
Parental Efficacy scale	0.74 [0.49 to 1.12]
P value	0.155
C-index	0.59 [0.50 to 0.69]

6.4.1.8 Streptococcus mutans

As the levels of salivary *S. mutans* counts in Sweep 1 increased, the odds of a child showing experience of caries at Sweep 2 increased, with children in the highest tertile of salivary *S. mutans* counts in Sweep 1 showing an OR 16.09, 95% CI 5.69 to 45.63 (p <0.001) compared to those who had no *S. mutans* detected (Table 6-20). Univariably, this was the most highly predictive variable.

Table 6-20: Unadjusted Odds Ratios & 95% Confidence Intervals for caries experience (Sweep 2) and C-index according to *S. mutans* (Sweep 1)

Explanatory Variable	Caries experience		Unadjusted OR [95% CI]
	N	%	
Microbiology			
S. mutans (CFU/ml)			
T1 = not detected	[10/49]	20.4	1 [ref]
T2 = 1 to 3.9 x 10 4	[25/46]	54.4	4.64 [1.88 to 11.48]
T3 = > 3.9 x 10 4	[33/41]	80.5	16.09 [5.69 to 45.63]
P value			<0.001
C-index			0.77 [0.69 to 0.85]

6.4.2 Multivariable logistic regression analyses

In the last section, univariable analysis identified risk factors within the five domains (oral hygiene practices, early feeding habits, use of dental services, current diet and parental attitudes & beliefs) that were associated with caries in Sweep 2.

Following the modelling strategy determined by the hypothetical pathway in Figure 4.3 those risk factors with p value <0.20 and lower level of CI of C-index >0.50 were offered to a multivariable model one domain at a time- the results of which are available in Appendix L. The 'winning candidates' at this stage were offered to a final multivariable model in which all risk factors from all domains were analysed simultaneously. Due to S. *mutans* being more proximal in the hypothetical pathway, it was expected to show stronger associations with caries than other risk factors and possibly eliminating the relatively more distal factors. Hence, two final models were run- one with all the relevant risk factors excluding *S.mutans* counts, and the other with the inclusion of *S.mutans* counts- the results of these two final models are presented here.

Those risk factors measured in Sweep 1 that were independently associated with caries experience at Sweep 2 are shown in Table 6-21. Age and gender-adjusted logistic regression models showed the risk factors independently associated with caries at Sweep 2 to be living in a deprived area, less than twice daily toothbrushing, irregular brushing habits before bedtime, rinsing out toothpaste after tooth-brushing, age when bottle feeding was completely stopped, poor sugar intake restrictions enforced at home, higher amounts of NMES consumption and older age at first visit to dentist. When *S. mutans* counts at Sweep 1 was added to the model, *S. mutans* counts showed a strong association with caries at Sweep 2, with the *S. mutans* levels being the most important risk factor for caries at Sweep 2. However, *S. mutans* levels did not completely dominate the risk and most of the behavioural risk factors in the previous model and socioeconomic position continued to remain independently associated with caries at Sweep 2.

The C-index of the final model excluding *S. mutans* counts was 0.83, which rose to 0.89 when *S. mutans* counts was added to the model. The ROC plots are shown in Figure 6.3 a. The greater distance of the curve from the diagonal line indicates the greater predictive ability of the model, with the distance further increasing when *S. mutans* counts were added to the model (Figure 6-4b).

Table 6-21: Adjusted Odds Ratios & 95% Confidence Intervals for caries experience (Sweep 2) according to riskfactors measured in Sweep 1

Explanatory Variables	Adjusted OR [95% CI] excluding *S. mutans* counts	Adjusted OR [95% CI] including *S.mutans* counts
Gender		
Male	1 [ref]	1 [ref]
Female	1.51 [0.60 to 3.84]	0.85 [0.23 to 3.12]
P value	0.385	0.807
Age of child (years)	0.70 [0.19 to 2.50]	1.72 [0.35 to 8.54]
P value	0.577	0.508
National SIMD[30] Quintile		
Q1 (most deprived)	2.09 [0.52 to 8.50]	1.80 [0.32 to 10.17]
Q2	0.38 [0.05 to 2.63]	0.18 [0.01 to 2.64]
Q3	1.45 [0.27 to 7.69]	0.74 [0.08 to 6.70]
Q4 & Q5 (least deprived)	1 [ref]	1 [ref]
P value	0.111	0.149
Daily toothbrushing frequency		
Less than twice	2.88 [0.78 to 10.67]	2.19 [0.36 to 13.18]
Twice or more than twice	1 [ref]	1 [ref]
P value	0.113	0.393
Brush before bedtime		
Always	1 [ref]	1 [ref]
Sometimes / Occasionally	1.31 [0.45 to 3.85]	0.97 [0.22 to 4.28]
Hardly ever/ Never	0.12 [0.01 to 1.93]	0.02 [0 to 0.82]
P value	0.177	0.065
Method for toothpaste removal		
Swallow	4.12 [1.10 to 15.49]	9.93 [1.69 to 58.42]
Spit	1 [ref]	1 [ref]
Rinse out	4.40 [1.44 to 13.46]	3.42 [0.73 to 16.03]
P value	0.008	0.020
Breast or bottle feeding		
Breast fed only	1 [ref]	1 [ref]
Bottle fed only	0.95 [0.26 to 3.51]	1.02 [0.23 to 4.58]
Breast and bottle fed	0.47 [0.11 to 2.05]	0.26 [0.04 to 1.77]
P value	0.462	0.282

[30] Scottish Index of Multiple Deprivation 2009 at National level

Explanatory variables	Adjusted OR [95% CI] excluding *S. mutans* counts	Adjusted OR [95% CI] including *S.mutans* counts
Age bottle feeding completely stopped		
Never used a baby bottle	0.38 [0.04 to 3.52]	1.42 [0.10 to 19.77]
Under 12 months	3.64 [0.99 to 13.34]	3.23 [0.52 to 20.05]
Between one year and two years	1 [ref]	1 [ref]
Over two years or Still using baby bottle	0.83 [0.30 to 2.31]	1.67 [0.42 to 6.62]
P value	0.143	0.646
Night time bottle feeding up to two years		
Every/ Most days	2.25 [0.83 to 6.06]	4.01 [1.03 to 15.58]
Some days	1.15 [0.27 to 4.86]	2.39 [0.31 to 18.52]
Hardly ever/ Never	1 [ref]	1 [ref]
P value	0.276	0.128
Frequency sugar restricted		
Every/ Most days	1 [ref]	1 [ref]
Some days	2.20 [0.71 to 6.81]	0.86 [0.19 to 3.90]
Hardly ever/ Never	8.86 [1.03 to 76.27]	7.47 [0.44 to 126.83]
P value	0.080	0.348
NMES (% food energy)	1.06 [0.98 to 1.14]	1.08 [0.98 to 1.20]
P value	0.134	0.130
Age at first dental visit		
Never been to dentist	1.36 [0.32 to 5.79]	2.62 [0.39 to 17.41]
Within first year after birth	1 [ref]	1 [ref]
After first year of birth	2.87 [1.11 to 7.42]	4.53 [1.16 to 17.68]
P value	0.090	0.092
***S. mutans* (CFU/ml)**		
T1 = not detected		1 [ref]
T2 = 1 to 3.9 x 10 4	NA	4.48 [1.07 to 18.83]
T3 = > 3.9 x 10 4		36.31 [5.90 to 223.29]
P value		0.001
C-index for full model	0.83 [0.77 to 0.90]	0.89 [0.84 to 0.95]

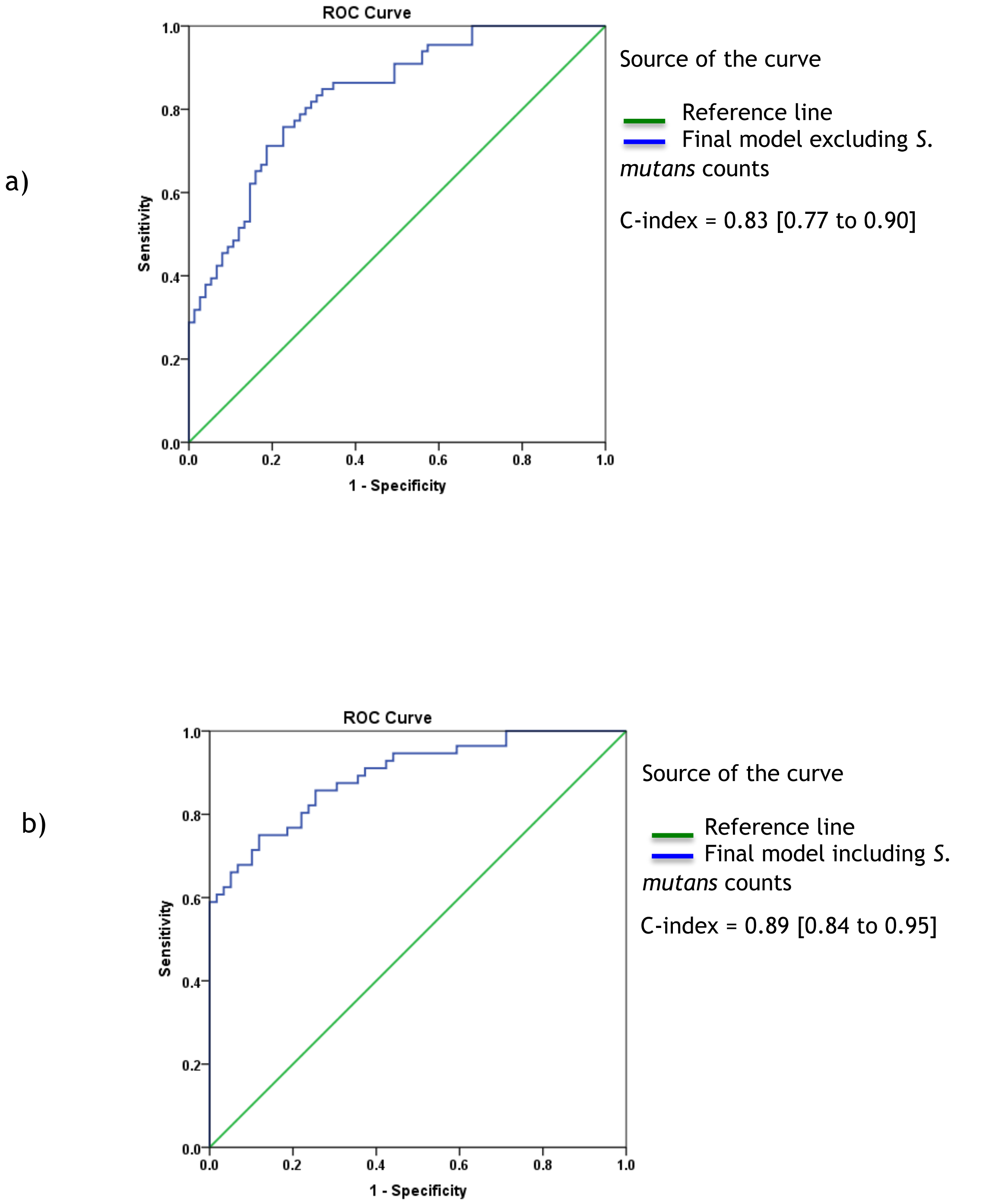


Figure 6-4: ROC plot indicating the C-index for a) final model excluding *S. mutans*, b) final model including *S.mutans* counts

6.5 Explaining the socioeconomic gradient in caries

The following section addresses objective 3 (ii) and explores the effect of different measures of SEP on caries experience, and which of the behavioural risk factors (if any) found to be associated with caries (***Section 6.4.2***) may help explain the socioeconomic gradient in caries.

Binary logistic regression was used to produce odds ratios and 95% confidence intervals for caries experience according to all SEP measures that had a p value <0.20 and C-index >0.50 in the univariable analysis, and these were adjusted for age, gender and the risk factors found to be associated with caries in Sweep 2 (***Section 6.4.2***): first one domain at a time and then a final model which considered all factors from all domains.

Table 6-22 illustrates that irrespective of the socio-economic measure used in the analysis, there is a strong socioeconomic gradient in caries experience at Sweep 2, with those in the most disadvantaged groups showing the highest risk of caries. The gradient is partly attenuated when adjusted for reported daily toothbrushing frequency, toothbrushing before bedtime, method of toothpaste removal after toothbrushing, age baby was completely weaned from bottle feeding, frequency of sugar restricted, NMES intake (% food energy) and age at first dental visit (Table 6-23). Controlling for the effect of the aforementioned factors, there was still a 1.89 times higher chance of a child living in the most deprived SIMD quintile showing caries at Sweep 2 than a child in the least deprived SIMD quintile. Similarly, children from households with more than 50% of the income obtained from benefits were 2.59 times more likely to have caries experience than those from households with little (a quarter of income) or no benefits. Those from households rented from local Authority/Council/Housing association were also 1.84 times more likely to have caries experience compared to children with parents who owned their house.

However, it is difficult from these results to determine which SEP measure might prove to be a stronger indicator of inequality than the others due to the differences in distribution across categories of the measures (Shaw et al., 2007).

All SEP measures were then standardized to determine if one measure of SEP would prove to be a stronger indicator of inequality than the others.

Table 6-22: Unadjusted and adjusted Odds Ratios and 95% Confidence Intervals for caries experience (Sweep 2) according to socioeconomic position-Adjusted for child characteristics, oral hygiene practices, early feeding habits, current diet.

SEP Variables	Unadjusted OR [95% CI]	Adjusted OR [95% CI][31]	Adjusted OR [95% CI][32]	Adjusted OR [95% CI][33]	Adjusted OR [95% CI][34]
National SIMD Quintile					
Q1 (most deprived)	3.45 [1.24 to 9.64]	3.35 [1.19 to 9.42]	3.22 [0.99 to 10.45]	3.47 [1.02 to 11.83]	3.85 [1.21 to 12.23]
Q2	1.07 [0.28 to 4.12]	1.03 [0.26 to 4.06]	0.73 [0.16 to 3.27]	0.85 [0.18 to 4.09]	1.11 [0.25 to 4.89]
Q3	1.58 [0.48 to 5.20]	1.54 [0.47 to 5.08]	1.36 [0.34 to 5.48]	1.88 [0.46 to 7.70]	1.42 [0.37 to 5.43]
Q4 & Q5 (least deprived)	1 [ref]	1 [ref]	1 [ref]	1 [ref]	1 [ref]
P value	0.015	0.017	0.018	0.040	0.015
Gross annual household Income					
< £10,000	4.07 [1.66 to 9.99]	3.88 [1.57 to 9.60]	3.72 [1.40 to 9.88]	2.99 [1.11 to 8.06]	3.29 [1.30 to 8.30]
£10,000 – £19,999	2.56 [1.13 to 5.79]	2.57 [1.13 to 5.84]	2.33 [0.92 to 5.92]	1.73 [0.68 to 4.43]	2.29 [0.98 to 5.34]
£20,000 & more	1 [ref]	1 [ref]	1 [ref]	1 [ref]	1 [ref]
P value	0.005	0.007	0.025	0.093	0.026
Main-Carer's highest Level of Education					
Level 0[35]	8.40 [2.36 to 29.90]	8.35 [2.34 to	6.77 [1.72 to 26.63]	4.76 [1.20 to 18.89]	8.20 [2.26 to 29.81]
Level 1[36]	3.00 [1.14 to 7.90]	2.96 [1.12 to 7.80]	2.55 [0.88 to 7.34]	1.65 [0.54 to 5.04]	2.51 [0.92 to 6.83]
Level 2[37]	3.00 [1.17 to 7.70]	2.99 [1.16 to 7.70]	2.38 [0.86 to 6.55]	2.27 [0.82 to 6.26]	2.76 [1.05 to 7.23]
Level 3[38]	1 [ref]	1 [ref]	1 [ref]	1 [ref]	1 [ref]
P value	0.009	0.009	0.051	0.128	0.014

31 Adjusted for age and gender

32 Adjusted for daily toothbrushing frequency, brush before bedtime and method for toothpaste removal

33 Adjusted for age bottle feeding completely stopped and frequency of sugar restricted

34 Adjusted for NMES intake (% energy)

35 Level 0 - no qualification or pre-school leaving qualification
36 Level 1 - O grade, standard grade, GCSE or equivalent
37 Level 2 - Higher grade, A level, GSVQ advanced HNC, HND, SVQ Levels 4 or 5 or equivalent
38 Level 3 - first degree, higher degree or professional qualification

Table 6.22 continued

SEP Variables	Unadjusted OR [95% CI]	Adjusted OR [95% CI][39]	Adjusted OR [95% CI][40]	Adjusted OR [95% CI][41]	Adjusted OR [95% CI][42]
Current or last occupation of Main-Carer					
Never worked or permanently unemployed due to disability	2.86 [0.59 to 13.96]	2.81 [0.57 to 13.82]	0.70 [0.09 to 5.36]	0.48 [0.04 to 5.35]	1.63 [0.28 to 9.37]
NS-SEC 3[43]	3.86 [1.31 to 11.38]	3.76 [1.27 to	2.32 [0.69 to 7.83]	2.81 [0.72 to 10.97]	3.00 [0.99 to 9.12]
NS-SEC 2[44]	0.84 [0.24 to 2.91]	0.79 [0.22 to 2.82]	0.60 [0.15 to 2.38]	0.73 [0.17 to 3.13]	0.89 [0.25 to 3.12]
NS-SEC 1[45]	1 [ref]	1 [ref]	1 [ref]	1 [ref]	1 [ref]
P value	0.013	0.013	0.103	0.104	0.099
Housing Tenure					
Rented- not private[46]	3.91 [1.88 to 8.16]	3.88 [1.86 to 8.11]	3.27 [1.49 to 7.18]	2.67 [1.19 to 6.03]	3.33 [1.56 to 7.10]
Rented Privately	0.82 [0.26 to 2.60]	0.82 [0.26 to 2.61]	0.60 [0.17 to 2.15]	0.60 [0.17 to 2.13]	0.76 [0.23 to 2.54]
Owned/mortgaged	1 [ref]	1 [ref]	1 [ref]	1 [ref]	1 [ref]
P value	<0.001	0.001	0.003	0.015	0.003
Percentage of income from benefits					
Half or more	3.98 [1.95 to 8.12]	4.01 [1.96 to 8.20]	4.16 [1.90 to 9.11]	2.63 [1.19 to 5.85]	3.63 [1.76 to 7.51]
None or About a quarter	1 [ref]	1 [ref]	1 [ref]	1 [ref]	1 [ref]
P value	<0.001	<0.001	<0.001	0.017	0.001

[39] Adjusted for age and gender

[40] Adjusted for daily toothbrushing frequency, brush before bedtime and method for toothpaste removal

[41] Adjusted for age bottle feeding completely stopped and frequency of sugar restricted

[42] Adjusted for NMES intake (% energy)

[43] NS-SEC 3 - Routine & manual

[44] NS-SEC 2 - Intermediate

[45] NS-SEC 1 - Higher Managerial, Administrative & Professionals

[46] Rented from local Authority/Council/Housing association

Table 6-23: Unadjusted and adjusted Odds Ratios and 95% Confidence Intervals for caries experience (Sweep 2) according to socioeconomic position- Adjusted for age at first dental visit and full adjusted model.

SEP variables	Unadjusted OR [95% CI]	Adjusted OR [95% CI][47]	Fully Adjusted OR [95% CI][48]
National SIMD Quintile			
Q1 (most deprived)	3.45 [1.24 to 9.64]	4.19 [1.34 to 13.09]	1.89 [0.48 to 7.49]
Q2	1.07 [0.28 to 4.12]	0.94 [0.22 to 3.94]	0.23 [0.03 to 1.64]
Q3	1.58 [0.48 to 5.20]	1.56 [0.41 to 5.95]	1.33 [0.26 to 6.93]
Q4 & Q5 (least deprived)	1 [ref]	1 [ref]	1 [ref]
P value	0.015	0.005	0.049
Gross annual household Income			
< 10,000	4.07 [1.66 to 9.99]	3.48 [1.39 to 8.73]	1.82 [0.56 to 5.73]
10,000 – 19,999	2.56 [1.13 to 5.79]	2.24 [0.97 to 5.19]	1.21 [0.40 to 3.70]
20,000 & more	1 [ref]	1 [ref]	1 [ref]
P value	0.005	0.021	0.585
Main-Carer's highest Level of Education			
Level 0[49]	8.40 [2.36 to 29.90]	8.63 [2.32 to 32.17]	4.68 [0.93 to 23.36]
Level 1[50]	3.00 [1.14 to 7.90]	3.56 [1.30 to 9.72]	1.62 [0.44 to 6.00]
Level 2[51]	3.00 [1.17 to 7.70]	3.51 [1.32 to 9.35]	2.17 [0.66 to 7.12]
Level 3[52]	1 [ref]	1 [ref]	1 [ref]
P value	0.009	0.008	0.259

[47] Adjusted for age at first dental visit

[48] Adjusted for age, gender, daily toothbrushing frequency, brush before bedtime, method for toothpaste removal, age bottle feeding completely stopped, frequency of sugar restricted, NMES (% energy) and age at first dental visit

[49] Level 0 - no qualification or pre-school leaving qualification
[50] Level 1 - O grade, standard grade, GCSE or equivalent
[51] Level 2 - Higher grade, A level, GSVQ advanced HNC, HND, SVQ Levels 4 or 5 or equivalent
[52] Level 3 - first degree, higher degree or professional qualification

SEP variables	Unadjusted	Adjusted	Fully Adjusted
Current or last occupation of Main-Carer			
Never worked or permanently unemployed due to disability	2.86 [0.59 to 13.96]	1.49 [0.25 to 8.77]	0.05 [0.001 to 1.65]
NS-SEC 3[55]	3.86 [1.31 to 11.38]	3.36 [1.11 to 10.20]	1.22 [0.20 to 7.58]
NS-SEC 2[56]	0.84 [0.24 to 2.91]	0.76 [0.21 to 2.74]	0.41 [0.07 to 2.54]
NS-SEC 1[57]	1 [ref]	1 [ref]	1 [ref]
P value	0.013	0.032	0.136
Housing Tenure			
Rented- not private[58]	3.91 [1.88 to 8.16]	3.54 [1.67 to 7.49]	1.84 [0.74 to 4.59]
Rented- privately	0.82 [0.26 to 2.60]	0.74 [0.23 to 2.43]	0.44 [0.11 to 1.85]
Owned/mortgaged	1 [ref]	1 [ref]	1 [ref]
P value	<0.001	0.001	0.111
Percentage of income from benefits			
Half or more	3.98 [1.95 to 8.12]	3.63 [1.76 to 7.51]	2.59 [1.01 to 6.63]
None or About a quarter	1 [ref]	1 [ref]	1 [ref]
P value	<0.001	0.001	0.047

[53] Adjusted for age at first dental visit

[54] Adjusted for age, gender, daily toothbrushing frequency, brush before bedtime, method for toothpaste removal, age bottle feeding completely stopped, frequency of sugar restricted, NMES (% energy) and age at first dental visit

[55] NS-SEC 3 - Routine & manual

[56] NS-SEC 2 - Intermediate

[57] NS-SEC 1 - Higher Managerial, Administrative & Professionals

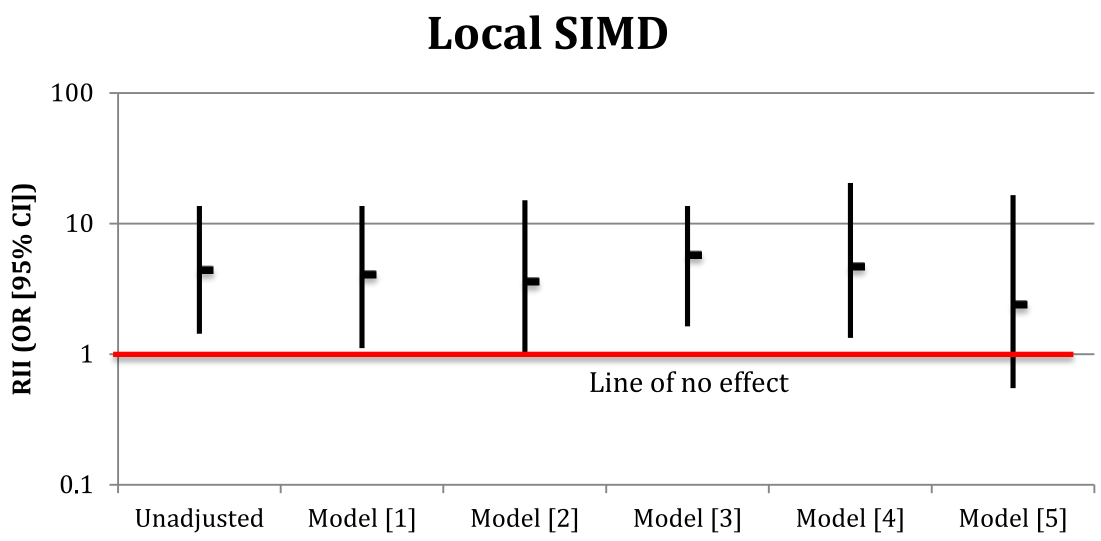
[58] Rented from local Authority/Council/Housing association

6.5.1 Relative Index of Inequality

The Relative Index of Inequality (RII) is a measure used to compare inequalities across populations that use the same socioeconomic measure or across the different socioeconomic measures within a population (Shaw et al., 2007). In the present study, RII analyses were carried out to determine if SEP measures when standardised; one measure of SEP would prove to be a stronger indicator of inequality than the others. In addition, it was used to explore the contribution of the risk factors found to be associated with caries in Sweep 2 from multivariable analysis (***Section 6.4.2)*** in explaining the observed socio-economic inequalities.

Figures 6-4 to 6-6 illustrates the RII (OR [95% CI]) for caries experience at Sweep 2 in the most disadvantaged children relative to the most advantaged. All measures of SEP demonstrated high RIIs for caries experience. While the inequality was greatest using gross household income as the measure of SEP, the RII attenuated significantly when adjusted for all risk factors found to be associated with caries in Sweep 2 (daily toothbrushing frequency, method for toothpaste removal, age bottle feeding was completely stopped, night time bottle feeding at two years, sugar restrictions enforced at home, NMES (% energy) and age at first dental visit). In contrast, the RII for national SIMD remained relatively stable even after adjusting for the aforementioned factors.

While the risk factors, age bottle feeding completely stopped, night time bottle feeding at two years and frequency of sugar restricted explained the gradient most across the SEP measures, the risk factors, NMES consumption (percentage of energy) and age at first dental visit increased the inequality. Adjusting for all risk factors from multivariable analysis associated with caries in Sweep 2 resulted in attenuation of RIIs across the five measures of socioeconomic position ranging from 36% to 81% for caries experience (Table 6-24). Despite some attenuation in the RII in all measures of SEP after adjustment for risk factors that were found to be associated with caries, clear social gradients in caries experience persisted with all measures of SEP (Figures 6.4 to 6.6). This suggests the persistence of residual confounding from underestimated or unmeasured factors like the wider social determinants.



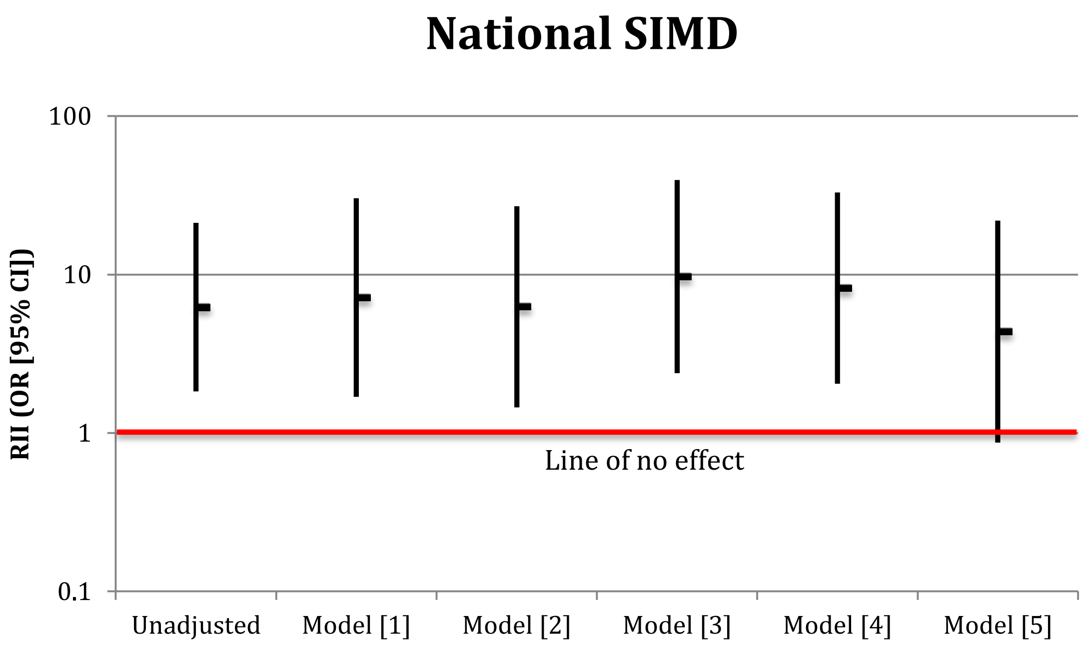


Figure 6-5: Relative Index of Inequality (RII) for the relation of a) Local SIMD to caries experience b)National SIMD to caries experience (RII (OR [95% CI are on log scale)

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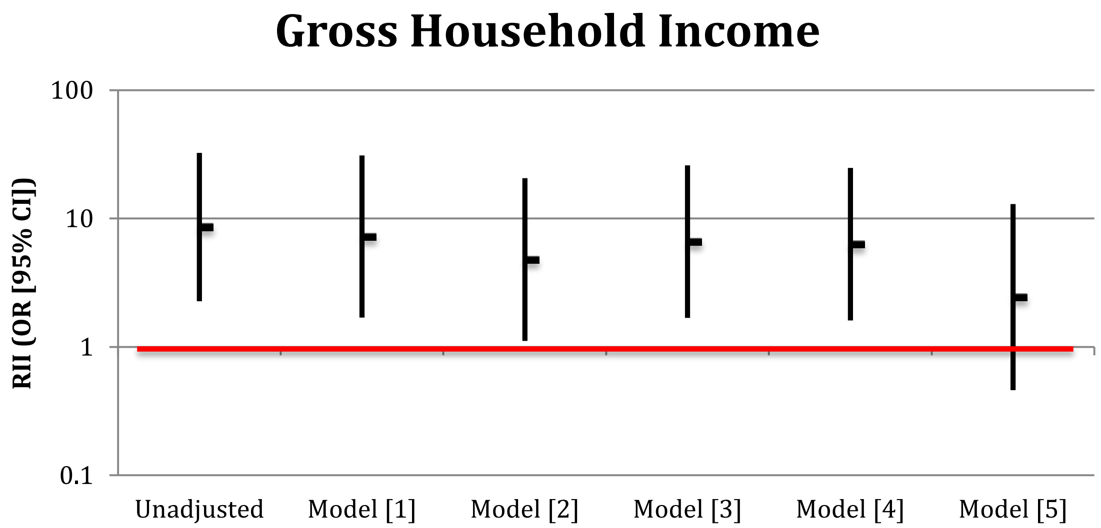
1 Adjusted for daily toothbrushing frequency, brush before bedtime and method of toothpaste removal
2 Adjusted for age bottle feeding completely stopped and frequency of sugar restricted
3 Adjusted for age at first dental visit
4 Adjusted for NMES (% food energy)
5 Adjusted for daily toothbrushing frequency, brush before bedtime, method for toothpaste removal, age bottle feeding completely stopped, frequency of sugar restricted, NMES (% energy) and age at first dental visit



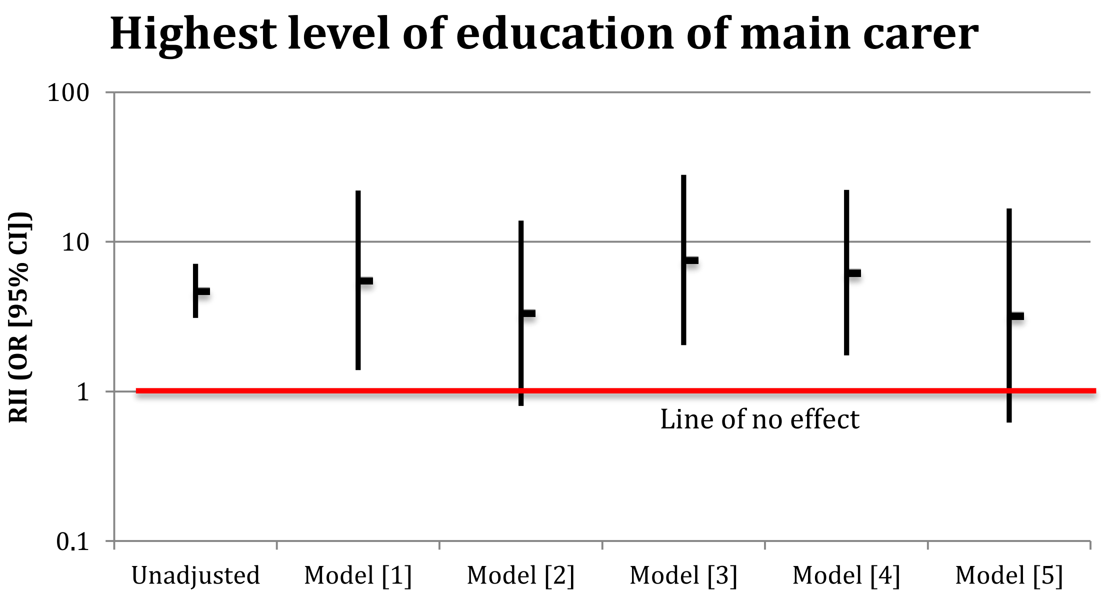


Figure 6-6: Relative Index of Inequality (RII) for the relation of a) Gross household income to caries experience b) Highest level of education of main-carer to caries experience
(RII (OR [95% CI are on log scale)

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1 Adjusted for daily toothbrushing frequency, brush before bedtime and method of toothpaste removal
2 Adjusted for age bottle feeding completely stopped and frequency of sugar restricted
3 Adjusted for age at first dental visit
4 Adjusted for NMES (% food energy)
5 Adjusted for daily toothbrushing frequency, brush before bedtime method for toothpaste removal, age bottle feeding completely stopped, frequency of sugar restricted, NMES (% energy) and age at first dental visit

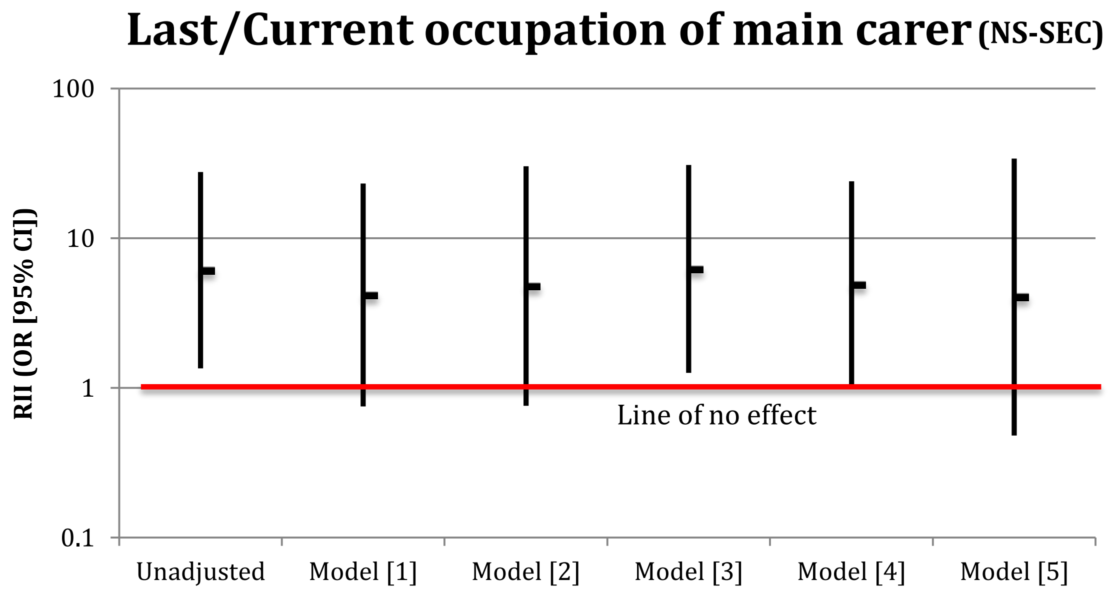


Figure 6-7: Relative Index of Inequality (RII) for the relation of Last/current occupation of main-carer to caries experience (RII (OR [95% CI are on log scale)

Table 6-24: Percentage reduction in RII for behavioural factors associated with caries in Sweep 2

	% reduction in RII[1]	% reduction in RII[2]	% reduction in RII[3]	% reduction in RII[4]	% reduction in RII[5]
Local SIMD	9	23	-40	-8	59
National SIMD	-15	-1	-66	-38	36
Gross Household Income	18	50	50	30	81
Highest level of education of main-carer	-22	37	-77	-41	40
Last/Current Occupation of main-carer	38	26	-2	24	40

Note: Negative value indicate that adjusting for risk factors widens the inequality

1 Adjusted for daily toothbrushing frequency, brush before bedtime and method of toothpaste removal
2 Adjusted for age bottle feeding completely stopped and frequency of sugar restricted
3 Adjusted for age at first dental visit
4 Adjusted for NMES (% food energy)
5 Adjusted for daily toothbrushing frequency, brush before bedtime, method for toothpaste removal, age bottle feeding completely stopped, frequency of sugar restricted, NMES (% energy) and age at first dental visit

6.6 Sample size considerations for larger cohort study

The most recent UK-wide Birth Cohort, Life Study has so far recruited a sample of 50,000 which is thought to provide adequate power to detect moderate causal effects of risk factors (environmental, psychosocial, genetic) that are moderately common (prevalence ≥10%) on multiple health outcomes of prevalence ≥2% (including asthma, obesity, autist disorders and learning difficulties) (Dezateux et al., 2013). However, the study is planned to recruit upto 90,000 babies and their families to provide high statistical power to examine the interplay between a range of biological, behavioural and environmental factors in various health outcomes; and also to account for potential drop-out and lower participation rates over time as the study runs from pregnancies to adulthood. Other large-scale studies like the Avon Longitudinal Study of Parents and Children (ALSPAC) have had no formal power calculations and attempted to enrol all pregnant women within a frame of expected date of delivery resident in the study area (Avon) (Golding, 1990).

Similar to the planned larger cohort study in this body of work, efforts underway in Australia in designing a birth cohort to investigate multiple risk factors in the development of caries in young children to inform interventions estimated their sample size to be 450 infants and their parents (de Silva-Sanigorski et al., 2010). A formal power calculation was carried out to detect a difference of 9% or greater in the prevalence of caries between various rurality groups with a power of 80% .

As discussed in ***Section 2.4.4***, pilot studies are important to check on an array of glitches in the preparation of a large-scale study. However, the MRC urges caution when determining effect size estimate from a pilot study for power or sample size calculations as pilot studies underestimate variation (Craig et al., 2008). The rule of thumb of ‘use an n of 30 or greater to estimate a parameter’ may not alleviate the issue, unless the effect size is large.

The sample size calculation for the larger cohort study will depend on a number of statistical and non-statistical factors, including the hypotheses to be tested, the variables of interest, desired accuracy and precision of effect estimates, recruitment rate, response rate, attrition and drop-out rate of the participants, geographical, demographical and social differences within the population and in their response to interventions. Additionally, only a part of the data that was collected is presented in this body of work, the analyses of some important and novel biological variables (cortisol, AMPs, sIgA etc) that are planned to be analysed in the larger cohort study where not intended to be explored in this body of work.

The sample size for the larger cohort study may be sensitive to the choice of all of the above factors. However, the effect sizes from the multivariable models from this body of work can be used as a base to perform a power calculation to identify the sample size for a larger cohort study when more clarity is available on the various factors discussed above.

Chapter 7: Discussion

Currently the evaluation of the *Childsmile* programme involves routine monitoring of data for all four components of *Childsmile* collated electronically through the Health Informatics Centre (HIC) and the Dental Health Services Research Unit (DHSRU), University of Dundee (*Childsmile* Research and Evaluation Team (CERT), 2009). Presently no data on biological or behavioural risk factors are available at the individual child level.

The purpose of the current study was to test the feasibility of collecting data to design a larger cohort study in future. This prospective larger cohort study will collect data at individual level incorporating behavioural and biological risk factors which may be linked to information available at community level to inform, direct and evaluate future *Childsmile* interventions to improve oral health and reduce oral health inequalities in Scotland.

7.1 Key Findings

This is one of the first contemporary studies in the UK to have piloted the collection of longitudinal data on biological, behavioural, psychosocial and socioeconomic factors (simultaneously using validated measures) to identify risk factors for dental caries in young children, using a theoretical causal model. The results of this pilot study demonstrated that it was feasible to recruit a large number of preschool children and their parents from the most deprived areas of Glasgow via nursery schools and collect clinical and questionnaire data over time across educational establishments. This is of relevance to other health-related research studies.

This pilot cohort study collected data that were valid, robust and analysable, and the results of the analyses were meaningful. However, because of the pilot nature of the study, interpretation of the findings may need to be made with caution.

The risk factors that emerged from the multivariable analyses were in the majority proximal factors. The results concur with previous finding that cariogenic bacteria at baseline is the most proximal risk factor associated with caries (Litt et al., 1995). Other proximal behavioural factors that were found to show an independent association with caries in Sweep 2 included higher NMES intake, irregular brushing habits immediately before bedtime, rinsing out toothpaste after tooth-brushing, night time bottle feeding at two years and older age at first visit to dentist. Notably, two risk factors (brushing immediately before bedtime and age at first dental visit) not featured in previous longitudinal studies were found to be independently associated with caries at Sweep 2. The only distal risk factor that emerged from the multivariable analyses was living in deprived socioeconomic circumstances.

The study confirmed the well-known association between socioeconomic circumstances and caries in young children (NDIP, 2014; Reisine and Psoter, 2001), uniquely using a range of socioeconomic measures, at individual- and area-level. As suggested by previous research, children living in the most deprived circumstances were consistently more likely to have caries experience than those from affluent backgrounds, irrespective of the socio-economic measure used (Grindefjord et al., 1995, Rodrigues and Sheiham, 2000, Reisine and Psoter, 2001).

This study is among the few prospective longitudinal studies that attempted to explain the socioeconomic gradient in caries in young children (Slade et al., 2006; Gao et al., 2010) using a range of indicators of socioeconomic position (SEP). The persisting socioeconomic gradient in caries experience after adjustment for relevant risk factors, using a comprehensive set of SEP measures, suggests the complexity in the determinants of these inequalities and possibly the independent influence of SEP on caries (Gao et al., 2010). The results support the findings of Slade et al. (2006) who attempted to explain socioeconomic inequalities in young children using a case-control study design.

The area-based measure of SEP, Scottish Index of Multiple Deprivation (SIMD) was the strongest and the most stable indicator of inequality in caries prevalence whereas the gradient for gross household income was attenuated by behavioural risk factors. Consistent with the findings of a previous studies, in young children (Slade et al., 2006), adolescents (Mashoto et al., 2010; Jung et al., 2011) and adults (Sanders et al., 2006; Donaldson et al., 2008) the socioeconomic gradient in caries was not fully explained after adjusting for relevant risk factors. This may imply a direct causal effect of SEP. However, residual confounding from risk factors underestimated and unaccounted cannot be ruled out.

The findings from this pilot study will be used to design a larger birth cohort study to robustly investigate modifiable risk factors of caries, identify 'causes of the causes' (Marmot, 2010) and design and test potential interventions to tackle and reduce inequalities.

7.2 Strengths

7.2.1 Study design

Systematic reviews synthesising evidence on risk factors associated with caries in young children have previously highlighted a paucity of studies using the ideal study design (longitudinal) (Harris et al., 2004, Parisotto et al., 2010, Hooley et al., 2012). The longitudinal design of this study, although pilot in nature enabled tracking changes over time, establishing putative risk factors previously identified in cross-sectional studies and suggesting the direction of causal association (Song and Chung, 2010).

7.2.2 Sample size

Whilst the sample size for feasibility or pilot studies may depend on the variability in the key parameters, a sample size of 30 to 50 participants (per arm in case of a randomised pilot) are often chosen, and considered sufficient (Hertzog, 2008, Chief Scientist Office, 2014). The sample size for this pilot study was relatively large and on a par with many published full-scale longitudinal studies (Karjalainen et al., 2001, Warren et al., 2009). It therefore permitted the investigation of associations between variables (with adequate caution) and generated meaningful results.

7.2.3 Participation rate

Previous studies show parents, teachers and children from more deprived areas to be substantially less likely to participate in population-based studies (Galea and Tracy, 2007; Goodman and Gatward, 2008) with participation rates eight to nine percent lower in the most deprived areas when compared to the least deprived areas (Goodman and Gatward, 2008). Based on previous estimates, the recruitment rate for this study was as anticipated at the time of sampling (approx. 40%) (Tsakos et al., 2012). Comparatively, it was higher than that achieved in other pilot studies (Survey of sugar intake among children in Scotland, 2008, Wyse et al., 2011). A slightly higher than the target sample of 200 families was successfully recruited and the response was similar across all socioeconomic groups. Additionally, this study recruited and retained a large percentage of the traditionally ‘hard-to-reach’ families from the most deprived 20% areas. This is thought to be mainly due to the nursery school-based recruitment strategy that provided access to a large cross-section of children from deprived areas (Pescud et al., 2014, Bonevski et al., 2014); and the nursery-staff support in recruitment that might have influenced participation (Wolfenden et al., 2009).

This study had an excellent follow-up rate (87% of those with complete data in Sweep 1 were followed up) one year later compared to other longitudinal studies that followed a similar sample (in age and SEP) (Litt et al., 1995; Warren et al., 2009). Contrary to the common challenge of differential participation in many cohort studies (Mattila et al., 2000; Booker et al., 2011; Bovenski et al., 2014), there was no evidence of differential participation between Sweep 1 and Sweep 2 participants according to SEP. The children who were lost to follow-up were mainly those who had changed home addresses.

7.2.4 Methodological credibility

This study collected robust data on a range of risk factors (social, pyschological, behavioural and biological factors) using measures that were previously tested and validated in earlier studies (Adair et al., 2004; Conway et al., 2005; The Avon Longitudinal Study of Parents and Children (ALSPAC), 2007; Survey of sugar intake among children in Scotland, 2008; Scottish Executive, 2009). SEP was measured using a range of measures, both at the area- and individual-level to fully capture the construct (as far as possible), unlike many other studies where the research interest was primarily in inequalities (Slade et al., 2006; Mashoto et al., 2010; Polk et al., 2010; Jung et al., 2011). It also allowed for comparisons to be made between the various measures, through standardization of the measures (RII analyses).

Further methodological rigour was employed through training and calibration of the clinical examiner according to BASCD criteria (Pitts et al., 1997) for measuring caries experience and the Royal College of Paediatrics guidelines (RCPCH, 2013) for measuring heights and weights; and data quality control.

Limited longitudinal studies have investigated the simultaneous contribution of such a broad range of risk factors to caries, whilst considering the impact of inequalities contemporaneously. Of those that included multiple risk factors, some did not feature biological factors (Mattila et al., 2000, Rodrigues and Sheiham, 2000, Karjalainen et al., 2001, Ismail et al., 2009) and/or psychosocial factors (Wendt et al., 1996, Mattila et al., 2000, Rodrigues and Sheiham, 2000, Warren et al., 2009) and when these factors were included in the study, there was bias associated with the use of non-validated measures (Litt et al., 1995; Fontana et al., 2011). Although pilot in nature, this study has created a unique dataset, which includes multiple measures of risk factors across various levels associated with caries that had never previously been recorded among young children from Glasgow. It has provided tools, methods and preliminary data to test multiple novel hypotheses in relation to salivary immune response, childhood obesity and bacterial metagenomics that could be reproduced in a larger study.

7.2.5 Statistical modelling

Whilst multivariable statistical analyses are appropriate for studies of multiple risk factors seeking to identify risks in caries (Harris et al., 2004), a model constructed through techniques such as step-wise logistic regression have limitations. Such an approach is based purely on statistical associations (Wendt et al., 1996, Mattila et al., 2000, Wigen et al., 2011) and has limited appreciation for the theoretical basis of the inter-relationships between the multiple risk factors (Victora et al., 1997). Additionally, the conclusions from step-wise regression are generally irreplicable due to its dependence on sampling error (Victora et al., 1997). Unlike most previous longitudinal studies, the analysis in this study, in addition to using longitudinal data, was guided by a conceptual model that included social, psychological, behavioural and biological factors of caries in children (Wendt et al., 1996; Mattila et al., 2000). A decision on which factors to include in the model was balanced between statistical associations with some degree of theoretical interpretation, based on the hypothetical model describing the relationships between risk factors (Victora et al., 1997).

The use of measures for quantifying socioeconomic inequalities have somewhat been limited in caries epidemiology (Blair et al., 2013, Do et al., 2010). The present study used the Relative Index of Inequality (RII) to compare inequalities across different SEP measures and to explain socioeconomic inequalities in caries. The use of RII ensured that the inequality estimates were less influenced by the extremes of the socioeconomic spectrum (Shaw et al., 2007), as it considered the average distribution of caries across the socioeconomic strata as opposed to measures such as range, rate ratios, rate differences or odds ratios (Regidor, 2004). To the author's knowledge, no previous studies have attempted to explain socioeconomic inequalities in caries using this methodology (Sanders et al., 2006; Slade et al., 2006; Donaldson et al., 2008; Mashoto et al., 2010; Polk et al., 2010; Jung et al., 2011).

7.2.6 Multidisciplinary approach

This study had the advantage of being supported by a multidisciplinary team, who also lead on the strategic evaluation of the *Childsmile* programme. The evaluation has a number of research components including (but not limited to) behaviour change, impact on health services and health outcomes (oral and general) (*Childsmile* Research and Evaluation Team (CERT), 2009). Experiences and data collected from this study will be used to design (and power) a larger cohort study that will inform and disentangle the relative contribution of *Childsmile* interventions to oral health improvement; and reduce dental health inequalities in Scotland.

7.3 Limitations

7.3.1 Sample size and Characteristics

This was a feasibility study and the main purpose of the study was to pilot tools, test methodologies and consider sample size requirements for a future larger cohort study. This meant that the study was not adequately powered to detect subtle effects; estimates would be less precise and likely to be biased- although exact methods of calculating confidence intervals were employed to mitigate this (Burton et al., 1998).

Children from more deprived areas (SIMD) were intentionally oversampled to account for an expected lower participation rate (Goodman and Gatward, 2008) which may have biased the estimates.

7.3.2 Bias associated with self-reported questionnaire data

Self-reported data is subject to potential biases of recall and social desirability (Levin, 2005). Although this is a maxim in analytical epidemiology, to ensure consistency in self-reports, a number of questions (where possible) were used in assessing one variable. Statements around parental attitudes and beliefs may have been particularly susceptible to social desirability bias. Hence, all statements used in measuring parental attitudes and beliefs adapted for this study have been assessed for consistency in previous studies (Adair et al., 2003, Skaret et al., 2008, Eckert et al., 2010). Furthermore, items measuring attitudes and beliefs related to oral hygiene and diet were evaluated for consistency by assessing the relationship between these variables and self-reported behaviours (toothbrushing and NMES consumption). Parents who reported unfavourable attitudes and beliefs were found to positively correlate with unfavourable behaviours.

Diet diaries are generally agreed to be the most precise method of dietary assessment, although they carry inherent limitations of underreporting (Wrieden et al., 2003). The semi-quantitative Food Frequency Questionnaire (FFQ) used in this study has been shown to slightly overestimate energy compared to that recorded in diet diaries. Nonetheless, this FFQ has been shown to provide valid data on micronutrient levels expressed as a percentage of energy in studies of more than 100 participants (Survey of sugar intake among children in Scotland, 2008).

7.3.3 Misclassification of teeth

The method used to measure caries experience at Sweep 1 may have introduced some misclassification. However, a single dentist who was trained and calibrated according to BASCD criteria examined all children in Sweep 2, which was the main outcome measure used in this body of work. Nonetheless, the misclassification of teeth in Sweep 2 may not be completely ruled out as intra-examiner reliability was not assessed.

7.3.4 Estimation of mutans streptococci (MS) levels from saliva

Mutans streptococci is generally agreed to colonise tooth surfaces, with plaque samples collected from the tooth surface being the most reliable method for the estimation of MS (Parisotto et al. 2010). However, the bacterial species and their numbers vary extensively based on the site in the oral cavity where plaque samples are collected and the length of time since last toothbrushing (Simon-Soro et al., 2013). Furthermore, MS have been detected in the saliva of pre-dentate infants (Wan et al., 2001). Along with the relative ease of sampling and processing, saliva has been an accepted and widely used method for the quantification of MS, although their numbers in unstimulated saliva are not representative of those in plaque samples (Dasanayake et al., 1995).

Compared to plaque samples, saliva samples have a smaller predictor value for caries development (Seki et al., 2003). Plaque samples have also consistently yielded higher numbers of MS (Seki et al., 2003, Thenische et al., 2006, Malcolm, 2013), which in turn is reported to be associated with higher caries risk (Leal and Mickenautsch, 2010). Nevertheless, systematic reviews have shown consistent evidence of an increased risk of caries among two to five year olds from studies assessing MS in saliva and plaque (Thenische et al., 2006; Leal and Mickenautsch, 2010).

The present study could not use the plaque samples collected in this study, which was reserved for later analysis of oral metabiogenome content. Unstimulated saliva was used for the quantification of MS. A previous *Childsmile* study using similar methodology (TaqMan® QPCR) has shown a positive correlation between the numbers of *S. mutans* detected in unstimulated saliva and plaque (p = < 0.0001) (Malcolm, 2013).

7.3.5 Statistical modelling

This study used a traditional regression-based approach to establish associations between risk factors and caries. This approach was limited in analysing factors in sequence of their temporal order and clarifying the operational pathways through which risk factors interacted to cause caries (Newton and Bower, 2005). Utilizing statistical methodologies such as structural equation modelling (SEM) or path analysis would permit modelling of causal pathways and simultaneous testing of interactions between a range of socioeconomic, psychosocial, behavioural and biological factors and caries (Aleksejūnienė et al., 2009). However, SEM is a large sample technique and a sample size of at least 200 is recommended for reliable results (Litt et al., 1995).

7.3.6 Data linkage

Given the strong evidence that fluoride varnish substantially reduces caries increment (Marinho et al., 2013), all data (clinical and questionnaire data) were planned to be record-linked to *Childsmile* databases that held information regarding fluoride varnish applications at *Childsmile* practice and nurseries/schools. However, the record linkage was not achieved within the limited time frame prior to data analyses. It is now planned to be included in future analyses.

7.3.7 Dietary assessment method

Weighed diet diaries are generally agreed to be the most precise method of dietary assessment, in spite of it being prone to underreporting and alterations in diet (Wrieden et al., 2003). The use of FFQs for dietary assessment in this study had some limitations (Wrieden et al., 2003). Specifically, the FFQs did not capture information about the frequency of NMES intake through a day, timing of sugar consumption, such as at night, and the number of eating/drinking events involving NMES per day that have all been shown to have an association with the development of dental caries than quantity of sugar consumed (Marshall et al, 2007; Anderson et al, 2009). Diet diaries could provide a greater depth of information about the dietary behaviour and their relationship with caries experience.

Nonetheless, FFQs were thought to be an appropriate tool for dietary assessment for the purposes of this study. In particular, compared to a diet diary it was relatively less of a burden to the participants (Wrieden et al., 2003), thus potentially not risking compliance and loss to follow-up (Booker et al., 2011). The FFQs used in this study had the advantage of being administered by multiple modes (by interviewer face-to-face, over the telephone or can be self-administered by the participant (main-carer)) (Survey of sugar intake among children, 2008). They were also relatively less expensive to be used at population level for a larger epidemiological study (Wrieden et al., 2003).

7.3.8 Community-level influences

The hypothetical pathway that drove the design, data collection and the analyses did not include community-level influences on caries (Fisher Owens et al., 2007). Hence, this study was unable to explain behaviours in the context of the wider social determinants. Although the use of area-based measure of SEP may partially reflect the physical and social environment, it gave limited information on the contextual qualities that may cause inequalities in caries. Variables preferably using qualitative research to measure social capital, community cohesion, social support and network will improve the understanding of influences on caries and the causes that actually drive socioeconomic inequalities (Newton and Bower, 2005).

7.4 Feasibility

This section discusses the results related to objectives 1(i) to (iv).

The terms ‘feasibility’ and ‘pilot’ are often used interchangeably. According to the National Institute for Health Research (NIHR) Evaluation, Trials and Studies

Coordinating Centre, feasibility studies are ‘pieces of research’ conducted to test specific aspects of the design of the proposed main study, and pilot studies seek to test whether all of the components of the proposed main study work together (Chief Scientist Office, 2014). The Medical Research Council (MRC) guidance for developing complex interventions explicitly recommends the use of a series of feasibility and pilot studies targeted at each of the uncertainties in the study design to avoid problems associated with sample size, recruitment and retention, acceptability and compliance (Craig et al., 2008).

Longitudinal cohort studies are important for understanding aetiological mechanisms underlying disease and associated inequalities. However, they carry inherent limitations of logistical issues of expense and time, particularly in the case of prospective designs and following up of high-risk groups (Song and Chung, 2010). Furthermore, they may be prone to bias associated with loss to follow-up (selection bias), threatening the internal validity of the study (Song and Chung, 2010). Piloting can assess feasibility, acceptance and compliance for continued participation, if measures are valid and repeatable, time and budget problems and potential personnel and data management issues in a miniature version of the proposed main study (van Teijlingen and Hundley, 2001). This will in turn ensure that valuable resources are used effectively. Large-scale population-based cohort studies like the Avon Longitudinal Study of Parents and Children (ALSPAC) was the result of five years of planning and piloting (Golding, 1990).

Within the present pilot study, the following aspects of a longitudinal cohort design was assessed:

- Recruitment
- Response
    i. Clinical data
    ii. Questionnaire data
- Quality of data collected
- Follow-up

7.4.1 Recruitment

Most large-scale birth cohort studies like the National Survey of Health and Development (NSHD), 1970 British Cohort Study (1970), the ALSPAC (Golding, 1990) and the Born in Bradford Study (2008) have utilised the support of local community health professionals like midwives or nurses to enrol eligible families as these studies began early in pregnancy or at birth. Other studies like the Millennium Cohort Study (MCS) (Plewis and Ketende, 2006) and the Growing Up in Scotland (GUS) (Bradshaw et al., 2008) study recruited the sample as a cross-sectional survey (Survey of sugar intake among children in Scotland, 2008) via the Department of Work and Pensions sent opt-out letters, when children were a few months old. The present pilot cohort study targeted a sample of preschool aged children, and hence the sample was drawn from the population of nursery schools previously included in the NDIP. As discussed previously, the NDIP inspected a representative sample of three-year-olds in 2009/10, accounting for 20% of the three-year-old population (McMahon et al., 2010; McMahon et al., 2011). The methods used for recruitment via nursery schools, adopted from Tsakos et al. (2012) proved to be an effective route for identifying eligible families for a study of this nature. Although the nurseries were used to participating in the annual NDIP (McMahon et al., 2010), this was a considerable undertaking for the nursery schools in Sweep 1 as engagement of educational establishment staff was critical and their time was an important factor.

Low recruitment rates are a barrier to conducting population-based studies (Galea and Tracy, 2007; Booker et al., 2011), particularly when more deprived groups of the community are involved (Bovenski et al., 2014). The present study anticipated a low recruitment rate and approached a sufficient number of eligible participants weighted towards more deprived areas to ensure that the target sample size was reached, a strategy supported by Galea and Tracy (2007). Comparatively, the recruitment rate was relatively higher than those achieved for other pilot studies (Survey of sugar intake among children in Scotland, 2008, Wyse et al., 2011),

Whilst a large number of studies have reported recruitment of lower SEP groups into research as highly challenging (Galea and Tracy, 2007; Goodman and Gatward, 2008; Bonevski et al., 2014), this study found school-based recruitment as an effective strategy in recruiting the traditionally ‘hard to reach’ group. This echoed the findings of Pescud et al. (2014), who specifically recommended schools as an effective platform for accessing and recruiting lower SEP families into health-based research. Additionally, as evidenced from a narrative synthesis of non-randomised studies to identify strategies to increase recruitment in school-based research, the present study used multiple scheduled face-to-face meetings with the principal contact at schools to build and maintain relationships and engage the staff with the study. These staff members have been found to be highly influential in parental decisions about participation in research (Wolfenden et al., 2009). The opt-out (passive consent) procedure has been found to be an effective strategy for yielding a higher participation rate, particularly among more deprived and ethnic minorities in many national cohort studies (Plewis and Ketende, 2006). Only seven and two percentages have opted out of the MCS (Plewis and Ketende, 2006) and the Growing Up in Scotland (GUS) study (Bradshaw et al., 2008) respectively. Nonetheless, a systematic review of the literature to identify effective strategies for the recruitment of ‘hard-to-reach’ groups have found mixed evidence for the effectiveness of the numerous strategies that have been reported in the literature. The authors suggest a ‘comprehensive and multipronged approach’ based on the study population and highlight the importance of involving community groups and organizations in improving participation and retention (Bonevski et al., 2014).

As noted in the pilot Survey of Sugar intake study (2008), the researcher or the head teacher speaking to the parents in advance of distributing the information sheets did not improve the recritment rate compared with nurseries where no such methods were used. However, the presence of a designated oral health promoting staff member at one of the nurseries was shown to considerably improve the response. Notably, this nursery was located in the most deprived area and had limited facilities at the time to accommodate data collection. Nevertheless, the staff in-charge at the nursery was extremely co-operative, prompt in all communications and highly experienced with children and parents (40 years in experience as a nursery-staff). This suggests the critical role of a championing individual in obtaining good response (Wolfenden et al., 2009; Boveski et al., 2014)

It is not known if the recruitment rate obtained in this study could have been further improved. It is speculated that there may have been some non-responders due to the increased number of assessments and in particular the collection of biological samples, which has been reported previously to potentially yield relatively lower participation rates (Galea and Tracy, 2007). Additionally, feedback obtained from the nursery-staff highlighted the increased number of studies/inspection programmes carried out via nursery schools (For example: Vision screening programme, The strength & difficulties study, NDIP, *Childsmile* fluoride varnish programme etc). The increased number of requests to consent/opt-out from various programmes throughout the year might also have had an effect on the recruitment rate of this study (due to cumulative intrusion in personal lives) (Galea and Tracy, 2007).

7.4.2 Response rate

Clinical: Once participating, the response rate[59] to clinical data collection was high (87% in Sweep 1, 80% in Sweep 2) and reflected the utility of an educational establishment setting for data collection and the flexibility of the researcher to collect data using varying routes and schedules. These strategies have been supported in a systematic review of the literature (Bovenski et al., 2014) and successfully used in the Determinants of young Adult Social well-being and Health (DASH) study- a large longitudinal study of 'hard to reach' school children that examines the influence of socioeconomic circumstances on a range of health outcomes (Maxwell et al., 2012).

Questionnaire: The mode of questionnaire administration has been shown to affect response rate and data quality (Bowling, 2005). A large majority of large-scale UK studies use face-to-face interviews to collect questionnaire data as it has shown to yield higher response rates and better quality of data collected. However, the time and costs involved in training interviewers may be

[59] Response rate is defined as the number of those who completed the clinical examination/questionnaire divided by the total who consented (Last, 2001).

Last JM (Ed.). A Dictionary of epidemiology. 4th edition. 2001, New York, Oxford University Press considerable. Moreover, it is relatively more prone to social desirability bias than self-administered questionnaires (Bowling, 2005).

The MCS has used a mixed mode of questionnaire administration (face-to-face and self-administered) depending on the variable of interest to mitigate the limitations of each mode. However, the ALSPAC have successfully used self-administered mode of questionnaire completion with good response rates of 82% and 80% in the first two years of its running (Golding, 1990). The response rate for questionnaires in this study was high (82% in Sweep 1) and comparable to those achieved in the ALSPAC, despite primarily comprising of families from deprived areas. The strategies that may have contributed to this high completion rate are multiple and were informed by a Cochrane Collaboration review namely: offering incentives, offering additional copies of questionnaires and inclusion of free-post envelope, timely reminders by educational-staff and researcher telephone follow-up (Edwards et al, 2007).

However, two items on the questionnaire that were poorly completed were questions related to household income and receipt of benefits. These questions are sensitive in nature and have been previously reported to yield item non-response in other studies (Bowling, 2005) and generally reported more in self-administered questionnaires than with face-to-face interviews (Bowling, 2005).

7.4.3 Quality of data collected

The results demonstrated the feasibility of collecting (clinical and questionnaire) data via nurseries/schools. The nursery schools can provide access to a large cross-section of children from across the socioeconomic spectrum and a good setting to engage ‘hard-to-reach’ groups and follow-up one year later.

Microbiology: Distributions of microbiological and immunological data were as expected and in line with published norms (Leal and Mickenautsch, 2010; Malcolm, 2013). However, limitations associated with the use of unstimulated saliva compared with other methods for the estimation of MS has been discussed under ***Section 7.3.4***.

The ALSPAC, NCDS and MCS in the UK have included self-administered saliva sample collection from older children and adults for DNA extraction, which is posted back by respondents. However, there have been limited large-scale cohort studies that have collected saliva sample of preschool aged children (Bartington et al., 2009). The MCS recently showed the feasibility and acceptability of adopting self-administered saliva sample collection to test for infections under the guidance of trained interviewers in a large cohort of preschool children (Bartington et al., 2009). Whilst the response to saliva collection was good (81%), families of ethnic minority and deprived backgrounds were less likely to provide a sample despite the provision of translated study information sheets and the help of family members or professional interpreter support. Although the study did not report on who declined to provide a saliva sample (mother, child or both), several other studies report significant participant non-compliance among young children (two to five-year-olds) specifically (Kaitz et al., 2012). Contrastingly, this study found no difficulty in collecting saliva using the Salimetrics Children's Swab (SCS). The 100% compliance rate in saliva and plaque collection demonstrates the acceptability of the methods used to both nursery-staff and the children themselves.

Although the SCS was used to absorb saliva from the mouth of every participant for up to 60 seconds, there were insufficient volumes of saliva for some assays. This might reflect the variation in salivary flow rate among the participants, which in turn has been related to oral clearance rate, pH buffering capacity and neutralization of acid production to maintain tooth integrity (Vadiakas, 2008). Future studies may need to make more careful decisions on prospective salivary assays to perform, approximate volumes that may be required and in turn a longer collection time of up to a maximum of 90 seconds (Kudielka et al., 2003). Although this longer saliva collection time has previously been used in some studies of preschool children (Kudielka et al., 2003), it was thought in this study that more than 60 seconds of saliva collection was demanding of a young child. Multiple collections or self-administered saliva sample collection which is mailed back are good alternatives that could be considered (Bartington et al., 2009).

Clinical data: The distribution of clinical data (caries and anthropometric) were as expected and showed similar trends to those published previously (Macpherson et al., 2012; ISD Scotland, 2013a; NDIP 2014).

The simultaneous collection of heights, weights, microbiological samples and caries data in an educational setting demonstrated the feasibility of conducting population-based studies in this setting over time. The potential for collecting these data simultaneously via the NDIP or CHSP is considerable into the future. This will address the common risk factor (CRF) approach recommended to address shared risk factors associated with various chronic health conditions like obesity and oral diseases (Watt and Sheiham, 2012). The prevalence of these chronic health conditions is highest among the most deprived and adopting the CRF approach has potential for reducing inequalities and increasing effectiveness and efficiency of these routine inspection programmes (Watt and Sheiham, 2012).

Diet: As discussed previously, weighed diet diaries are generally agreed to be the most precise method in dietary assessment. Although not the most preferred method for large-scale studies due to the costs associated with data preparation and analysis, the use of diet diaries in the ALSPAC for 10% of the child sample demonstrates their possible use on a large-scale (Emmett, 2004).

The present study found FFQs to be feasible and appropriate for this age group of children as diet is usually stable and established by three years of age (Emmett, 2004). The distributions of nutrients obtained using the FFQ were in line with those obtained in the Survey of sugar intake among children in Scotland (Masson et al., 2010). The results illustrate that the FFQs were detailed and appropriate to identify relationships between diet (NMES as a percentage of food energy) and caries. This concurs with the findings of Masson et al. (2010).

There remains a possibility that the levels of NMES intake recorded using the FFQ may have been influenced by the time of the year when the study was run. The FFQs were sent out in the early summer (June), which resulted in recording of ice-lollies, ice creams and other similar foods, and drinks that are preferred to be consumed over warmer summer months. However, the FFQ assessed habitual food intake over three months which means food intake over the months of April and May were included in the recording. It is thought that the combined effects of food consumed over the cooler months and one warmer month working in opposing directions would have given a true representation of the diet.

The Scottish Collaborative Group has recently developed an electronic version of the FFQ among adults. The electronic version showed good agreement and repeatability with the paper version, and acceptability among the participants (Kyle J et al., 2012). Other studies from the USA have developed interviewer administered web-based FFQs with audio questions and touch screen photo responses incorporated for use among low literacy groups (Zoellner et al., 2005). While these technology-assisted FFQs make administering and subsequent data entry more cost and time efficient for use in large-scale (Zoellner et al., 2005), further work is warranted for their use among young children.

About your study child and About your family questionnaires: There was sufficient variability in the questionnaire responses. The relationship between the variables and caries were as expected.

Among all the items that were used to measure parental attitudes and beliefs, '***If my child uses fluoride toothpaste, it will prevent tooth decay'*** was the item that showed the most notable variation in responses. However, this item was not used in building scales as it showed poor inter-item and item to scale correlations for this sample, and in turn reduced the Cronbach's alpha of the scale. While the original study (Adair et al., 2004) from where the items were adapted used this item in building a scale, the internal reliability for the scale was poor (Cronbach's alpha: 0.52).

In contrast to the present study and Adair et al. (2004), a longitudinal Norwegian study of three-year-olds used this item to build a scale with good internal reliability (Cronbach's alpha >0.70) (Skeie et al., 2010). They combined the items into slightly different scales to the original authors (Adair et al., 2004) and the present study. The scales were built by selecting items that were previously shown to have predictive validity in the Norwegian population (Skaret et al., 2008). Although this was not possible at the start of the present study, it has now provided with sufficient data (longitudinal) to possibly refine the scales based on predictive validity for future use.

7.4.4 Follow-up

The short period of follow-up in longitudinal studies has been suggested for the potential lack of association between some risk factors and caries (Burt and Pai, 2001). A length of three years has been recommended for longitudinal studies on dental caries of permanent teeth (Rugg-Gunn et al., 1984). However, this recommendation may be less relevant for caries in primary teeth as the progression of caries in primary teeth is more rapid than in permanent teeth (Ismail et al., 2009), therefore a shorter follow-up period of one year for assessing changes was considered acceptable (Rodrigues and Sheiham, 2000, Anderson et al., 2009).

A high retention rate is vital to prevent bias and ensure the internal validity of the study. Large-scale birth cohorts like the Danish National Birth Cohort have achieved high retention rates of 92% in the 5th year of the study (Olsen et al., 2001). Recent birth cohorts in the UK, like the ALSPAC and the MCS have shown relatively good levels of retention of approximately 75% over the first seven years of the study (Plewis and Ketende, 2006, Golding, 2004). These high retention rates have been reported to be primarily due to effective communication between the fieldworker and participants and publicity of the study results (Olsen et al., 2001; Golding et al., 2004).

The present pilot study had a successful follow up (87% of those with complete data in Sweep 1) of what are conventionally thought to be ‘hard to reach’ groups. The strategies that may have contributed to this high retention rate are multiple and were informed by a systematic review of retention strategies in population-based cohort studies namely: offering incentives and using alternative modes of data collection (Booker et al, 2011). Additionally the present study found that fostering a positive relationship with families (when possible over phone calls and face-to-face data collection) and flexible schedules for data collection to be crucial for ensuring high retention rates, a strategy supported by the UK-based DASH study (Maxwell et al., 2012) and a systematic review of the literature to engage and retain 'hard-to-reach' groups (Bovenski et al., 2014).

7.5 Dental caries experience

The prevalence of caries experience at both time points was higher than those reported in nationally representative epidemiological surveys (McMahon et al., 2011, Macpherson et al., 2012; NDIP 2014). This was not unexpected as the children in this study were on average a year older in Sweep 1 and lived in more deprived areas of Glasgow city compared to those in the NDIP. While the figures of the NDIP are not directly comparable with the results of this study, caries experience reported in the most recent NDIP are discussed below for context.

Nineteen percent of children were reported to have caries experience in a representative sample of three-year-olds in the Greater Glasgow and Clyde Health Board through the NDIP in the preceding year (2009/10) to data collection for the present study (McMahon et al., 2011). This is the first study to report caries experience among preschool children (as opposed to ante-preschool children) in Glasgow city (Sweep 1) and the proportion of children with caries experience in Sweep 1 was 35.8%.

The prevalence of caries experience (47.7%) and mean $d_3$mft score (1.9) in Sweep 2 was higher than those reported in the recent 2014 NDIP for Glasgow City Community Health Partnership (CHP). The proportion of children showing caries for Glasgow city CHP was 41.2%, with mean $d_3$mft score of 1.8 in the 2013/14 NDIP (2014). The higher proportion of caries in this body of work could be explained by the sampling method that was used in this study, i.e. over-sampled children from deprived areas to ensure a reasonable distribution of SEP and prevalence of caries.

7.6 Risk factor models for caries

The following sections discuss the results in relation to objectives 3 (i) and (ii)

As discussed in ***Section 7.3.1***, statistical testing of results from a pilot study may be considered inappropriate due to the limitations associated with sample size and the generalizability of results. The sample size in this pilot study was adequate to produce statistical models. However, caution should be exercised when interpreting results.

In the following sections, when discussing the results of the analyses comparisons are made with results from other relevant studies and surveys. However, such comparisons are difficult, and in some cases potentially unhelpful due to the difference in population characteristics, particularly socioeconomic circumstances, sampling methodologies, sample sizes and response rates. The comparisons will therefore be made primarily for context rather than to draw inferences about the sample in relation to other populations.

7.6.1 Risk factors associated with caries in young children

The following section discusses the results in relation to objective 3 (i).

The hypothetical model outlined previously in Figure 4.2 integrated factors operating at various levels to understand the complex aetiology of caries in young children. Although the hypothetical framework was used to guide the analyses, all the risk factors (except one) that emerged from the multivariable analyses were largely proximal factors, with more distal factors dropping out.

Overall, the results demonstrated a combination of socioeconomic, behavioural and biological factors at baseline to be associated with caries a year later. The results from this body of work are in line with associations previously reported in the literature (Harris et al., 2004); some identified in previous cohort studies reviewed in ***Section 2.4.2*** (Grindefjord et al., 1996; Wendt et al., 1996; Rodrigues and Sheiham, 2000; Radford et al., 2000) and some from cross-sectional studies (Al Ghanim et al., 1998; Shiboski et al., 2003; Willems et al., 2005).

The results interpreted in the light of the conceptual model demonstrate that cariogenic bacteria (levels of salivary *S.mutans)* are the immediate proximal risk factor associated with caries. This is in agreement with previous findings from other cohort studies that found higher levels of *S. mutans* in saliva to be most closely associated with caries in young children (Litt et al., 1995; Thenisch et al., 2006). However, a direct causal temporal relationship between *S. mutans* and dental caries has not been established. This is partly due to the complexity of oral microbial ecology, with mutans streptococci also present in healthy mouths (Wan et al., 2001) and sometimes absent inspite of having caries experience (Becker et al., 2002; Aas et al., 2008). Although pilot in nature, further analysis grouping children according to no caries experience at both Sweeps, caries experience at only Sweep 2 and caries experience at both Sweeps may help establish a temporal relationship between the levels of *S. mutans* and caries experience.

In addition to the levels of *S.mutans*, there was evidence of five oral health related behavioural risk factors (higher amounts of NMES intake, rinsing out toothpaste after tooth-brushing, irregular brushing habits before bedtime, night time bottle feeding at two years and older age at first visit to dentist) to be independently associated with caries.

The study revealed greater risk of caries with higher intakes of NMES as a percentage of food energy, independent of other risk factors. Additionally, fewer children adhered with the Department of Health recommendations of no more than 11% of food energy to be derived from NMES (percentage of food energy). These findings are consistent with those reported by the Survey of Sugar intake among children in Scotland (2008). However, the present study had the advantage of using longitudinal data and objectively measuring dental caries using the standardised BASCD criteria (Pitts et al., 1997) compared to the aforementioned survey. A recent systematic review showed a clear relationship between sugar intake (NMES) and caries, with moderate evidence that caries was higher when NMES intake was above 10% of food energy (Moynihan and Kelly, 2014). This has further been supported by an ecological study that showed a log-linear relationship between caries and sugar intake, when sugar intake increased from 0% to 10% of daily food energy. (Sheiham and James, 2014). This evidence has led to calls for the updating of the WHO sugar consumption guidelines to less than five percent of food energy to minimize the risk of dental caries and other chronic diseases, including obesity and diabetes throughout the life-course (Sheiham and James, 2014).

The model also featured two oral hygiene practices, rinsing out toothpaste after toothbrushing and irregular bedtime brushing habits at baseline to be independently associated with caries at Sweep 2. The SIGN guidelines (2014) recommend that toothbrushing is carried out last thing at night before bed and at least once during the day. Brushing immediately before bedtime keeps fluoride levels high while salivary flow (and in turn the protective effects of saliva) drops during sleep (SIGN, 2014). However, there have not been many studies carried out in young children that has considered, irregular bedtime brushing habits as a risk factor in caries. A thorough search of the literature only found one poorly conducted Taiwanese case-control study to have ever considered irregular bedtime brushing habit as a risk factor for caries in young children (Tsai et al., 2006). The risk factor, rinsing after toothbrushing although not featured in previous cohort studies, has previously been shown to reduce the caries-preventive effect of fluoride toothpastes in trials of young Swedish children (four-year-olds) and Scottish older children (mean age 12.5) (Chestnutt et al., 1998, Sjögren et al., 1995).

The influence of bottle feeding on caries in young children remains somewhat debated, as bottle use has been equally common among young children without caries (Gussy et al., 2006). The evidence has been inconsistent and primarily based on cross-sectional studies (Al Ghanim et al., 1998; Shiboski et al., 2003). Although comparability of bottle feeding related factors are difficult due to the variation in the different measures used across studies, most cohort studies did not have variables related to bottle feeding in the multivariable analyses (Grindefjord et al., 1995, Grindefjord et al., 1996, Fontana et al., 2011, Warren et al., 2009). In contrast, this study showed night time bottle feeding at two years of age to be independently associated with caries experience. In the present study 17% of the children were still being bottle fed at four years of age. However, no association was evident between night time bottle feeding at four years and caries experience at Sweep 2, as shown in previous studies (Rodrigues and Sheiham, 2000). A critical period for caries development associated with bottle use have been proposed by Reisine and Douglas (1995) which may be before the age of four years. Litt et al. (1995) found that the use of the bottle at night time was associated with sugar intake more than the use of the bottle itself. The mothers who reported night time bottle use were also more likely to have children with a higher sugar intake which in turn was associated with caries (Litt et al., 1995).

The finding that older age at first dental visit was associated with caries concurs with the findings of the Survey of Sugar intake among children in Scotland that showed children who were reported to be treated for decay were more likely to have been older when they first attended the dentist and attended the dentist for the first time because of trouble with their teeth (Masson et al., 2010). Similar findings were reported in a large retrospective cohort study in North Carolina that showed children (with public dental insurance) who had their first anticipatory dental visit by one year of age more likely to have preventive visits in future than restorative/emergency visits (Savage et al., 2004). It is not known what factors explain a child to remain caries free after early anticipatory dental attendence. It is partly likely that remaining caries free may be the effect of oral health anticipatory guidance received by parents (Savage et al., 2004). There also remains a possibility that parents who are well motivated to provide the best possible dental care for their children may be the ones who attend an early anticipatory dental appointment (Savage et al., 2004).

Finally, the results of the multivariable analyses confirmed the well-known association between socioeconomic circumstances and caries in young children (Reisine & Poster, 2001; Thomson and Mackay, 2004; Gao et al., 2010; McMahon et al., 2011; NDIP 2014). While almost all socioeconomic measures were strongly associated with caries in the univariable analysis (except for years of main-carer's education after secondary school), after adjusting for all socioeconomic factors, only national SIMD remained independently associated with caries (multivariable analyses within domains- Appendix L). This suggests that area- and individual-level measures of socioeconomic position were correlated (Braveman et al., 2005). It further indicates that area-level measures may provide information on socioeconomic circumstances not captured by individual-level measures as individuals live in communities where the social environment of the area may influence health over and above that of individual SEP. Additionally, national SIMD was found to be independently associated with caries at Sweep 2 independent of a range of more proximal behavioural risk factors and *S.mutans* counts. This concurs with the findings of other studies in young children that found increased neighbourhood deprivation as an independent risk factor for caries over and above the individual-level SEP and behavioural measures (Willems et al., 2005, Broomhead et al., 2014). Similar findings have also been shown previously among Scottish infants, with children residing in areas of highest deprivation (DEPCAT) reported to have significantly more caries than their affluent peers irrespective of mutans streptococci levels (Radford et al., 2000). Broomhead et al. (2014) recently showed 60% of the variation in mean $d_3$mft among five-year-olds to be explained by the 2010 Index of Multiple Deprivation (Broomhead et al., 2014).

The neighbourhood effect of increased risk is suggested to reflect the social environment e.g. reduced access to health promoting shops and services, environments conducive to fear, psychosocial stress and anxiety, lack of supportive social networks, low social capital and community cohesion (Willems et al., 2005; Sisson, 2007). These 'stressors' in an area may collectively interplay to increase risk of caries. While deprived individuals residing in relatively affluent areas have been shown to have the impact of low income compensated, no impact on dental health was observed when Australian affluent adults lived in relatively deprived areas (Sanders et al., 2006).

This body of work found no independent association between parental attitudes and beliefs at baseline and caries at follow-up, although this association was evident in the cross-sectional multivariable analysis (Appendix H) and longitudinal univariable analysis. It is not clear if utilizing advanced statistical methodologies such as structural equation modelling (SEM) or path analysis would have produced different results. Previous studies among young children from South-east Asia (Gao et al., 2010, Qiu et al., 2014) and the USA (Litt et al., 1995) using SEM have shown parental attitudes to be indirectly linked to caries through behaviours as per the Theory of Planned Behaviour. As discussed earlier in ***Section 7.3.5*** the regression-based approach used in this study lacked potential to estimate the risk factors in the sequence of their operational order and treated distal and proximal risk factors equally distant from outcome. The interlinking effect of parental attitudes and beliefs may have been dismissed.

7.6.2 Socioeconomic gradient in caries experience

The following section discusses the results in relation to objective 3 (ii).

The study confirmed the well-known association between socioeconomic circumstances and caries in young children (Reisine and Psoter, 2001; NDIP, 2014), uniquely using a range of socioeconomic measures, at individual- and area-level. There was a strong socioeconomic gradient in caries experience at both Sweeps, with children living in the most deprived socioeconomic circumstances being the most affected. Large differences in caries experience were evident between the most deprived and the least deprived, with 58% of those from the most deprived SIMD quintile having caries experience compared to 29% in the least two deprived SIMD quintiles (SIMD Q4 and Q5). Although not directly comparable, similar trends were reported in the recent NDIP (2014) that showed 47% of the five-year-olds from the most deprived quintile to have caries experience compared with 17% of those in the least deprived areas (SIMD Q5).

7.6.3 Explaining the socioeconomic gradient

As reviewed in ***Section 2.3.6***, different theories have offered varying explanations for the observed socioeconomic inequalities in oral health (Sisson 2007). According to these, inequalities arise because of unfavourable material circumstances, health-debilitating behaviours or due to various psychosocial factors. However, there is limited evidence in the dental literature examining how different behavioural, psychosocial and material factors together influence oral health inequalities in young children (Sisson, 2007).

The results of this study found that a range of behavioural risk factors found to be prospectively associated with caries experience did not fully explain higher prevalence of caries among children from deprived socioeconomic circumstances. The findings are consistent with Slade et al. (2006), who investigated if behavioural risk factors explained the socioeconomic inequality among Australian five-year-olds. However, Slade et al. (2006) used a case-control study design which is not the ideal design for studies of a chronic prevalent disease like caries. It proves difficult to assess the confounding effects of factors other than SEP and is prone to bias of case selection. The present study had an advantage of using longitudinal data and assessing the effect of a wide variety of risk factors on socioeconomic inequalities in caries compared with the aforementioned study. Additionally, the present study assessed the effect of diet objectively using a validated measure and SEP was measured in several different ways, both at the individual- and area-levels.

In the recent past, several other studies have attempted to identify the factors that explain oral health inequality in adolescents (Polk et al., 2010, Mashoto et al., 2010, Jung et al., 2011)) and adults (Sanders et al., 2006; Donaldson et al., 2008), with the majority investigating the role of oral health related behaviours and few, the role of psychosocial factors (Jung et al., 2011). Overall, all of the studies so far have been cross-sectional in design and have used a limited number of SEP indicators (typically, education and income), thereby limiting any comprehensive analysis on the relationship between SEP and oral health. However, the findings of the majority of the studies in spite of using varying dental health outcomes (self-reported, subjective, objective), SEP indicators and conducted in differing populations, are consistent with the findings of this study that socioeconomic inequalities were only partially explained by behavioural factors.

All measures of SEP demonstrated a substantial gradient with caries. However, it was difficult from the raw data to determine which SEP measure was the strongest measure of inequality in this sample. The Relative Index of Inequality (RII) was used to standardize the distribution of the SEP variables to allow comparison across measures (Shaw et al., 2007). While this approach has commonly been used to explain socioeconomic inequalities in general health (Singh-Manoux et al., 2004; Batty et al., 2006), no previous studies trying to explain socioeconomic inequalities in caries in young children have used this approach. Its use in caries epidemiology has been limited and more recently used to quantify and evaluate the trends over time in socioeconomic inequality in caries in young children (Do et al., 2010; Blair et al., 2013).

Overall, the results of the RII analyses showed higher risk of caries in the most deprived group relative to the least deprived, across SEP measures. The RIIs were relatively high compared to studies of the general health literature (Singh-Manoux et al., 2004; Batty et al., 2006). The extent of socioeconomic inequalities in caries varied by the SEP measure used (Sanders et al., 2006a), with national SIMD being the strongest indicator of inequality in caries in this sample. The variation in RIIs across SEP measures when adjusting for different groups of risk factors suggested that the various measures of SEP used in this study are tapping different mechanisms that contribute to socioeconomic inequalities in caries (Sanders et al., 2006a).

Area-based measures of SEP are increasingly being used to measure effects of SEP on various health outcomes, target resources and plan services (Sanders et al., 2006b; Brewster et al., 2013; NDIP, 2014). The results of this study suggest that SIMD may be a convenient measure of SEP that may be stable and effective in revealing socioeconomic inequalities in caries in young children. These findings are in agreement with previous studies that used both area-level as well as individual-based measures of SEP and found that the inequalities in caries were steeper when area-based measures were used (Locker, 1993, Thomson and Mackay, 2004, Locker, 2000).

It is suggested that when SEP is considered as the primary variable of interest, investigating the effect of both, individual as well as area-level socioeconomic circumstances is important as different measures of SEP represent different aspects of social position, thus relating and contributing differently to health inequalities (Braveman et al., 2005). SIMD used in this study was a composite measure and thus reflected a number of different aspects of socioeconomic circumstances ***(Section 2.3.2.2)***. It supplemented the individual-level measures, by including the contribution of contextual attributes as a determinant of inequalities (Broomhead et al., 2014). In this study, a higher percentage of participants were classified using the SIMD than other measures like income and occupation, thereby minimizing the degree of bias due to missing information (Locker et al, 2000). Measuring SEP by SIMD will also have the benefit of tracking inequalities through time and by location (Blair et al., 2013), possibly mapping inequalities, which can then be used to inform intervention/policy. Nonetheless, it might be informative for the purpose of designing interventions to know the specific characteristics associated with those living in a deprived area that might underlie inequalities.

The study showed that individual behaviours were important risk factors of caries, but did not fully explain the relationship between SEP and caries. The risk factor that was most associated with inequalities across the SEP measures was age at first dental visit. Inequalities indicated by all measures of SEP (except gross household income) increased when adjusted for age at first dental visit. The findings imply that first dental attendance for anticipatory care may be disproportionately less among the lower SEP groups. Dental registrations of infants up to two-year-olds in Scotland have remained low at 47% (ISD Scotland, 2013b) and failure to attend rates have been found to be greater in areas of high deprivation (Deas et al., 2010). Consistent with the findings of this study, dental attendance pattern was found to be a factor contributing to the socioeconomic gradient in dental health among Singaporian preschool children, with SEP affecting dental attendance both directly and indirectly (Gao et al., 2010). Through the use of structural equation modelling, the indirect effect of SEP on dental attendance was demonstrated to be through parental knowledge, attitudes and beliefs regarding the importance of anticipatory dental attendance. Additionally, positive parental attitudes, which are associated with better dental attendance (Tickle et al., 2000) and in turn better dental health have been shown to be held by higher socio-economic groups (Skeie et al., 2010, Van den Branden et al., 2012).

Similarly, adjusting for NMES intake (as a percentage of energy) was shown to increase inequality. Consistent with previous findings from Scotland, this suggests higher intake of NMES was disproportionately distributed among the lower SEP groups (Bradshaw et al., 2008, Masson et al., 2010). Although diet may partly be behaviourally influenced, it is also determined by the availability, access and affordability of health promoting foods, which are largely related to location and economic means (Sisson, 2007). This implies that reducing inequalities in caries may require developing strategies, which look beyond the proximal causes of caries to address the underlying determinants of oral health and related behaviours (Watt, 2007). This will involve addressing the social, economic and environmental causes of behaviours (Sheiham et al., 2011).

The results of this study demonstrate that while it will be good to target behaviours using behavioural interventions, they may not fully reduce socioeconomic inequalities in caries on their own (Watt, 2007). The findings also suggests the presence of other determinants of inequalities related to caries, which were either not measured adequately or unmeasured in this study. It is suggested that psychological stress (measured but not analysed in this body of work) experienced by those in low SEP due to unstable living conditions, insecurity, poor social capita, cohesion etc. may provide an explanation between low SEP and dental caries (Sisson, 2007). Evidence supports chronic stress as influencing health promoting behavioural decision making and additionally affecting the immune response (Sisson, 2007, Boyce et al., 2010). A psychobiological pathway to explain the social gradient in caries in young children has been proposed after lower SEP was found to be associated with increased levels of salivary cortisol and cariogenic bacteria (Boyce et al. 2010).

Most of the previous studies attempting to explain socioeconomic inequalities have not accounted for fluoride varnishes or fluoride exposure from sources other than toothpastes except Polk et al. (2010). However, the study was conducted among adolescents and in a country with no universal dental care plan and hence may not be applicable to Scottish young children. Scotland is unique with interventions applied universally, but in proportion to the level of deprivation. As described previously in ***Section 2.3.9.1.3***, *Childsmile* school targets 20% of the nursery and P1 children in priority schools for fluoride varnish applications. Data collection in a few of the nurseries took place soon after (one to two weeks) children had fluoride varnish applications through the *Childsmile* programme and this may have had an effect on the results of this study.

7.6.4 Tackling oral health inequalities

This study indicates that the risk factors and causes of socioeconomic inequalities in caries may have roots in early life. Given that, childhood socioeconomic circumstances are significantly related to caries experience in childhood, adolescence (Nicolau et al., 2003) and adulthood (Thomson et al., 2004), independent of adult SEP, preventive interventions that tackle the determinants of inequalities should be required to commence in early life (Scottish Government, 2008; Marmot et al., 2010).

Tackling oral health inequalities has been a key focus of health policies (Marmot et al., 2010, Marmot and Bell, 2011, Scottish Government, 2008, Sheiham et al., 2011). The effectiveness of behavioural interventions in reducing inequalities in dental caries have been questioned, when oral hygiene practices post-intervention were found to increase socioeconomic inequality among children (Schou and Wight, 1994). This individualist downstream behavioural approach to health inequalities has been criticised for being ineffective, ‘victim blaming’, over-simplifying the problem, not being cost-effective and diverting resources from more effective ‘upstream’ measures (Watt, 2007). Such an approach fails to address the wider social determinants and the environment (including economic, cultural, and political factors) in which behavioural choices are made (Sheiham et al., 2011).

According to Marmot (2005) ‘if the major determinants of health are social, so must be the remedies’. Consequently, there has been a pressing emphasise for more ‘upstream action’ – to develop preventive policies and public health strategies from a more social and structural view of the determinants of health (Watt, 2007). However, the Scottish Government’s (2008) recent policy, including the Early Years Framework suggests the need for both upstream and downstream interventions - to create an environment where health-debilitating behaviours are not formed and to help those that are victims to health-debilitating behaviours, respectively. It further underscores the need for early preventive intervention and targeted support to those in need to ensure maximized health outcomes and minimized health inequalities among children. This follows the ‘proportionate universalism’ strategy proposed for tackling health inequalities by which actions are applied universally, but with the scale and intensity matched to the level of disadvantage (Marmot et al., 2010). Since oral health inequalities mirror those in general health, it is recommended that a collaboration between oral and general health to address shared risk factors (behavioural and common social determinants) associated with various chronic health conditions (like cardiovascular diseases, obesity and oral diseases) may reduce inequalities, creating supportive environments for promoting health in the whole population, and increase effectiveness and efficiency of health promotion (Watt and Sheiham, 2012).

Based on the WHO recommendations, a set of guiding principles for developing oral health strategies has been summarised to improve oral health and address inequalities including - empowering and engaging communities through holistic participatory approaches, intersectoral, partnership and multi-strategy working, evidence based practice and policy implemented with robust evaluation approaches (Watt, 2007).

Chapter 8: Conclusions and Recommendations

8.1 Conclusions

This pilot study set out to assess the feasibility of recruiting and following up a cohort of preschool children, collect data to investigate behavioural, biological and social factors associated with dental caries and their role in explaining socioeconomic inequalities in dental caries.

Successful tools and methods, lessons learnt and data collected from this study will be used to design a large birth cohort study that will permit a detailed evaluation of the current *Childsmile* programme. The prospective cohort study will collect data at an individual level to identify modifiable risk factors and the 'causes of the causes' to inform and direct the development of future interventions within the *Childsmile* programme to improve oral health overall and tackle oral health inequalities in Scotland.

8.1.1 Feasibility

The following conclusions have been drawn from this study relating to the feasibility of recruiting and following up a cohort of preschool children to collect clinical and questionnaire data:

1. The method used for recruitment via nursery schools proved to be an effective platform for identifying eligible families across the socioeconomic spectrum and recruiting and engaging a large majority of the conventionally 'hard-to-reach' groups.

2. The anticipated recruitment rate at the beginning of the study was achieved in spite of the study largely recruiting from more deprived areas. Socioeconomic position was not found to influence refusal to participate in this study. The nursery-staff engagement with the study and their influence on the families of the children was thought to be important in recruitment.

3. The time of the year when the study is carried out may affect recruitment rates. Recruitment rates were relatively lower among nursery schools that ran over summer holidays.

4. Once participating in the study, compliance was high (above 80%) for clinical data collection in both Sweeps and questionnaire data collection in Sweep 1. A large proportion of the most deprived groups were successfully engaged and retained in this study. Socioeconomic background was not found to influence compliance. Again, this high compliance rate largely reflects the utility of the educational establishment setting, where clinical data collection was possible for relatively large numbers of children at a time. Compliance may also have been influenced by the rapport between staff and parents that aided good questionnaire returns in Sweep 1. Other factors that may have influenced the high compliance may be related to timely reminders and the flexibility offered by the researcher in data collection.

5. It was feasible to simultaneously conduct dental clinical examination, collect saliva and plaque samples and measure heights and weights across educational establishment settings over time.

6. The collection of unstimulated saliva using the Salimetrics Children's Swab was found to be an easy, acceptable, relatively stress free and reliable technique for use among young children. There was sufficient quantity and quality in the data collected on which microbiological, some immunological and stress markers were measured and validated. The use of TaqMan® QPCR was found to be more reliable and relatively faster for the estimation of *S.mutans* than conventional culture methods. The quality of other clinical (dental caries and anthropometric) data collected was good.

7. The collection of plaque samples using CPITN probes was an easy and acceptable technique. The quality of the data collected has not been assessed in this body of work.

8. Overall, the completeness and quality of the data collected through questionnaires was high, particularly in relation to 'About the Study Child' and 'About your family'. The completeness and quality of data collected did not appear to be influenced by socioeconomic factors. The Semi-Quantitative Food Frequency Questionnaire (FFQ) developed by the Scottish Collaborative Group was found to be detailed and appropriate for use among preschool children to provide robust data (NMES as a percentage of food energy) to identify a relationship between diet and caries in a future large-scale setting. The completeness of the FFQ was slightly lower than those of 'About the Study Child' and 'About your family'.

8.1.2 Risk factor models for caries

This was a pilot study and caution is warranted in extrapolating the findings beyond the scope of the data collected. The conclusions from risk factor models may be more or less indications (than definite conclusions) that need further investigation in a larger study. The prevalence of caries was high and in line with national studies, and followed a socioeconomic gradient. The following conclusions have been drawn from the risk factor models for caries produced in this pilot cohort study:

1. The level of *Streptococcus mutans* in saliva at baseline was the most proximal risk factor associated with caries a year later, independent of other risk factors.

2. A range of 'modifiable' oral health related behavioural factors (higher NMES intake, irregular brushing habits immediately before bedtime, rinsing out toothpaste after tooth-brushing, night time bottle feeding at two years and older age at first visit to dentist) were found to be independently associated with caries a year later.

3. The only distal risk factor and socioeconomic measure that was associated with caries a year later was the area-based measure of socioeconomic position (SEP), Scottish Index of Multiple Deprivation (SIMD) at national

level. Other individual-level measures of SEP were found to be correlated with SIMD.

The Relative Index of Inequality (RII) showed that each measure of SEP explained a different aspect of SEP that contributed differently to inequalities in caries. The results emphasised the importance of using multiple measures of SEP in epidemiological studies.

There was some attenuation in the RII after adjustment for relevant risk factors. However, the socioeconomic gradient in caries persisted, suggesting that perhaps other more upstream factors may be important in causing inequalities.

Sample size for the larger cohort study will depend on a number of statistical and non-statistical factors, including the hypotheses to be tested, variables of interest (social/ environmental/ behavioural/ biological/ genetic etc), desired accuracy and precision of the effect estimates, geographical and social differences within the population and in their response to interventions. These factors are not fully clear at this stage. Nevertheless, the effect sizes from the multivariable models in this study will serve as a starting point to perform a power calculation to identify the sample size that will be required in the larger cohort study.

8.2 Recommendations

After having tested the feasibility of conducting a cohort study among preschool children and following them up one year later, examining factors associated with caries and their role in explaining socioeconomic inequalities in caries, the following recommendations were drawn to guide the design of a larger cohort study that will robustly evaluate the outcomes of *Childsmile* and inform and guide future direction of the programme:

1. The utilization of nurseries is recommended as an appropriate platform for recruitment of preschool children and collecting data (caries, heights, weights, microbiological samples and questionnaire) from them. Data can be collected from a large cross-section of children across the socioeconomic spectrum. Most importantly, it is recommended as a

platform of choice to engage the conventionally 'hard–to-reach' groups. However, transition points like moving from nursery school to primary school may be associated with constraints of access in the follow-up, which can be labour and resource intensive.

2. Where possible, the sampling of nursery schools should take into account the presence of a key staff with interest in oral health who can actively champion the research study.

3. It is recommended that recruitment of participants into the study via nursery schools are completed prior to the various academic breaks at schools. Term-time breaks, particularly those that are long are likely to lower recruitment rates.

4. Those studies planning to collect health-related data (anthropometric, dental) from school children on a regular basis (routinely) in an educational setting should consider coordinating data collection activities into a single school visit. This minimizes disruption during the school term and may have cost efficiencies.

5. Saliva is a valuable specimen on which numerous investigations can be performed. It is recommended that future studies make careful decisions on prospective salivary assays to perform and approximate volumes of saliva that may be required. It is further recommended that a longer collection time of up to a maximum of 90 seconds may be needed in some children (if compliant) if the swab does not appear to be saturated with saliva. Assays of greatest importance must be predetermined in the event that an inadequate volume of saliva is available for all.

6. It was occasionally not possible to clinically examine (collect microbiological samples) children before the snack break and toothbrushing session at nurseries. It will be valuable to check if the data was compromised by this practicality issue.

7. It is recommended that multiple strategies including timely reminders, telephone follow-up and flexibility in the mode of data collection and schedule be used to improve response rates.

8. The use of postal services and scheduled data collection events at the Glasgow Dental hospital & School are recommended alternative cost cutting methods to pack delivery, questionnaire collection and data collection at schools.

9. 'About your study child' and 'About your family' questionnaires are considered suitable for use in large-scale from the completeness and quality of data that was obtained.

10. The FFQs used in this study provided robust nutritional data for preschool children and are recommended for use in large-scale. This study did not have face-to-face sessions with the majority of the main-carers, as the completed questionnaires were returned through the schools. No attempt was made to contact the main-carer again to have blank lines completed. It would be beneficial to have a follow up protocol for incomplete FFQs for future studies.

11. The Standard Operating Procedures available in Appendices I and K are recommended for use in collecting microbiological samples and measuring heights & weights respectively.

12. Risk factors and causes of socioeconomic inequalities in caries are complex and have roots in early life. The explanation of socioeconomic inequalities in caries among young children remains to be answered. It is likely that the explanations for the observed inequalities in caries lie in the 'cause of the cause' hypothesis (Marmot, 2010), with the behavioural risk factors being widely accepted causes for caries that are socially determined. Further work is required to include community-level upstream factors into the hypothetical model developed in this study like those recommended by the World Health Organization (Peterson et al., 2003), and more recently by Watt and Sheiham (2012); which emphasise

the political context and structural mechanisms generating social hierarchy of individuals.

13. Future studies are recommended to utilize sophisticated statistical modelling techniques like multilevel modelling, path analysis or structural equation modelling and multiple measures of SEP to more fully explain the pathways and mechanisms involved in inequalities.

8.3 Further work

Although pilot in nature, this study has created a unique dataset, which includes data on multiple risk factors across various levels associated with caries. Further work from the data collected in this study will significantly advance our understanding of caries development. These include:

1. The children in this study should be matched to their Community Health Index (CHI) number, which will aid in linking with routine data held within the Information Services Division Scotland. Of priority should be the unachieved fluoride varnish linkage in this body of work.

2. There is potential in the dataset to look at changes in behaviours and parental attitudes over time according to socioeconomic position and how these affect caries development.

3. There is immense potential in understanding the biological pathways that may underlie socioeconomic inequalities in caries. A metagenomic approach using the plaque samples collected at both Sweeps is now being used to investigate the variation in the ecology of the complex bacterial flora and *S. mutans* genotypes over time, their role in caries and with SEP. Additionally, the immunological factors and cortisol validated in this body of work may further our understanding of the host-pathogen interactions in health and diseased state (work currently underway).

4. Results from recent systematic reviews are equivocal of a relationship between obesity and caries in the primary dentition and have called for longitudinal studies that take into account the large number of

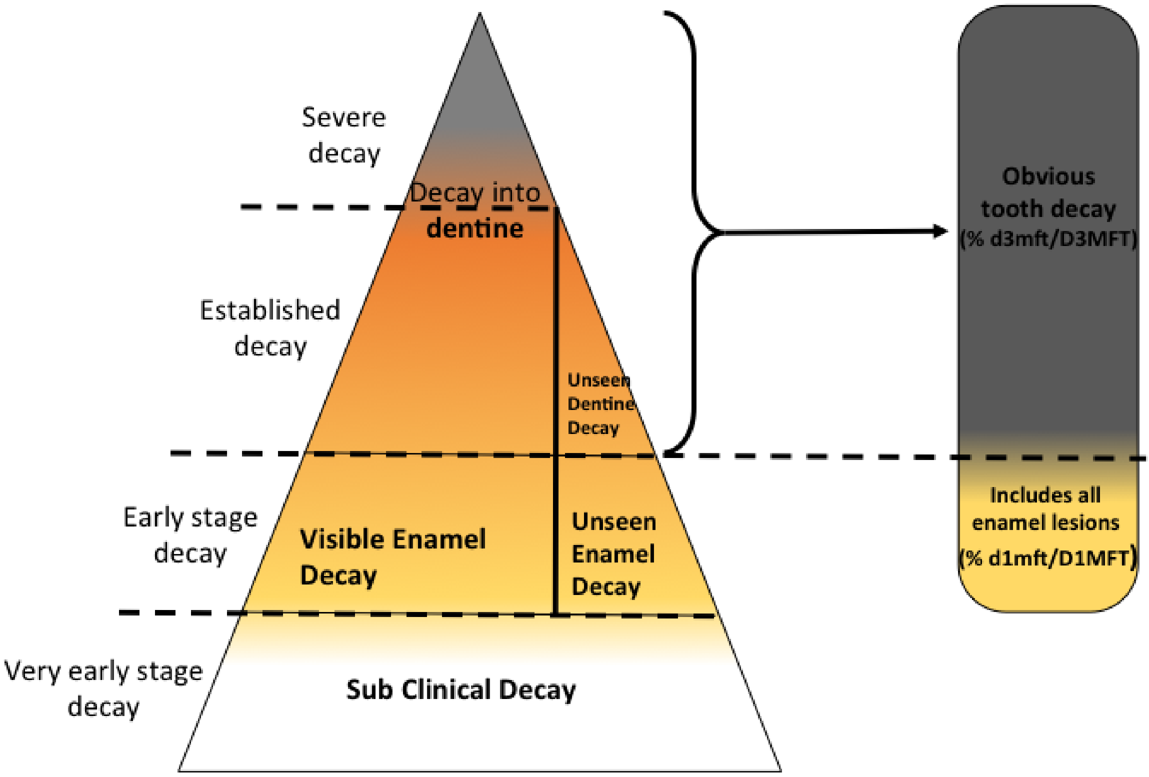
confounding factors. The dataset from this study offers a good opportunity to investigate the relationship between the two conditions robustly.

5. Robust and rich dietary data is available that can be used to test multiple hypothesis- For e.g. the possibility of an association between obesity and intake of energy, NMES, total fat and saturated fatty acids- although limited information on physical activity is available.

It is thought that the analyses of all the data collected in this study will further our understanding of caries development and inequalities in young children. It will give more clarity into the variables and relationships that are worthy of further investigation within a larger cohort study. The present study has set the scene for future epidemiological work on the aetiology and social patterning of dental caries in children in Scotland, at the individual child level from which future interventions can be designed to further improve oral health and reduce inequalities.

Appendices

Appendix A: Caries Iceberg



The caries diagnostic threshold represented in the form of an iceberg. Adapted from Pitts (2004)

Appendix B: Pack 1

a) Covering letter

University of Glasgow | Dental School

9 June 2011

Dear Parent/Guardian,

Invitation to participate in a research project about pre-school children's dental health

We are looking at pre-school age children in nursery schools and we would like to inform you about a research study. We would also like to revisit these children when they start school.

Firstly, we would like to collect a small volume of your child's saliva and dental plaque and we would also like to measure their height and weight. Secondly, we would like to ask you some questions that might help us understand why some children get tooth decay and others do not. This will include questions about your child's diet, your family circumstances and dental hygiene.

Once you have completed the first part of the study (pre-school stage) you will be entered into a prize draw with a chance to win supermarket vouchers worth £100 as 1st prize, £50 as 2nd prize and £25 as 3rd prize; and again if you complete the second part of the study (Primary 1 stage). Your nursery will also receive a cash sum towards their funds.

Please also find attached an information sheet with more details about the study. If you would like to contact me please feel free to do so using the contact details below.

We will be sending you a second pack in around one week's time, containing a Consent Form for you to complete if you wish to take part in our study.

We would stress that participation in the study is voluntary and you can withdraw your consent at any point.

Once we have collected all of the information we need from you, you will be entered into the first prize draw.

Thank you very much for taking the time to read this information.

Yours Sincerely,

Andrea Sherriff

Dr Andrea Sherriff
Senior Lecturer
Glasgow University Dental School
Tel: 0141 211 9801
Email: andrea.sherriff@glasgow.ac.uk

b) Participant information leaflet

University of Glasgow | Dental School

Participant Information Leaflet

A pilot cohort study to investigate risk factors for dental caries

Dr Andrea Sherriff
Senior Lecturer
Glasgow University Dental School
378 Sauchiehall Street
Glasgow, G2 3JZ
Tel: 0141 211 9801
Email: andrea.sherriff@glasgow.ac.uk

WHY HAS YOUR CHILD BEEN CHOSEN?
Your child is invited to take part in this research study as they are in their pre-school year at nursery. Please take time to read the following information carefully.

WHAT IS THE PURPOSE OF THIS STUDY?
The purpose of this study is to understand why some children get tooth decay and others do not. It is important for our study that we collect as much information as possible on why tooth decay happens. This means collecting saliva and dental plaque from children as well as information about their diet, their dental hygiene and their family circumstances. We ask these questions because they are very important for our study and we would like to assure you that this information will be kept completely confidential.

WHAT DOES YOUR CHILD HAVE TO DO? ARE THERE ANY RISKS?
1. We wish to collect a small volume of saliva by placing a cotton wool swab under your child's tongue for a few seconds.

2. A small amount of dental plaque will be collected from the mouth. The plaque will then be placed in fluid and the bacteria will be analysed at a later date.

3. Your child will have their height and weight measured in light indoor clothing with their shoes removed.

Your child should not experience any discomfort and there are no risks associated with any of the procedures. These will be collected at your child's nurseries in the presence of their teacher.We wish to collect samples and heights and weights on two occasions, once in the pre-school year at nursery and once in Primary 1.

We would also like to know about changes in children's diet, dental hygiene and family circumstances as they move from nursery school to primary school, as this may help us further understand why some children develop tooth decay and others do not. We will ask you to provide us with this information at both times preferably at your child's nursery/school or other convenient location you may choose.

DOES YOUR CHILD HAVE TO TAKE PART?
It is up to you to decide whether or not your child should take part in this study. If you decide you would like your child to take part in the study, you will be given this information sheet and consent form to sign and keep. You will have an opportunity to ask questions and have them answered to your satisfaction. If you choose for your child not to take part in the study, your child will not be disadvantaged in any way. **Once the samples, height and weight and Study Data have been collected your name will be entered into a prize draw to win supermarket vouchers worth £100 as 1st prize, £50 as 2nd prize and £25 as 3rd prize.**

University of Glasgow | Dental School

WHAT ARE THE BENEFITS OF YOUR CHILD TAKING PART?
Your child's participation in this study may help improve dental care for children in the future, through better understanding of why some children get tooth decay and others do not. However, your child will not directly benefit from taking part in this study.

WHAT WILL HAPPEN TO MY SAMPLES AND PERSONAL DATA?
Your child's samples and associated personal data ("Study Data") will be stored securely, processed and used for investigations at the University of Glasgow. Any preserved saliva samples and Study Data may be stored for up to 15 years. All samples will only be used for the purposes described.
The studies that we are doing on your child's saliva and dental plaque will not suggest any clinical diagnosis or treatment for any disease and it is not the purpose of this research to provide you or your child with test results.

By signing the Consent Form you consent to the Study Dentist and his or her staff collecting and using your child's personal data for the study. This includes your participant profile, your questionnaire responses on your child's diet and dental hygiene; and your child's date of birth, sex, postcode and tooth decay score (the tooth decay score is collected as part of the normal dental exam). Your consent to the use of your child's Study Data does not have a specific expiration date. By signing this form you consent to the use of Study Data as described in this form.

HOW WILL MY CONFIDENTIALITY BE PROTECTED?
Special precautions are taken to ensure the research study is carried out with a high degree of confidentiality. If you agree to participate in the study, a code that is specific to your child will be used to label your child's samples and Study Data and identify all results that are recorded at the University of Glasgow. This coding of all information is to ensure that the results are kept confidential by keeping your child's identity and the results separate. **Samples will not include your name, address or Hospital number.** Only the Study Dentist and health care professionals who are part of the research team have access to the code key that will connect your Study Data to you. The Study Dentist is responsible for handling of your child's Study Data in accordance with applicable Data Protection law(s). Please note, the results of the study may be published in medical literature, but your child will not be identified.

CAN I WITHDRAW MY CONSENT?
You are free to withdraw your consent for use of the samples or Study Data at any time. If you choose to withdraw your consent, your child will not be disadvantaged in any way, including dental treatment and care your child is entitled to receive. If you withdraw your consent, your child's sample(s) will be destroyed. The University may still use Study Data obtained before you withdrew your consent.

WHO SHOULD I CONTACT FOR INFORMATION OR HELP?

Dr Andrea Sherriff
Senior Lecturer
Glasgow Dental School & Hospital
378 Sauchiehall Street
Glasgow, G2 3JZ
Tel: 0141 211 9703

Mrs Silda Sadique
Clinical Academic Fellow
Glasgow Dental School & Hospital
378 Sauchiehall Street
Glasgow, G2 3JZ
Tel: 0141 211 9746

Prof Lorna Macpherson
Professor of Dental Public Health
Glasgow Dental School & Hospital
378 Sauchiehall Street
Glasgow, G2 3JZ
Tel: 0141 211 9750

Thank you for taking the time to read this participant information leaflet.

Appendix C: Pack 2

Covering letter and Consent form

University of Glasgow | Dental School

17 June 2011

Dear Parent/Guardian,

Participation in a research project about pre-school children's dental health

About a week ago, we sent you some information about our study. If you would like to take part in the study, please complete and sign the enclosed Consent Forms.

We would like you to complete and sign both of the consent forms, put one into the envelope addressed to your child's head teacher, Mrs Avril Williamson (you can keep the other) and then send this back to the nursery in your child's nursery bag before the **25th of May. You'll notice that by signing the consent form you are giving us permission to visit your child at their nursery for a clinical examination, and again in Primary 1.**

At present, we have not organized a date to visit your child's nursery. Once we have, your nursery will let you know.

If you would like to be entered into the prize draw with a chance to win supermarket vouchers worth £100 as 1st prize, £50 as 2nd prize and £25 as 3rd prize, please complete the enclosed Prize Draw slip. Once we have seen your child and collected the information from you, you will be entered into the prize draw.

For now please:

- Sign both consent forms
- Fill in your details on the prize draw slip
- Place one consent form and the prize draw slip in the envelope provided.
- Send back to the nursery in your child's bag before the **1st of July**.

Thank you again for your help.

Yours Sincerely,

Andrea Sherriff

Dr Andrea Sherriff

Senior Lecturer
Glasgow University Dental School
Tel: 0141 211 9801
Email: andrea.sherriff@glasgow.ac.uk

University of Glasgow | Dental School

Participant ID

A pilot cohort study to investigate risk factors in dental caries

CONSENT FORM

Dr Andrea Sherriff
Senior Lecturer
Glasgow University Dental School
378 Sauchiehall Street
Glasgow, G2 3JZ
Tel: 0141 211 9801
Email: andrea.sherriff@glasgow.ac.uk

Please initial boxes

I confirm that I have parental responsibility for the child above and have read and understood the information sheet. I have had the opportunity to consider the information, ask questions and have had these answered satisfactorily. I have read and understand the information sheet for the above study. ☐

I understand that my participation is voluntary and that I am free to withdraw at any time, without giving any reason, without my dental care or legal rights being affected. ☐

I agree to myself and my child taking part in the above study. ☐

I confirm that I have received a copy of this information and signed consent form to keep. ☐

I give permission for my child's health information to be used in a non-identifiable format through publications, reports and presentations of this research study. ☐

____
Name of Study Child

Name of Subject's Parent	Signature	Date
Name of Research Team Member	Signature	Date

One copy to be retained by parent and one copy to be retained in study file.

Appendix D: Pack 3

Covering letter

University of Glasgow | Dental School

27th April 2011

Dear Parent or Guardian,

Filling booklets for a research project about pre-school children's dental health

Thank you for agreeing to take part in our study. We really appreciate your help.

You will find 3 booklets inside the envelope given to you by the nursery. Please fill in **all three booklets** and return them to the nursery in the enclosed addressed envelope by **Tuesday the 10th of May**.

Filling in the booklets should only take 30-45 minutes of your time.

- The first booklet: **About your Study Child** is about the Study Child.
- The second booklet: **Diet Questionnaire** is about the Study Child
- The third booklet: **About you and your family** is about the family of the Study Child

Once we receive your completed booklets and we have visited your child at nursery, you will be entered into our Prize Draw for £100, £50 or £25 of supermarket vouchers.

If you have not returned the completed booklets by Tuesday 10th May to the nursery, our researcher, Silda Sadique, will contact you to offer help in filling in the booklets if you need it.

Once again, thank you very much for your time.

Yours Sincerely,

Andrea Sherriff

Dr Andrea Sherriff
Senior Lecturer
Glasgow University Dental School
Tel: 0141 211 9801/ 9750
Email: andrea.sherriff@glasgow.ac.uk

Appendix E: About your Study Child questionnaire

University of Glasgow | Dental School

CHILDSMILE

Participant Code: | | | | | | |

About your Study Child

To help us with our study, we would like to ask you about your child's toothbrushing habits, how they were fed when they were younger and your views on dental health. We would like you to answer for the child who is part of this study (the Study Child). There are no right or wrong answers.

This questionnaire should only take around 10 minutes to complete.

Your responses will be treated confidentially.

Thank you very much for your help.

How to complete this questionnaire

- Most of the questions can be answered by putting a tick in the box next to the answer you want to give.
- If you make a mistake, just cross out your first tick and add another one in the box next to the right answer.
- A few questions require you to write your answers in the spaces provided.

Please remember to put all 3 booklets in the envelope, seal it and hand it to your child's nursery head teacher by the

If you have any questions or require further information please contact:

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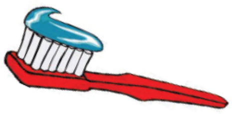
Clinical Academic Fellow
Glasgow Dental School & Hospital
378 Sauchiehall Street
Glasgow, G2 3JZ
Tel: 0141 211 9746
email: silda.sadique@glasgow.co.uk

2

Toothbrushing



First a few questions about your Study Child's toothbrushing habits and the toothpaste they use.

1. From what age were your child's teeth brushed regularly? *(Tick 2 boxes if necessary)*

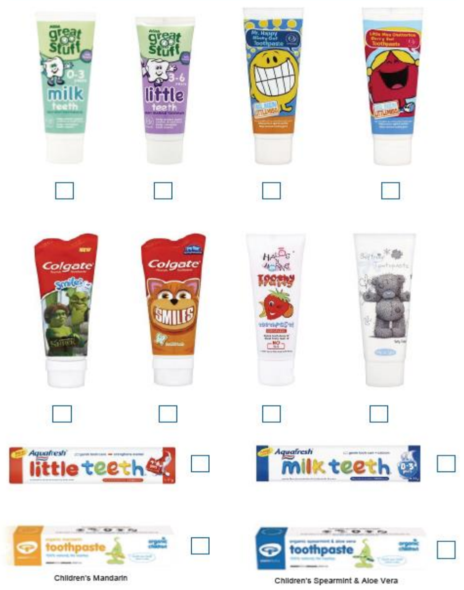
When the first tooth came into the mouth	
Under 12 months	
Between 13 months and 2 years	
Between 2 years, 1 month and 3 years	
Between 3 years, 1 month and 4 years	
Not yet brushing regularly	

2. How often are your child's teeth brushed? *(Please tick only one box)*

Not every day	
Once a day	
Twice a day	
More than twice a day	

3

3. Which toothpaste(s) are used to brush your child's teeth?
Please tick the box or boxes below/beside each brand of the toothpaste that your child currently uses. If the toothpaste your child uses is not in the list, please write the name of the toothpaste your child uses here:



4

Asda Smartprice

Asda Essential Care Sensitive TotalCare Whitening Toothepaste

Sainsbury's Little Ones Fruity

Sainsbury's Little Ones Mild Mint

5

4. Who brushes your child's teeth? *(Please tick only one box)*

Always brushed by the child	☐
Always brushed by an adult	☐
Sometimes brushed by an adult, sometimes by the child.	☐

Other (*Please describe*) ___________________________

5. Which of the options below **best** describes the method your child uses to remove toothpaste after brushing? *(Please tick only one box)*

Swallows toothpaste	☐
Spits out toothpaste	☐
Spits out and rinses out toothpaste	☐

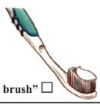
6. Are your child's teeth brushed immediately before bedtime? *(Please tick only one box)*

Always	☐
Sometimes	☐
Occasionally	☐
Hardly ever	☐
Never	☐

7. Which of the pictures below **best** show the amount of toothpaste used when your child's teeth are brushed? *(Please tick only one box)*

"smear" ☐

"pea-size" ☐



"half a brush" ☐

"full brush" ☐

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Dental visits

8. How old was your child when he/she went to the dentist for the **first time**?

Please write the age here: __________

Or tick here if your child has not yet visited a dentist ☐

9. Why did your child go to the dentist for the **first time**? *(Please tick only one box)*

He/she was having trouble with his/her teeth	☐
We had a note from the nursery dentist	☐
He/she went for a check up	☐
He/she just went to get used to going to the dentist	☐
An organised Childsmile visit	☐
Never been to the dentist	☐

Other: ___________________________

10. On average, how often does your child attend a dentist for a **routine** check-up? *(Please tick only one box)*

More frequently than once every six months	☐
Once every six months	☐
At least every 12 months but not as often as every 6 months	☐
Less often than every 12 months	☐
Never	☐

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Early feeding habits

11. When your child was a baby, did you breast or bottle feed him/her? *(Please tick only one box)*

Breast fed	
Bottle fed	
Breast **and** bottle fed	

Other: (*Please describe*) ____________________________

12. How old was your child when he/she **completely stopped** using a baby bottle with a teat? *(Please tick only one box)*

Never used a baby-bottle	
Under12 months	
Between 1 year and 2 years	
Between 2 years, 1 month and 3 years	
Between 3 years, 1 month and 4 years	
Still using a baby bottle	

13. At what age did your child start eating solid food? *(Please tick only one box)*

Under 2 months	
2-3 months	
4-6 months	
7-12 months	
Over 12 months	

14. **Other than water**, how often did your child take a drink to bed during the night:

a) in the **first two years**?

Every day	
Most days	
Some days	
Hardly ever	
Never	

b) since he/she was **4 years old**

Every day	
Most days	
Some days	
Hardly ever	
Never	

15. How often do you restrict the amount of sugary foods your child eats? *(Please tick only one box)*

Every day	
Most days	
Some days	
Hardly ever	
Never	

Finally, we are interested in your views about dental health. Please tell us how strongly you agree or disagree with each of the following statements below.

Please tick one box for each statement.

		Strongly agree	Agree	Neither agree nor disagree	Disagree	Strongly disagree
16	My child losing a baby tooth due to tooth decay would be upsetting					
17	It is worthwhile to give my child sweets/biscuits to behave well					
18	As a family, we are confident we can reduce the chances of our child getting tooth decay					
19	Most children eventually develop dental cavities					
20	It is the responsibility of the dentist to prevent my child getting tooth decay					
21	As a family, we intend controlling how often the child has sugary foods or drinks between meals					
22	It is not worth it to battle with my child to brush his/her teeth twice a day					
23	It is often too stressful to say no to my child when they want sweets					
24	If my child uses fluoride toothpaste, it will prevent tooth decay					
25	As a parent, it is my responsibility to prevent my child getting tooth decay					

26. During the school term, how much time does your child spend watching television or playing computer games? *(Please tick only one box)*

None (No time at all)	
Less than 1 hour per day	
1-2 hours per day	
3 or more hours per day	

Thank you very much for completing this questionnaire. Your help is very much appreciated.

Please put all the 3 booklets in the envelope, seal it and hand it to your child's nursery head teacher by the

If you have any questions or require further information please contact:

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378 Sauchiehall Street
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Tel: 0141 211 9746
email: silda.sadique@glasgow.co.uk

Appendix F: About your Family questionnaire

Thank you very much for completing this questionnaire. Your help is very much appreciated.

Please remember to put <u>all 3 booklets</u> in the envelope, seal it and hand it to your child's nursery head teacher by the

If you have any questions or require further information please contact:

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Tel: 0141 211 9746
email: silda.sadique@glasgow.co.uk

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University of Glasgow | Dental School

CHILDSMILE

Participant Code: | | | | | | |

About You and Your Family

To help us with our study, we would like to know a little about you and your family. This questionnaire should only take around 5-10 minutes to complete. There are no right or wrong answers.

Your responses will be treated confidentially.

Thank you very much for your help.

How to complete this questionnaire

- Most of the questions can be answered by putting a tick in the box next to the answer you want to give.
- If you make a mistake, just cross out your first tick and add another one in the box next to the right answer.
- A few questions require you to write your answers in the spaces provided.

Please remember to put all 3 booklets in the envelope, seal it and hand it to your child's nursery head teacher by the

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email: silda.sadique@glasgow.co.uk

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14. What is the **Study Child's** racial or ethnic group? *(Please tick only one box)*

White: Scottish	
White: Other British	
White: Any other white background (Please specify below)	
Asian, Asian Scottish or Asian British: Pakistani	
Asian, Asian Scottish or Asian British: Indian	
Asian, Asian Scottish or Asian British: Chinese	
Asian, Asian Scottish or Asian British: Any other Asian background (Please describe below)	
Mixed: Any mixed background	
Black, Black Scottish or Black British: Caribbean	
Black, Black Scottish or Black British: African	
Black, Black Scottish or Black British: Any other black background (Please specify below)	
Any other ethnic group	

Please describe here ____________________

7

10. Is your home:

Being bought/ mortgaged	
Owned with no mortgage	
Rented from Local Authority/ council	
Rented from Housing Association	
Rented- Privately unfurnished	
Rented- Privately – furnished	
Rented from an employer	

Other: (Please describe) ______________________________

The following 4 questions are about the Study Child

11. What is the **postcode** of the **Study child's** home address? ____________________
Please write the FULL postcode eg G66 5UR

12. How many children are in the **Study child's** family, including the Study Child? ________

13. In your family, is the **Study Child**: *(Please tick only one box)*

First born	
Second born	
Third born	
Other	

Other: (*Please describe*) ______________________________

1. **Are you the main carer, or one of the main carers of the Study Child?**
(Please tick only one box)

Yes	
No	

2. What is your relationship with the **Study Child**? *(Please tick only one box)*

Mother	
Father	
Grandparent	

Other: *(Please describe)* ______________________________

3. How old are **you**?

Please write your age in years: ____________________

4. What is your marital status? *(Please tick only one box)*

Married	
Cohabiting/ living with partner	
Single	
Widowed	
Divorced	
Separated	

5. Do you smoke? *(Please tick only one box)*

Yes	
No	

6. How many years of full time education have you completed **after** leaving secondary school?

Please write here: ________ *Years.*

If you did not finish secondary school please tick here ☐

7. What educational qualifications do you have? (*Please tick ALL boxes that apply to you*).

School leaving certificate/ NNQ Access Unit	
O Grade, Standard Grade, GCSE, CSE, Senior Certificate	
GSVQ Foundation or Intermediate, SVQ Level 1 or 2, SCOTVEC Module or equivalent, NNQ Higher Still Access 3 Cluster, Intermediate 1 or 2	
SCE Higher Grade/ NNQ Higher or Advanced Higher/ CSYS/ A level, Advanced Senior Certificate	
GSVQ Advanced, SVQ Level3, ONC, OND, SCOTVEC National Diploma	
City and Guilds	
HNC/HND/SVQ Level 4 or 5	
First Degree/ Higher Degree	
None of these qualifications	

Other: (*Please describe*)

____

8. Which of the options from the table below would you place your **total** household income (including yourself) from all sources, **before tax and other deductions.**
(Please tick only one box)

Weekly income before tax		Annual income before tax
Less than £77		Less than £3,999
£78 - £115		£4,000 - £5,999
£116 - £154		£6,000 - £7,999
£155 - £192		£8,000 - £9,999
£193 - £230		£10,000 - £11,999
£231 - £289		£12,000 - £14,999
£290 - £346		£15,000 - £17,999
£347 - £385		£18,000 - £19,999
£386 - £442		£20,000 - £22,999
£443 - £500		£23,000 - £25,999
£501 - £558		£26,000 - £28,999
£559 - £615		£29,000 - £31,999
£616 - £673		£32,000 - £34,999
£674 - £730		£35,000 - £37,999
£731 - £788		£38,000 - £40,999
£789 - £846		£41,000 - £43,999
£847 - £903		£44,000 - £46,999
£904 - £961		£47,000 - £49,999
£962 and more		£ 50,000 and more

9. What proportion of your household income (including your own) would you say comes from benefits? *(Please tick only one box)*

Child benefit only	
None	
About a quarter	
About a half	
About three quarters	
All	

Appendix G: Sweep 1 Data record sheet

PARTICIPANT ID

Gender	Height	Weight	Clothing	Shoes removed	Time	Saliva	Plaque	Observer Name	Questionnaire
F/M	cm	kg	L/H	Y/N					

Comments: ____

AMR index:

55	54	53	52	51	61	62	63	64	65
85	84	83	82	81	71	72	73	74	75

A (Active dentinal decay):

M (Missing due to decay):

R (Restored due to decay):

AMR = A+M+R=

Appendix H: Cross-sectional analyses

Unadjusted and adjusted Odds Ratios & 95% Confidence Intervals for caries experience and C-index (SWEEP 1) according to demographic variables

Explanatory Variables	N	% caries experience	Unadjusted OR [95% CI]	Adjusted OR [95% CI][60]
Gender				
Male	[37/100]	37	1 [ref]	1 [ref]
Female	[31/90]	34.4	0.90 [0.49 to 1.62]	0.89 [0.36 to 2.18]
P value			0.714	0.796
C-index [95% CI]			0.51 [0.43 to 0.60]	0.83 [0.76 to 0.90][61]
Age			0.75 [0.32 to 1.74]	1.01 [0.31 to 3.31]
P value			0.505	0.986
C-index [95% CI]			0.53 [0.44 to 0.62]	
Ethnicity				
White: Scottish	[42/122]	34.4	0.98 [0.46 to 2.06]	
Others[62]	[14/40]	25	1 [ref]	
P value			0.947	
C-index [95% CI]			0.65 [0.56 to 0.74]	
Number of children in family				
1	[11/36]	30.6	1 [ref]	
2	[24/67]	35.8	1.27 [0.53 to 3.02]	
3 or more	[19/57]	33.3	1.14 [0.46 to 2.79]	
P value			0.862	
C-index [95% CI]			0.52 [0.43 to 0.62]	
Rank of child in family				
First born	[21/70]	30	1 [ref]	
Second born	[24/57]	42.1	1.70 [0.82 to 3.53]	
Third born or later	[9/31]	29.0	0.96 [0.38 to 2.42]	
P value			0.290	
C-index [95% CI]			0.57 [0.47 to 0.66]	
Age of Main carer				
<25 years	[8/17]	53.3	1.65 [0.53 to 5.12]	
26 -35 years	[29/80]	50.7	1.48 [0.74 to 2.97]	
36 years and over	[19/66]	41.0	1 [ref]	
P value			0.471	
C-index [95% CI]			0.55 [0.46 to 0.65]	
Main Carer marital Status				
Married/Cohabiting	[37/115]	32.2	1[ref]	
Single parent	[19/48]	39.6	1.38 [0.69 to 2.78]	
P value			0.365	
C-index [95% CI]			0.53 [0.44 to 0.63]	
Main-Carer smoking status				
Current Smoker	[19/48]	39.6	1.40 [0.7 to 2.81]	
Not current smoker	[37/116]	31.9	1 [ref]	
P value			0.346	
C-index [95% CI]			0.54 [0.44 to 0.63]	

[60] Fully adjusted multivariate model

[61] C- index for the fully adjusted multivariate model

[62] Other Whites, Asians, Blacks and Mixed

Unadjusted and adjusted Odds Ratios & 95% Confidence Intervals for caries experience and C-index (SWEEP 1) according to Socioeconomic Position

Explanatory	N	% caries	Unadjusted OR	Adjusted OR [95%
Local SIMD[64] Quintile				
Q1 (most	[28/66]	42.4	1.63 [0.74 to 3.62]	
Q2	[17/38]	44.7	1.79 [0.73 to 4.40]	
Q3	[8/39]	20.5	0.57 [0.21 to 1.56]	
Q4 & Q5 (least	[14/45]	31.1	1 [ref]	
P value			0.081	
C-index [95% CI]		0.61 [0.52 to 0.69]		
National SIMD[65] Quintile				
Q1 (most	[43/102]	42.2	3.06 [1.07 to 8.76]	
Q2	[5/25]	20	1.05 [0.26 to 4.19]	
Q3	[14/35]	40	2.80 [0.86 to 9.18]	
Q4 & Q5 (least	[5/26]	19.2	1 [ref]	
P value			0.058	
C-index [95% CI]		0.60 [0.52 to 0.68]		
Gross annual household Income				
< £10,000	[21/41]	51.2	4.04 [1.70 to 9.59]	
£10,000 – £19,999	[16/47]	34	1.99 [0.84 to 4.68]	
£20,000 & more	[13/63]	20.6	1 [ref]	
P value			0.007	
C-index [95% CI]		0.65 [0.56 to 0.74]		
Percentage of income from benefits				
Half or more	[28/64]	43.8	2.56 [1.28 to 5.12]	
None or About a	[21/90]	23.3	1 [ref]	
P value			0.028	
C-index [95% CI]		0.61 [0.52-0.71]		
Current or last occupation of Main-Carer				
Never worked or	[5/9]	55.6	5.94 [1.08 to	
NS-SEC 3[66]	[20/42]	47.6	4.32 [1.25 to	
NS-SEC 2[67]	[4/24]	16.7	0.95 [0.21 to 4.35]	
NS-SEC 1[68]	[4/23]	17.4	1 [ref]	
P value			0.014	
C-index [95% CI]		0.69 [0.58 to 0.80]		
Main-Carer's years of Full-time Education				
Not completed	[19/51]	37.3	1.31[0.65 to 2.65]	
Further education	[33/106]	31.1	1 [ref]	
P value			0.446	
C-index [95% CI]		0.53 [0.43-0.63]		
Main-Carer's highest Level of Education				
Level 0[69]	[12/25]	48	4.09 [1.31 to	
Level 1[70]	[19/45]	42.2	3.24 [1.18 to 8.90]	
Level 2[71]	[16/54]	29.6	1.87 [0.68 to 5.10]	
Level 3[72]	[7/38]	18.4	1 [ref]	
P value			0.05	
C-index [95% CI]		0.63 [0.54 to 0.72]		

[63] Fully adjusted model
[64] Scottish Index of Multiple Deprivation 2009 at Local Health Board level (NHS Greater Glasgow & Clyde)
[65] Scottish Index of Multiple Deprivation 2009 at National level
[66] NS-SEC 3 - Routine & manual
[67] NS-SEC 2 - Intermediate
[68] NS-SEC 1 - Higher Managerial, Administrative & Professionals
[69] Level 0 - no qualification or pre-school leaving qualification
[70] Level 1 - O grade, standard grade, GCSE or equivalent
[71] Level 2 - Higher grade, A level, GSVQ advanced HNC, HND, SVQ Levels 4 or 5 or equivalent
[72] Level 3 - first degree, higher degree or professional qualification

Explanatory	N	% caries	Unadjusted OR	Adjusted OR [95%
Housing Tenure				
Rented- not	[31/64]	48.4	2.98 [1.45 to 6.12]	
Rented-Privately	[7/24]	29.2	1.30 [0.47 to 3.64]	
Owned/mortgaged	[18/75]	24	1 [ref]	
P value			0.01	
C-index [95% CI]		0.63 [0.54 to 0.72]		

Unadjusted and adjusted Odds Ratios & 95% Confidence Intervals for caries experience and C-index (SWEEP 1) according to Parental Attitudes and Beliefs

Explanatory Variables	Unadjusted OR [95% CI]	Adjusted OR [95% CI][75]
Positive Parental attitudes Scale	0.41 [0.22 to 0.78]	0.10 [0.01 to 0.71]
P value	0.007	0.021
C index	0.65 [0.56 to 0.73]	
Parental Efficacy scale	0.75 [0.50 to 1.12]	4.81 [1.41 to 16.41]
P value	0.164	0.012
C index	0.60 [0.50 to 0.69]	

[73] Fully adjusted model

[74] Rented from local Authority/Council/Housing association

[75] Fully adjusted model

Unadjusted and adjusted Odds Ratios & 95% Confidence Intervals for caries experience and C-index (SWEEP 1) according to Early feeding habits

Explanatory	N	% caries	Unadjusted OR	Adjusted OR [95%
Breast or bottle feeding				
Breast fed only	[12/36]	33.3	1 [ref]	
Bottle fed only	[37/94]	39.4	1.30 [0.58 to 2.91]	
Breast and	[8/35]	22.9	0.59 [0.21 to 1.69]	
P value			0.22	
C-index [95% CI]			0.57 [0.48 to 0.66]	
Age bottle feeding completely stopped				
Never used a	[3/12]	25	0.73 [0.18 to 2.93]	2.47 [0.34 to
Under 12	[17/31]	54.8	2.64 [1.11 to 6.26]	2.97 [0.91 to
Between 1 yr	[23/73]	31.5	1 [ref]	1 [ref]
Over 2 years or	[14/49]	28.6	0.87 [0.39 to 1.92]	0.57 [0.19 to
P value			0.076	0.082
C-index [95% CI]			0.60 [0.51 to 0.69]	
Night time bottle feeding up to 2 years				
Every/ Most	[23/62]	37.1	2.08 [0.99 to 4.39]	2.83 [1.04 to
Some days	[16/25]	64	6.28 [2.36 to	2.77 [0.75 to
Hardly ever/	[17/77]	22.1	1 [ref]	1 [ref]
P value			0.001	0.085
C-index [95% CI]			0.66 [0.57 to 0.75]	
Frequency sugar restricted				
Every/ Most	[34/120]	28.3	1 [ref]	
Some days	[16/32]	50.0	2.53 [1.14 to 5.62]	
Hardly ever/	[7/13]	53.9	2.95 [0.93 to 9.42]	
P value			0.026	
C-index [95% CI]			0.60 [0.51 to 0.70]	
Night time bottle feeding up to 4 years				
Every/ Most	[10/30]	33.3	0.99 [0.42 to 2.32]	
Some days	[9/20]	45.0	1.62 [0.62 to 4.23]	
Hardly ever/	[38/113]	33.6	1 [ref]	
P value			0.608	
C-index [95% CI]			0.53 [0.43 to 0.62]	
Age of commencement of solids				
< 4 months	[4/16]	25	0.84 [0.25 to 2.80]	0.31 [0.06 to
4-6 months	[31/109]	28.4	1 [ref]	1 [ref]
7 months and	[22/39]	56.4	3.26 [1.53 to 6.95]	2.90 [0.97 to
P value			0.006	0.041
C-index [95% CI]			0.62 [0.53 to 0.71]	

[76] Fully adjusted model

Unadjusted and adjusted Odds Ratios & 95% Confidence Intervals for caries experience and C-index (SWEEP 1) according to Current Diet

Explanatory Variables	N	% caries experience	Unadjusted OR [95% CI]	Adjusted OR [95% CI][77]
Current Diet				
NMES<11% food energy	[9/32]	28.1	1 [ref]	
NMES>=11% food energy	[47/129]	36.4	1.47 [0.63 to 3.43]	
P value			0.379	
C-index [95% CI]		0.53 [0.44 to 0.62]		
NMES (% food energy)			1.06 [1.01 to 1.11]	1.06 [1.00 to 1.12]
P value			0.024	0.053
C-index [95% CI]			0.59 [0.50 to 0.69]	
Crisps & Savoury Snacks (g/day)				
T1: <=9.63	[17/59]	28.8	1 [ref]	
T2: 9.64 to 18.38	[17/47]	36.2	1.40 [0.62 to 3.18]	
T3: 18.39 & above	[20/49]	40.8	1.70 [0.77 to 3.80]	
P value			0.419	
C-index [95% CI]			0.56 [0.47 to 0.66]	
Biscuits, Cakes & Pastries (g/day)				
T1: <=20.84	[17/51]	33.3	1 [ref]	
T2: 20.85 to 36.93	[19/51]	37.3	1.19 [0.53 to 2.68]	
T3: 36.94 & above	[17/51]	33.3	1.00 [0.44 to 2.28]	
P value			0.891	
C-index [95% CI]			0.52 [0.42 to 0.62]	
Confectionary (g/day)				
T1: <= 10.69	[16/49]	32.7	1 (ref)	
T2: 10.70 to 23.81	[21/58]	36.2	1.17 [0.53 to 2.61]	
T3: 23.82 & above	[18/51]	35.3	1.13 [0.49 to 2.58]	
P value			0.925	
C-index [95% CI]			0.52 [0.42 to 0.61]	
Non-Diet Soft Drinks (g/day)				
T1: <= 70.0	[13/57]	22.8	1 [ref]	
T2: 70.01 to 161.35	[17/41]	41.5	2.40 [1.00 to 5.76]	
T3: 161.36 & above	[22/48]	45.8	2.86 [1.24 to 6.63]	
P value			0.036	
C-index [95% CI]			0.62 [0.52 to 0.71]	
NMES (g/day)				
T1: <= 51.9	[16/56]	28.6	1 [ref]	
T2: 52.0 to 81.1	[19/50]	38	1.53 [0.68 to 3.46]	
T3: 81.2 & above	[21/55]	38.2	1.54 [0.70 to 3.42]	
P value			0.484	
C-index [95% CI]			0.55 [0.46 to 0.64]	
Total Sugars (g/day)				
T1: <=103.2	[19/53]	35.9	1 [ref]	
T2: 103.3 to 149.0	[19/54]	35.2	0.97 [0.44 to 2.15]	
T3: 149.1 & above	[18/54]	33.3	0.90 [0.40 to 1.99]	
P value			0.961	
C-index [95% CI]			0.51 [0.42 to 0.61]	

[77] Fully adjusted model

Unadjusted and adjusted Odds Ratios & 95% Confidence Intervals for caries experience and C-index (SWEEP 1) according to Oral hygiene practices

Explanatory Variables	N	% caries experience	Unadjusted OR [95% CI]	Adjusted OR [95% CI][78]
Daily toothbrushing frequency				
Less than twice	[20/40]	50	2.51 [1.21 to 5.23]	
Twice or more than twice	[35/123]	28.5	1[ref]	
P value			0.014	
C-index [95% CI]			0.59 [0.49 to 0.68]	
Brush before bedtime				
Always	[21/88]	23.9	1 [ref]	1 [ref]
Sometimes / Occasionally	[34/70]	48.6	3.01 [1.53 to 5.94]	4.73 [1.78 to 12.58]
Hardly ever/ Never	[2/7]	28.6	1.28 [0.23 to 7.07]	2.64 [0.30 to 23.24]
P value			0.006	0.008
C-index [95% CI]			0.63 [0.54 to 0.72]	
Age when toothbrushing started				
Under 12 months/First tooth erupted	[37/126]	29.4	1 [ref]	1 [ref]
After 12 months	[19/36]	52.8	2.69 [1.16 to 5.74]	1.64 [0.54 to 4.97]
P value			0.011	0.386
C-index [95% CI]			0.59 [0.50 to 0.68]	
Adult supervised tooth-brushing				
Yes	[50/139]	36	1 [ref]	
No	[7/26]	26.9	0.66 [0.26 to 1.67]	
P value			0.376	
C-index [95% CI]			0.53 [0.44 to 0.62]	
Method for toothpaste removal				
Swallow	[10/22]	45.5	2.13 [0.83 to 5.46]	5.04 [1.37 to 18.61]
Spit	[29/103]	28.2	1 [ref]	1 [ref]
Rinse out	[18/40]	45.0	2.09 [0.98 to 4.45]	1.84 [0.64 to 5.31]
P value			0.087	0.045
C-index [95% CI]			0.59 [0.50 to 0.68]	
Amount of toothpaste used				
Recommended: Pea-size	[34/113]	30.1	1 [ref]	
Others: Smear, Half a brush, Full brush	[23/52]	44.2	1.84 [0.93 to 3.63]	
P value			0.078	
C-index [95% CI]			0.57 [0.47 to 0.66]	

[78] Fully adjusted model

Appendix I: Standard Operation Procedures for Saliva and plaque sampling

SALIVA COLLECTION

A. THE EQUIPMENT

Salimetrics Children's Swab

The Salimetrics Children's Swab is recommended for collecting saliva samples from infants and children younger than six (6). Under ideal circumstances, collecting saliva for 60- 90 seconds yields on average >200 µL of useable sample.

B. THE PROTOCOL

1. Disinfect hands thoroughly using the hand sanitizer / disinfectant.

2. Use a fresh pair of gloves from the box of gloves. Do not use gloves that show damage or holes. Make sure that once the gloves are worn, only instruments used for sample collection and child mouth are touched. Do not attempt to maneuver the child by holding the child's head or clothes. If this happen, wear a fresh pair of gloves.

3. Remove one Salimetrics Children's Swab (SCS) from storage bag. Do not use if cuts or tears are present. Close bag immediately to protect the remaining swabs from contact with moisture. Store swabs in dry conditions. Re-introduce the swab as needed until the lower third (approx. 2in/5 cm) of the swab is saturated.

Please note: Localized secretions from specific areas in the mouth can affect results for analytes such as SIgA. It is therefore recommended that the swab be not be moved around in the mouth.

4. Immediately after collection, remove the cap from the swab storage tube and insert the **saturated** end of the swab into the labelled tube. Using sharp scissors cut off the remaining **'dry'** length (7.5 cm/3- in.) and re-cap tube.

   - **Note for transcriber/assistant:**
   Always start choosing storage tubes from left hand side bottom, moving up. Use the same code shown on the tube to label the column, *ID code* in the data collection sheet.

5. Organize the tubes into storage boxes /foam racks in order, following the continuous code.

6. Immediately after collection, storage tube should be frozen (- 20 to - 800C). If freezer is not immediately available, refrigerate samples (2- 80C) or place in cooler containing ice packs. **Freezing samples within 2 hours of collection is recommended.**

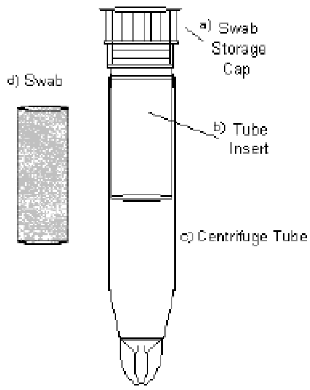
C. SALIVA EXTRACTION AND STORAGE

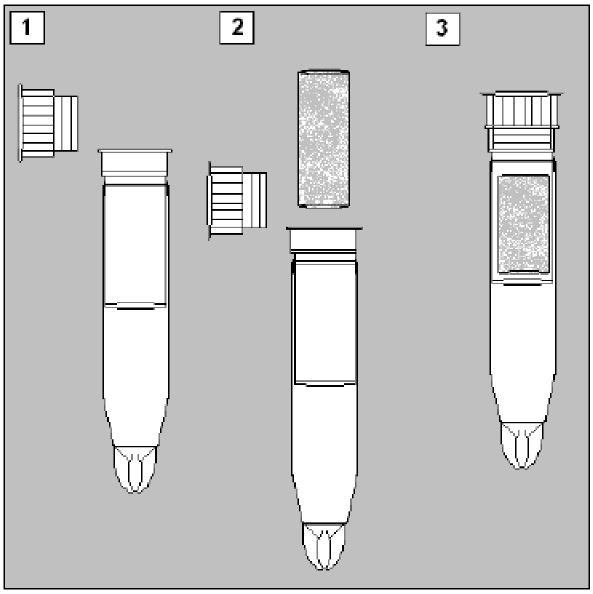
7. Centrifuge the storage tube by placing them in ALC refrigerated centrifuge PK 120 R for 10 minutes at 4100 rpm to extract the saliva. Remove the swab insert and discard. Recap the tube.
8. Pipette 100 µL into Bijoux bottles containing 900 µL of Phosphate saline buffer(PBS) and diluted serially to 10^{-3} for bacterial culturing.

9. Aliquot 50µL of saliva into a bijoux labelled for cortisol analysis and store at -70C.

10. Aliquot 50µL of saliva into a bijoux labelled for sIgA analysis and store at -70C.

11. Aliquot 100µL of saliva into x ml of lysis buffer and store at -70 C.





NOTES:

It is important that saliva collected using the Salimetrics Children's Swab (SCS) not evaporate. Make sure the cap is closed properly until you hear the click and sample is frozen **within two hours** after collection. If samples are to be stored for greater than three (3) months after collection, centrifuge **prior** to freezer storage to lessen the chance of evaporation.

PLAQUE COLLECTION

A. EQUIPMENT

CPITN probe (ball-ended probes)
Mouth mirror
Eppendorf containing 200 μL lysis buffer

B. PROTOCOL

1. Retract the cheek of the child using the mouth mirror.

2. Collect plaque using a CPITN probe from the buccal aspect (side of the teeth facing the cheek) of the interdental region (the area between the two teeth) of the molars in each quadrant.

3. Avoid trauma to the gingival tissues by keeping the instrument away from the gingivae or collecting any sub-gingival plaque.

4. **Scrape the tip of the ball-ended probe around the inner corners/edges of the eppendorf** or try to dip the end of the probe in the lysis buffer, contained within the eppendorf.

5. Close the cap of the ependorff tight (until it clicks) and shake well to disperse the plaque.

6. Place the eppendorf immediately in the ice box in a foam rack in the same order as the codes.

7. On return to GDHS, the foam rack of eppendorfs should be stored in the molecular biology lab freezer on Level 8 (opposite the CSSD lifts).

Appendix J: Steps used in culturing oral bacteria

The steps that were used in culturing bacteria are detailed in Figure 9-1

1. After saliva in extracted from Salimetrics Children's swab, vortex (Stuart scientific Anutovortex SA5) the sample for 30 seconds.

2. Pipette 100 μL into Bijoux bottles containing 900 μL of Phosphate saline buffer(PBS) and diluted serially to 10^{-3}

3. The diluted saliva in bijoux 10^{-3} and 10^{-2} are used to spiral plate in the following manner

PLATES	DILUTIONS
MSB	10^{-2} ,10^{-3}
Rogosa	10^{-2} ,10^{-3}
Bifido	10^{-2}
BA	10-^{3}
FAA	10^{-3}

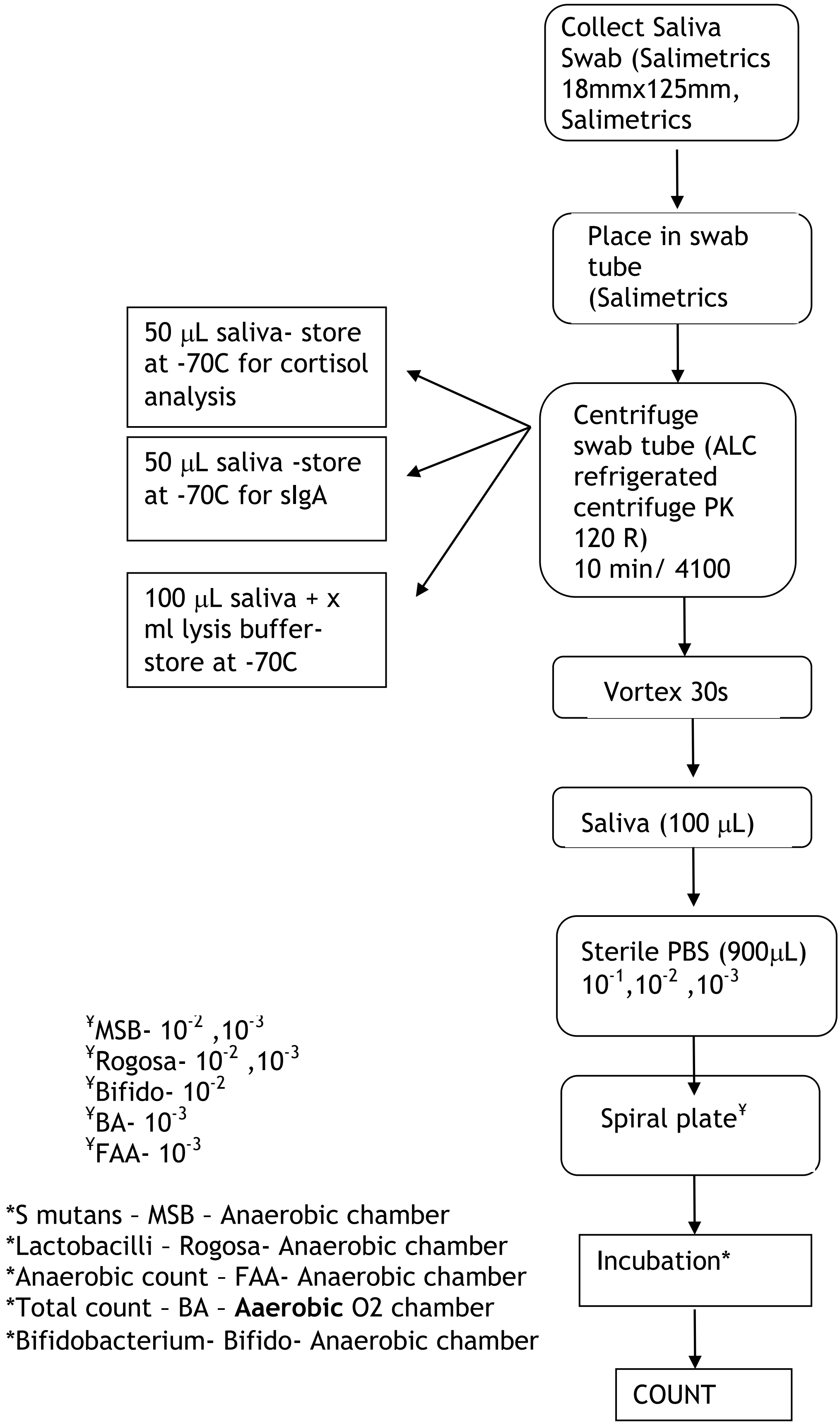


Figure 9-1- Saliva collection and processing

Appendix K: Standard Operation Procedures for anthropometric measurement

HEIGHT MEASURMENT

A. EQUIPMENT

The stadiometer used in this study is Seca 213 stand-alone portable stadiometer.

- Measuring range: 20 – 205 cm / 8 – 81"
- Graduation: 1 mm / 1/8"

B. PROTOCOL

1. Children should remove their shoes before being measured. If shoes are not removed, this should be recorded in the data collection sheet under the column, *Shoes removed.* Please inform you transcriber about this.

2. Raise the head plate to allow sufficient room for the child to stand underneath it.

3. The child should stand with their feet flat on the centre of the base plate, feet together and heels against the rod. The child's back should be as straight as possible, preferably against the rod, and their arms hanging loosely by their sides. Ideally heels, buttocks, shoulders and back of head touch the back plate. They should be facing forwards. Do not stretch up, measure on expiration.

4. Slide the headboard gently, but firmly on top of the head.

5. Position the head of the child looking straight ahead with the lower border of the bony orbit and the upper margin of the ear hole in the same horizontal line (Frankfort plane). This brings the crown of the head to be the highest point measured

6. Read out the height to the nearest millimeter to the transcriber.

WEIGHT MEASURMENT

A. THE EQUIPMENT

The scales used in this study is Seca 877 Electronic flat floor scale (Class III Approved)

- These scales display the weight in a window on the scales.
- The Seca 877 is switched on by pressing the top of the scales (e.g. with your foot).
- There is no switch to turn the scales off, they turn off automatically.
- The scales take 6 x 1.5v AA batteries

The scales have an inbuilt memory which stores the weight for 10 minutes. If during this time you weigh another object that differs in weight by less than 500 grams (about 1lb), the stored weight will be displayed and not the weight that is being measured. This means that if you weigh someone else during this time, you could be given the wrong reading for the second person.

So if you get an identical reading for a second person, make sure that the memory has been cleared. Clear the memory from the last reading by weighing an object that is more than 500 grams lighter (ie a pile of books, your briefcase or even the stadiometer). You will then get the correct weight when you weigh the second respondent. You will only need to clear the memory in this way if:

a) You have to have a second or subsequent attempt at measuring the same person.

b) Two children appear to be of a very similar weight.

B. THE PROTOCOL

1. Turn the display on by inserting the battery at the back of the scale. The readout should display Er 16 momentarily. If this is not displayed check the batteries. While the scales read Er 16 do not attempt to weigh anyone.

2. Ask the child to remove shoes, heavy outer garments such as jackets and cardigans and heavy jewellery, if any. Children should not hold or carry a teddy.

3. **Turn the scales on with your foot again**. Wait for a display of 0.0 kg before the child stands on the scales.

4. Ask the child to stand with their feet together in the centre and their heels against the back edge of the scales. Arms should be hanging loosely at their sides and head facing forward. Ensure that they keep looking ahead – children are easily distracted or sometimes tempted to look down at their weight reading. Ask them not to do this and assure them that you will tell them their weight afterwards if they want to know.

The posture of the child is important. If they stand to one side, look down, or do not otherwise have their weight evenly spread, it can affect the reading. For accurate readings, it is very important that respondents stand still. Ask the child to stand perfectly still – 'Be a statue'. **Take your hands of the child if you were attempting to stabilize the child.**

5. The scales will take a short while (3 seconds) to stabilize. If the child moves excessively while the scales are stabilizing you may get a false reading. If you think this is the case reweigh.

6. Read out the reading to the transcriber who will record the reading into the data collection sheet.

Notes:

Weighing uncooperative children:

For children who are uncooperative you may alter the protocol and first weigh an adult then weigh that adult holding the child as follows:-

Weigh the adult (eg: teacher at the nursery) as normal following the protocol as set out above and press the '2 in 1' button on the right hand side of the scale. This will show a 'NET' reading. Now the teacher may hold the child in hands and the scale will calculate and display the child's weight.

Recording measurements:

All measurements should be recorded in pencil. If a mistake is made when recording a measurement, it can be corrected.

<u>When you are storing the scales please make sure you remove the battery to stop the scales turning themselves on.</u>

Appendix L: Multivariable analysis within domain stage

Unadjusted and adjusted Odds Ratios & 95% Confidence Intervals for caries experience and C-index (SWEEP 2) according to demographic variables

Explanatory Variables	N	% caries experience	Unadjusted OR [95% CI]	Adjusted OR [95% CI][1]
Child characteristics				
Gender				
Male	[46/94]	48.9	1 [ref]	1 [ref]
Female	[38/80]	47.5	0.94 [0.52 to 1.72]	0.80 [0.41 to 1.57]
P value			0.850	0.521
C-index [95% CI]			0.51 [0.42 to 0.60]	0.61 [0.52 to 0.70][2]
Age			0.71 [0.30 to 1.70]	0.71 [0.27 to 1.86]
P value			0.439	0.484
C-index [95% CI]			0.54 [0.45 to 0.62]	
Main Carer marital Status				
Married/Cohabiting	[45/104]	43.3	1 [ref]	1 [ref]
Single parent	[23/41]	56.1	1.68 [0.81 to 3.47]	1.57 [0.74 to 3.34]
P value			0.165	0.244
C-index [95% CI]			0.55 [0.46 to 0.65]	
Main-Carer smoking status				
Current Smoker	[23/40]	57.5	1.77 [0.85 to 3.68]	1.46 [0.68 to 3.16]
Not current smoker	[46/106]	43.4	1 [ref]	1 [ref]
P value			0.130	0.333
C-index [95% CI]			0.56 [0.46 to 0.65]	

[1] Multivariate model adjusted for variables within the demographic domain

[2] C- index for the multivariable model within demographic domain

Unadjusted and adjusted Odds Ratios & 95% Confidence Intervals for caries experience and C-index (SWEEP 2) according to Socioeconomic Position

Explanatory Variables	N	%	Unadjusted OR [95% CI]	Adjusted OR [95% CI][1]
National SIMD[2] Quintile				
Q1 (most deprived)	[58/100]	58.0	3.45 [1.24 to 9.64]	3.04 [0.56 to 16.52]
Q2	[6/20]	30.0	1.07 [0.28 to 4.12]	0.24 [0.02 to 3.19]
Q3	[12/31]	38.7	1.58 [0.48 to 5.20]	1.03 [0.16 to 6.58]
Q4 & Q5 (least deprived)	[6/21]	28.6	1 [ref]	1 [ref]
P value			0.015	0.182
C-index [95% CI]			0.63 [0.55 to 0.71]	0.79 [0.69 to 0.88][3]
Gross annual household Income				
< £10,000	[22/34]	64.7	4.07 [1.66 to 9.99]	1.58 [0.23 to 10.86]
£10,000 - £19,999	[23/43]	53.5	2.56 [1.13 to 5.79]	0.49 [0.09 to 2.63]
£20,000 & more	[18/58]	31.0	1 [ref]	1 [ref]
P value			0.005	0.275
C-index [95% CI]			0.65 [0.56 to 0.74]	
Percentage of income from benefits				
Half or more	[38/59]	64.4	3.98 [1.95 to 8.12]	1.63 [0.37 to 7.21]
None or About a quarter	[25/80]	31.3	1 [ref]	1 [ref]
P value			<0.001	0.518
C-index [95% CI]			0.66 [0.57-0.76]	
Current or last occupation of Main-Carer				
Never worked or permanently	[5/9]	55.6	2.86 [0.59 to 13.96]	0.96 [0.08 to 11.87]
NS-SEC 3[4]	[27/43]	62.8	3.86 [1.31 to 11.38]	1.14 [0.20 to 6.51]
NS-SEC 2[5]	[7/26]	26.9	0.84 [0.24 to 2.91]	0.44 [0.08 to 2.34]
NS-SEC 1[6]	[7/23]	30.4	1 [ref]	1 [ref]
P value			0.013	0.543
C-index [95% CI]			0.68 [0.57 to 0.78]	
Main-Carer's highest Level of Education				
Level 0[7]	[14/19]	73.7	8.40 [2.36 to 29.90]	3.03 [0.32 to 28.46]
Level 1[8]	[21/42]	50.0	3.00 [1.14 to 7.90]	1.18 [0.16 to 8.90]
Level 2[9]	[24/48]	50.0	3.00 [1.17 to 7.70]	2.25 [0.44 to 11.49]
Level 3[10]	[9/36]	25.0	1 [ref]	1 [ref]
P value			0.009	0.521
C-index [95% CI]			0.65 [0.56 to 0.74]	
Housing Tenure				
Rented- not private[11]	[40/60]	66.7	3.91 [1.88 to 8.16]	0.91 [0.16 to 5.29]
Rented- privately	[5/17]	29.4	0.82 [0.26 to 2.60]	0.75 [0.09 to 5.91]
Owned/mortgaged	[23/68]	33.8	1 [ref]	1 [ref]
P value			<0.001	0.961
C-index [95% CI]		0.67		

[1] Adjusted for variables within the socioeconomic position domain

[2] Scottish Index of Multiple Deprivation 2009 at National level

[3] C- index for the multivariable model within the socioeconomic position domain

[4] NS-SEC 3 - Routine & manual

[5] NS-SEC 2 - Intermediate

[6] NS-SEC 1 - Higher Managerial, Administrative & Professionals

[7] Level 0 - no qualification or pre-school leaving qualification

[8] Level 1 - O grade, standard grade, GCSE or equivalent

[9] Level 2 - Higher grade, A level, GSVQ advanced HNC, HND, SVQ Levels 4 or 5 or equivalent

[10] Level 3 - first degree, higher degree or professional qualification

[11] Rented from local Authority/Council/Housing association

Unadjusted and adjusted Odds Ratios & 95% Confidence Intervals for caries experience and C-index (SWEEP 2) according to Early feeding habits

Explanatory Variables	N	% caries	Unadjusted OR [95% CI]	Adjusted OR [95% CI][1]
Breast fed only	[13/32]	40.6	1 [ref]	1 [ref]
Bottle fed only	[46/82]	56.1	1.87 [0.82 to 4.28]	1.28 [0.44 to 3.75]
Breast and bottle-fed	[11/33]	33.3	0.73 [0.27 to 2.01]	0.38 [0.10 to 1.39]
P value			0.061	0.066
C-index [95% CI]			0.60 [0.51 to 0.69]	0.77 [0.70 to 0.85][2]
Age bottle feeding completely stopped				
Never used a baby-bottle	[3/11]	27.3	0.56 [0.14 to 2.28]	0.56 [0.10 to 3.18]
Under 12 months	[19/27]	70.4	3.52 [1.35 to 9.19]	4.25 [1.41 to 12.81]
Between 1 yr and 2 yrs	[27/67]	40.3	1 [ref]	1 [ref]
Over 2 years or Still using	[21/42]	50.0	1.48 [0.68 to 3.22]	0.68 [0.25 to 1.82]
P value			0.038	0.029
C-index [95% CI]			0.63 [0.54 to 0.72]	
Night time bottle feeding up to 2 years				
Every/ Most days	[30/55]	54.6	2.08 [1.01 to 4.26]	2.41 [0.98 to 5.93]
Some days	[13/20]	65.0	3.21 [1.14 to 9.08]	1.60 [0.48 to 5.32]
Hardly ever/ Never	[26/71]	36.6	1 [ref]	1 [ref]
P value			0.034	0.155
C-index [95% CI]			0.62 [0.52 to 0.71]	
Frequency sugar restricted				
Every/ Most days	[42/107]	39.3	1 [ref]	1 [ref]
Some days	[19/29]	65.5	6.96 [1.43 to 33.83]	2.41 [0.89 to 6.56]
Hardly ever/ Never	[9/11]	81.8	2.94 [1.25 to 6.94]	14.32 [1.41 to 145.33]
P value			0.005	0.027
C-index [95% CI]			0.63 [0.54 to 0.72]	
Night time bottle feeding up to 4 years				
Every/ Most days	[14/22]	63.6	2.31 [0.89 to 5.98]	2.18 [0.61 to 7.85]
Some days	[12/21]	57.1	1.76 [0.68 to 4.54]	0.88 [0.29 to 2.64]
Hardly ever/ Never	[44/102]	43.1	1 [ref]	1 [ref]
P value			0.154	0.445
C-index [95% CI]			0.58 [0.48 to 0.67]	
Age of commencement of solids				
< 4 months	[7/14]	50.0	1.39 [0.45 to 4.27]	1.24 [0.32 to 4.75]
4-6 months	[41/98]	41.8	1 [ref]	1 [ref]
7 months and over	[22/34]	64.7	2.55 [1.13 to 5.73]	2.25 [0.84 to 6.06]
P value			0.076	0.271
C-index [95% CI]			0.59 [0.50 to 0.68]	

[1] Adjusted for variables within the early feeding habits domain

[2] C- index for the multivariable model within the early feeding habits domain

Unadjusted and adjusted Odds Ratios & 95% Confidence Intervals for caries experience and C-index (SWEEP 2) according to Current Diet

Explanatory Variables	N	% caries	Unadjusted OR [95%	Adjusted OR [95% CI][1]
Current Diet				
NMES<11% food energy	[10/28]	35.7	1 [ref]	1 [ref]
NMES>=11% food energy	[59/117]	50.4	1.83 [0.78 to 4.30]	0.87 [0.29 to 2.61]
P value			0.165	0.798
C-index [95% CI]			0.55 [0.45 to 0.64]	0.65 [0.56 to 0.74][2]
NMES (% food energy)			1.09 [1.02 to 1.16]	1.11 [1.02 to 1.20]
P value			0.007	0.018
C-index [95% CI]			0.60 [0.51 to 0.69]	
Crisps & Savoury Snacks (g/day)				
T1: <=9.63	[21/51]	41.2	1 [ref]	1 [ref]
T2: 9.64 to 18.38	[18/43]	41.9	1.03 [0.45 to 2.34]	1.05 [0.44 to 2.50]
T3: 18.39 & above	[29/48]	60.4	2.18 [0.98 to 4.87]	1.90 [0.81 to 4.45]
P value			0.106	0.258
C-index [95% CI]			0.59 [0.49 to 0.68]	

Unadjusted and adjusted Odds Ratios & 95% Confidence Intervals for caries experience and C-index (SWEEP 2) according to Parental Attitudes and Beliefs

Explanatory Variables	Unadjusted OR [95% CI]	Adjusted OR [95% CI]
Positive Parental attitudes Scale	0.54 [0.28 to 1.06]	0.43 [0.10 to 1.78]
P value	0.072	0.243
C index	0.60 [0.51 to 0.69]	0.60 [0.50 to 0.69] [133]
Parental Efficacy scale	0.74 [0.49 to 1.12]	1.18 [0.49 to2.84]
P value	0.155	0.715
C index	0.59 [0.50 to 0.69]	

[1] Adjusted for variables within the current diet domain

[2] C- index for the multivariable model within the current diet domain

Unadjusted and adjusted Odds Ratios & 95% Confidence Intervals for caries experience and C-index (SWEEP 2) according to Oral hygiene practices

Explanatory Variables	N	% caries	Unadjusted OR [95% CI]	Adjusted OR [95% CI][1]
Daily toothrushing frequency				
Less than twice	[24/34]	70.6	3.50 [1.53 to 8.00]	2.04 [0.76 to 5.42]
Twice or more than twice	[46/113]	40.7	1[ref]	1 [ref]
P value			0.003	0.155
C-index [95% CI]			0.61 [0.51 to 0.70]	0.73 [0.65 to 0.81][2]
Brush before bedtime				
Always	[28/80]	35.0	1 [ref]	1 [ref]
Sometimes / Occasionally	[39/61]	63.9	3.29 [1.64 to 6.60]	2.59 [1.15 to 5.81]
Hardly ever/ Never	[3/6]	50.0	1.86 [0.35 to 9.82]	1.19 [0.19 to 7.55]
P value			0.004	0.061
C-index [95% CI]			0.64 [0.55 to 0.73]	
Age when toothbrushing started				
Under 12 months/First tooth erupted	[51/116]	44.0	1 [ref]	1 [ref]
After 12 months	[19/30]	63.3	2.20 [0.96 to 5.04]	1.50 [0.59 to 3.82]
P value			0.062	0.395
C-index [95% CI]			0.56 [0.47 to 0.66]	
Adult supervised tooth-brushing				
Yes	[62/125]	49.6	1 [ref]	
No	[8/22]	36.4	0.58 [0.23 to 1.48]	
P value			0.255	
C-index [95% CI]			0.53 [0.44 to 0.62]	
Method for toothpaste removal				
Swallow	[12/21]	57.1	2.26 [0.86 to 5.94]	2.96 [1.03 to 8.50]
Spit	[33/89]	37.1	1 [ref]	1 [ref]
Rinse out	[25/37]	67.6	3.54 [1.57 to 7.96]	3.20 [1.34 to 7.64]
P value			0.006	0.011
C-index [95% CI]			0.64 [0.55 to 0.73]	
Amount of toothpaste used				
Recommended: Pea-size	[46/100]	46.0	1 [ref]	
Others: Smear, Half a brush, Full	[24/47]	51.1	1.23 [0.61 to 2.45]	
P value			0.567	
C-index [95% CI]			0.52 [0.43 to 0.62]	

[1] Adjusted for variables within the oral hygiene practices domain

[2] C- index for the multivariable model within the oral hygiene practices domain

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