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Prevalence, Incidence and Risk Factors for Early Childhood Caries Among Young African-American Children in Alabama

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PREVALENCE, INCIDENCE AND RISK FACTORS FOR EARLY CHILDHOOD CARIES AMONG YOUNG AFRICAN-AMERICAN CHILDREN IN ALABAMA

by

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CERTIFICATE OF APPROVAL

MASTER'S THESIS

This is to certify that the Master's thesis of

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for the thesis requirement for the Master of Science
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To My Parents and Siblings (Zaid, Meena and Harith)

In seeking wisdom, the first step is silence, the second listening, the third remembering,
the fourth practicing, the fifth -- teaching others

Solomon Ibn Gabirol
The Fountain of Life

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TABLE OF CONTENTS

LIST OF TABLES

Table

CHAPTER 1

INTRODUCTION

Dental caries experience of primary teeth in children under age 72 months, or what is known as Early Childhood Caries (ECC), is considered one of the most important, ongoing public health problems, not only in the United States of America, but throughout the whole world. Many statistics show that ECC is a major dental public health problem in the United States. For example, National Health and Nutrition Examination Survey III (NHANES III) data from 1988-1994 showed that about 8.4% of the children who were 2 years old and 40.0% who were 5 years old had at least one decayed or filled tooth[1]. Although the prevalence of dental caries experience among the U.S. population, including children, has decreased drastically in the last fifty years, ECC still presents a serious threat to child welfare[2].

National statistics in the United States show that ECC is most prevalent among children who are from low socioeconomic status families[3]. Furthermore, the findings of the national surveys demonstrate that ECC is more prevalent among specific racial and ethnic groups, such as African-Americans, Hispanics, and Native-Americans[3].

ECC is a relatively new term, used after it was first recommended by the National Institute of Dental and Craniofacial Research in 1999 to replace the old names that described its etiology, such as nursing caries and baby bottle booth decay[4]. ECC is a chronic, infectious, transmissible, and multifactorial disease that affects about 1% to 17% of preschool children in the developed countries and up to 70% of preschool children in the developing countries[5].

There are a limited number of studies that have investigated the prevalence and incidence of ECC among high-risk groups, in part because of difficulties related to recruitment and high attrition in case of longitudinal studies. In addition, there are relatively few studies which have been conducted to identify the effects of different risk factors associated with ECC. The most commonly known risk factors are low SES, lack of dental insurance, high-sugared snack intake, improper bottle use, higher levels of salivary *Streptococcus mutans* bacteria, and poor oral hygiene. However, the findings of these studies differ, in part because of different study designs, statistical analysis techniques, definitions of different dependent and independent variables, children's ages, and the presence of confounders, such as lack of dental coverage.

Because of the limited number of studies that have addressed the problem of ECC and the varied results, there is a need for new studies that investigate the different issues that are related to this "distressing syndrome", which starts most commonly in children from 1-3 years of age, and often is characterized by carious lesions on the maxillary deciduous teeth[6].

Since ECC is most common among specific racial and ethnic groups, this report will investigate this problem among a cohort of 3 to 22 month old African-American children at baseline and follow then for three years, who live in what is termed the "Black Belt" counties in Alabama. Worthy of mentioning, the word black in the term "Black Belt" refers to the color of the soil, which is a very rich and fertile. Although these counties are very rich in their agrarian resources, the people who live there are, as a group, the poorest people in the United States[7]. In 2012, the Institute of Rural Health Research at the University of Alabama stated that the children who live in the rural Alabaman counties suffer from serious health disparities, and have inadequate schools and health services[8].

In this study, data from an ongoing longitudinal study in Alabama were used and analyzed. This original "parent" study was designed to investigate the relationship between the acquisition of bacteria associated with the eruption of first primary molars in high-risk African-American children and the pathogenesis of dental caries. The purposes of this sub-study, which will involve collaboration between the School of Dentistry at the University of Alabama at Birmingham (UAB) and the College of Dentistry at the

University of Iowa, were to assess the prevalence and incidence of ECC, and the relationships between different risk factors and ECC in young African-American, Alabaman children aged 3 to 22 months at baseline.

The study of the prevalence of ECC among the enrolled children is valuable because it shows the percentage of the high risk, Alabaman children who had ECC and could be used in the future as a reference for other studies in other counties or states. Also, the determination of ECC incidence and caries increment will help us assess the risk of new caries among children within a specific time. Thus, it will give us insight into the progression of early lesions and the development of new carious lesions among these children. There are few studies that have assessed dental caries incidence among young children, and, as far as we know, there is no other study that has assessed the incidence of ECC among very young African-Americans.

In addition to the assessment of prevalence and incidence of ECC, this report will assess the relationships between ECC and several behavioral risk factors, such as the consumption of natural juice, sugar-added beverages and infant formula, and demographic risk indicators, such as age and sex. Also, the relationships between ECC and children's previous caries experience and their parents' caries experience will be assessed. Study of the effects of different behavioral risk factors and demographic risk indicators is also essential, since ECC is still an uncontrolled disease, and additional information is needed to understand the complex associations of risk factors with the development of ECC.

In Chapter Two (Literature Review), a review of the previously conducted studies which are relevant to the main themes of the study will be discussed in detail. The detailed findings and the main conclusions of these studies will be presented, giving the reader a general picture of what has been found previously and the main limitations which were associated with the design and the conduct of these studies.

CHAPTER 2

LITERATURE REVIEW

Introduction

The National Institute of Dental and Craniofacial Research recommended the use of the term Early Childhood Caries (ECC) in 1999. Prior to that, other terms that described the etiology of ECC were used, such as rampant caries, nursing caries, and baby bottle tooth decay[9].

A limited number of studies have been conducted to investigate the prevalence and incidence of ECC among high-risk groups as a result of difficulties in recruitment and assurance of the compliance of the study subjects. Studies that assess the relationship between ECC and risk factors, such as low SES, lack of dental insurance, high-sugared snack intake, improper bottle use, higher levels of salivary *Streptococcus mutans* bacteria, and poor oral hygiene are also limited and have had different findings because of different study designs, children's age, statistical techniques, definitions of dependent and independent variables, children's ages, and confounders, such as the presence of dental coverage.

This chapter will review the most relevant prevalence, incidence, and risk factors studies concerning Early Childhood Caries, which have been conducted in the United States and elsewhere. The main findings, conclusions, limitations and strengths of these studies will be discussed thoroughly in different sections of this chapter.

Definitions

Early Childhood Caries is the terminology that was recommended by the National Institute of Dental and Craniofacial Research workshop in 1999 to replace the old names that described its etiology, such as rampant caries, nursing caries, and baby bottle booth decay[9]. The American Academy of Pediatric Dentistry (AAPD) defines ECC as the presence of one or more decayed (non-cavitated or cavitated lesions), missing (due to caries), or filled tooth surfaces in any primary tooth in a child 71 months of age or younger[10]. Severe-ECC, on the other hand, is defined by the AAPD as the presence of smooth-surface caries in children younger than three years of age, or the presence of one or more cavitated, missing (due to caries), or filled smooth surfaces in primary maxillary anterior teeth or a decayed, missing, or filled score of ≥4 (age 3), ≥5 (age 4), or ≥6 (age 5) from age 3 through 5[10].

Non-cavitated lesions are also called d1 lesions. They are white chalky lesions that lack any visible loss of tooth structure, and they include demineralization of enamel, dentine, or both[11]. These lesions might develop into cavitated lesions, but can also remain unchanged or reverse to sound structure. That is why the word “non-cavitated” is preferred over “precavitated”, since the latter indicates that these lesions are going to develop into cavitated lesions in the future[11].

Cavitated lesions, or d2/d3 lesions, are carious lesions that include a break in the tooth structure. If they are confined to the enamel surface, then they are called d2, while if they include the dentine, then they are called d3[12].

Impact of ECC

Worldwide statistics show that a considerable percentage of children younger than age six years suffered from Early Childhood Caries[13]. In the United States, the National Health and Nutrition Examination Survey III (NHANESIII), 1988-1994, showed that 8.4% of the children who were two years old and 40% who were five years old had at least one filled or decayed tooth[14]. Also, the prevalence of ECC was higher among specific risk groups, such as children from low SES families, and those who were from specific racial and ethnic backgrounds, such as Hispanics, Native Americans, and African Americans. Recent data from NHANES, 1999-2002 showed that the prevalence of ECC among children aged 2, 3, 4, and 5 years was 10.9%, 20.9%, 34.4%, and 44.3%, respectively[15]. For example, in a study conducted by Currier-et al.[16], the prevalence of rampant caries among predominantly (98%) African-American children was assessed. Two-hundred and forty-six children aged 4 weeks to 9 years, divided into 6 age groups, as shown in Table 1, were recruited from Jefferson Maternal and Child Health Clinic (WIC) of the city of Richmond, Virginia, and examined over 10 weeks by the authors. All the children had dental examinations without the use of x-rays, mirror, or explorer[16]. The findings published in 1977 showed that 180 children had anterior teeth, 20 of them (11%) had dental caries on the anterior teeth (Table 1) and nine (5%) were diagnosed as having "Baby Bottle Syndrome", which was defined as the presence of dental caries on the labial or palatal of two or more maxillary incisors of children who had a history of using the bottle for feeding and pacification of children.

Table 1. Children with caries on the anterior teeth[16]

Age group	Total number of children	Total number of children with at least one anterior tooth erupted	Number of children with anterior tooth caries	Prevalence of anterior caries among children with at least one anterior teeth erupted
1-5 months	51	0	0	undetermined
6-11 months	23	8	0	0%
12-17 months	27	27	0	0%
18-35 months	55	55	5	9%
36-59 months	79	79	11	14%
60-108 months	11	11	4	36%
Total	246	180	20	11%

In addition to the effects of ECC on the child's overall health and oral health quality of life (OHQoL)[17], ECC is considered one of the most costly diseases in both developed and developing countries. It is reported that the annual cost of the overall dental care was over $108 billion in the U.S. in 2010, and there is no doubt that the cost of treating ECC constitutes a considerable portion of all dental care cost[18]. It is worthy to mention that, although many of the risk factors associated with ECC are well-known, there is still a need to investigate its incidence, prevalence, and risk factors further and among specific risk groups.

Prevalence of ECC

Definition

Prevalence, or what is sometimes known as the prevalence rate, is the proportion of a group of people that have a specific clinical condition at a given time[19]. The study of the prevalence of ECC is important because it provides a snapshot and allows comparisons between different study samples and potential risk groups. The prevalence of ECC is different in various places and groups around the world.

Prevalence of ECC Studies

Milnes'[20] conducted a review study of "all English-language articles that reported the prevalence of cavitated caries (d2 and d3) involving the primary maxillary incisors in preschool children." This article included many tables that contained summaries of studies that have been conducted in different places in the world. Table 1 contained prevalence studies from some countries in Europe, such as England, Sweden, and Finland with the description of the samples and definitions of nursing caries. The prevalence of nursing caries ranged from 1%-12% (Table 2). This wide disparity in the findings of these studies was attributed in part to different definitions of nursing caries, samples, study designs, and ages of children.

Table 2. Studies of nursing caries prevalence from Europe[20]

Country	Year	Investigators	Sample	Nursing Caries Criteria	Nursing Caries Prevalence
England	1967	Goose[21]	Random selection of 309 1- to 2-year-olds in 2 counties-pilot study to 1968 study by Goose and Gittus[41]	Comparison of child's mouth by health visitors to photographs of caries of labial surfaces of maxillary incisors in mild and advanced stages	6.8%
England and Wales	1968	Goose and Gittus[22]	Random selection of 5,549 1- to 2-year old children from 72 locations in England and wales	Comparison of child's mouth by health visitors to photographs of caries of labial surfaces of maxillary incisors in mild and advanced stages	5.9%
England	1966	Winter, Hamilton, James[23]	100 1- to 5-year-old children attending a welfare center; study assessed the comforter's role in rampant caries	Visual exam by dentist; caries diagnosed when cavity with dentin involvement was observed; rampant caries recorded when labial or palatal surfaces of 2 or more maxillary incisors involved	12.0%
England	1971	Winter, Rule, Mailer, James, Gordon[24]	601 12- to 60-month-old children from all social classes in London borough of Camden who were attending maternal and child welfare clinics	Visual exam by dentist; caries diagnosed when cavity with dentin involvement was observed; rampant caries recorded when labial or palatal surfaces of 2 or more maxillary incisors involved	8.0%
England	1973	Silver[25]	263 3-year-old children representing 81% of all 3-year-olds in Bishop's Storford	Labial (rampant) caries diagnosed if involvement of labial/palatal surfaces of 2 or more maxillary incisors	8.0%
England	1981	Silver[26]	252 3-year-old children representing 78% of all 3-year-olds in Bishop's Storford	Labial (rampant) caries diagnosed if involvement of labial/palatal surfaces of 2 or more maxillary incisors	1.0%

Table 2-Continued

England	1982	Holt, Joels, Winter[27]	555 12- to 60-month-old children from all social classes in London boroughs of Camden and Islington who were attending maternal and child welfare clinics for routine medical assessment	Visual exam by dentist; caries diagnosed when cavity with dentin involvement was observed; rampant caries recorded when labial or palatal surfaces of 2 or more maxillary incisors involved	3.0%
England	1982	Holt, Joels, Winter[27]	555 12- to 60-month-old children from all social classes in London boroughs of Camden and Islington who were attending maternal and child welfare clinics for routine medical assessment	Visual exam by dentist; caries diagnosed when cavity with dentin involvement was observed; rampant caries recorded when labial or palatal surfaces of 2 or more maxillary incisors involved	3.0%
England	1988	Holt, Joels, Bulman, Maddick[28]	565 12- to 60-month-old children attending maternal and child welfare clinics in Camden	Visual exam by dentist; caries diagnosed when cavity with dentin involvement was observed; rampant caries recorded when labial or palatal surfaces of 2 or more maxillary incisors involved	7.0%
England	1989	Silver[29]	230 3-year-old children representing 61% of all 3-year-olds in Bishop's Storford	Labial (rampant) caries diagnosed if involvement of labial/palatal surfaces of 2 or more maxillary incisors	4.0%
Sweden	1991	Wendt, Hallonsten, Koch[30,31]	632 12- to 14-month-old children living within the area of 4 child welfare centers in Jonkoping. A year later, 299 of these children were re-examined	Presence or absence at initial demineralization and cavitation	7.7%
Finland	1993	Paunio, Rautava, Helenius, Alanen, Sillanpaa[32]	1,018 3-year-old children selected by means of a stratified cluster sampling of all primigravid women in provinces of Turku and Pori	Extent of caries recorded for each tooth; caries of maxillary incisors alone or in combination with canines/molars; initial caries excluded	6.0%

Goose[21] conducted a pilot study about the prevalence of nursing caries, which was defined as the presence of cavitated lesions on the labial or palatal surfaces of the anterior maxillary teeth. The results of this study published in 1967 showed that the prevalence of nursing caries was 6.8% in 309 1- to 2-year-olds who were randomly selected from two different places in England. This study was extended by Goose et al.[22] and included more children (5,549) aged 1-2 years from 72 different places in England and Wales. In this latter study, which was published in 1968, the prevalence of nursing caries was 5.9%. One of the drawbacks of this study was that the dental examinations were done by untrained "health visitors", and relied on comparing the clinical cases with photographs of mild and severe nursing caries.

In London, a series of studies were conducted to assess the prevalence of nursing caries24,27,28. Three different samples of children 12 to 60 months of age from different socioeconomic classes were enrolled from maternal and childhood welfare clinics in the London boroughs of Camden in 1971, 1982, and 198824,27,28. In these studies, 601, 555, and 565 children were enrolled and visually examined by dentists, respectively. Children with caries on the labial or palatal surfaces of 2 or more maxillary incisors were diagnosed as having nursing caries. The findings of these studies showed that the prevalence of nursing caries was 8%, 3%, and 7% in 1971, 1982, and 1988, respectively, for ages 12 to 60 months. The increase in the prevalence of nursing caries in the last study was thought to be due to the use of sweetened pacifiers.

Another series of studies involving three different samples of 3-year-old children from Bishop's Storford near London were enrolled in 1973, 1981, and 198925,26,29. In these studies, 263, 252, and 230 children were enrolled, respectively. Children with caries on the labial or palatal surfaces of 2 or more maxillary incisors were diagnosed as having rampant caries. The findings of these studies showed that the prevalence of rampant caries was 8%, 1%, and 4% in 1973, 1981, and 1989, respectively. The increase in the prevalence of nursing caries in the last study29 in 1989 compared to the 1981 study26 also was thought to be due to the use of sweetened pacifiers.

Wendt et al.[30] conducted an epidemiological study to assess the prevalence of caries among one- and two-year-old Swedish children. In their study, 671 children aged 1 year were invited from four child welfare centers in Jonkoping, and 632 participated and had their dental examinations. Dental caries was defined at the cavitated and non-cavitated level. The results of this study published in 1991 showed that the prevalence of dental caries was 0.5% among 1-year old children. One year later, 326 children were selected randomly from the 632 children who had dental exams at age one. All these randomly selected children (age=2 years) had dental examinations (non-cavitated lesions included) and the prevalence of dental caries was 7.7%.

In Finland, Paunio et al.32 assessed the prevalence of nursing caries among 1,018 3-year-old children. A stratified cluster sample of all mothers who had their first visit to the maternity health care clinics was obtained. Children with caries on maxillary incisors alone or in combination with canines were diagnosed as having nursing caries. The results of this study published in 1993 showed that the prevalence was about 6.0%.

In his review, Milnes et al.20 included some studies from Australia and Southeast Asia. In Indonesia, Aldy et al.33 stated in their study which was published in 1979 that the prevalence of nursing caries was 46.0% among 100 children less than 5 years old who were recruited from a public hospital. Aldy et al.33 defined nursing caries as the presence of dental caries on the labial surface of 1 or more maxillary incisors. Brown et al.34 reported that the prevalence of nursing caries was 5.4% in 112 0- to 2-year-old children who attended maternal and child health clinics or Brisbane Children's Dental Hospital in "lower middle-class suburbs of Brisbane." However, the criteria were not defined.

A study from China was conducted by Deyu et al.35 to assess the prevalence of children with dental caries by age in Chengdu, China in 1982-1983 and 1990-1991. In 1982-1983, 149, 183, 196, 168, 191, and 198 children aged one, two, three, four, five, and six years old were examined, respectively. The prevalence of dental caries among these children was 2.7%, 20.2%, 37.8%, 72.0%, 77.5%, 82.8%, respectively. In 1990-1991, 92, 208, 192, 173, 140, 181 children aged one, two, three, four, five, and xix years old were examined, respectively. The prevalence of dental caries among these children was 5.4%, 18.3%, 50.5%, 75.7%, 85.0%, and 90.1%. One limitation of this study was that the authors did not mention whether the assessment of dental caries included non-cavitated lesions.

Similarly, Wei et al.[36] studied 977 5-year-old children (mean age=5.75) who were randomly selected from 9,000 5-year-old children in the kindergartens in Hong Kong and the Kowloon Peninsula (dates of examinations not specified). The study was published in 1993 and showed that 37% of the children were caries-free, so the prevalence was 63% for caries experience. However, the authors did not mention whether non-cavitated lesions were included or not.

Milnes et al.[20] also reviewed prevalence studies of nursing caries from Africa. Many studies showed that the prevalence of nursing caries was higher among black children compared to white children, despite the fact that the majority of the black children were breastfed compared to white children who were bottle fed. Cleaton-Jones et al.[37] studied the prevalence of dental caries in 499 white children aged 1-5 years old. The authors examined 32, 98, 114, 136, 119 children aged 1, 2, 3, 4, and 5 years, respectively. The results of this study, which was published in 1978, showed that the prevalence of dental caries was 37.5%, 53.1%, 78.9%, 67.6%, and 37.7%, respectively.

In Nigeria, Salako[38] examined 560 children 3-7 years of age who were in the School of Dental Science in Lagos, Nigeria. The study published in 1985 showed that the prevalence of nursing caries was 38.4%, with nursing caries defined as the obvious cavitation or sticking of the probe in a carious lesion. A study conducted in Tanzania by Kerosuo et al.[39] was designed to assess the prevalence of nursing caries in 442 infants 12-30 months of age who attended maternal and child health clinics in Mwanza (rural region) and Morogoro (urban region), Tanzania. The authors defined children with nursing caries as children with at least two maxillary carious incisors or “the presence of linear hypoplasia noted in association with nursing caries pattern.” The study was published in 1991, and showed that the prevalence of the overall nursing caries was 10.6%.

Raadal et al.[40] conducted a study to assess the prevalence of nursing caries among 275 Sudanese children who were 3.5-5.5 years of age from a public preschool in Khartoum, Sudan. The authors defined children with nursing caries as children with carious lesions in the labial or lingual surfaces of two or more maxillary incisors. The results of this study, which was published in 1993, showed that the prevalence of nursing caries” was 5.5%.

Two large studies from the Middle East were included in Milnes’[20] review of nursing caries articles. In Kuwait, 5,473 4- to 5-year-old children were recruited from public kindergartens in Kuwait City, Kuwait, and examined for the prevalence of nursing caries, which was defined according to nursing caries pattern[41]. The study was conducted by Soparkar at al.[41] and published in 1986. Soparkar at al.[41] stated that the prevalence of nursing caries was 11.5%. In Iraq, Yagot et al.[42] screened 2,389 children who were 12-53 months of age from a nursery school in Baghdad. The results of this study which were published in 1990 showed that the prevalence of nursing caries, which was defined as the presence of caries on the gingival third of the labial surface of any of the maxillary incisors, was 15.6%.

Milnes et al.[20] included many studies from North America which have been conducted to assess the prevalence of nursing caries. For instance, a study by Powell et al.[43] assessed the prevalence of nursing caries in 4,000 Los Angeles children who were 18 months to 5 years old. These children were recruited from a dental school, Children’s Hospital of Los Angeles, public health service clinics and private dental offices. The study results were published in 1976 and showed that 1.0% of the 4,000 children were diagnosed as having nursing caries, which was defined according to the history of child's nursing habits and clinical appearance of the teeth, as shown in Table 2. Similarly, Currier et al.[44] examined 180 predominantly preschool age African-Americans who were attending maternal and child health clinics in Richmond, Virginia. The results which were published in 1977 showed that the prevalence of nursing caries, which was defined as the presence of smooth surface caries in the anterior teeth, was 5.0%.

In Ohio, Johnsen et al.[45] assessed the prevalence of nursing caries in 200 3.5- to 5-year-old children who were enrolled in the Head Start program and were living in a fluoridated community. The results of this study, which were published in 1984, showed that the prevalence of nursing caries, which was defined as the presence of labiolingual carious lesions in the maxillary incisors, was 11.0%. Kelly et al.[46] assessed the prevalence of "baby bottle tooth decay" among Alaskan Native children and American Indians. In their study, they recruited 514 children 3-5 years of age (282 Native-American and 232 Alaskan Native) children who were enrolled in rural Head Start programs in 18 villages/tribes in Alaska and Oklahoma. Two examiners screened all the children who participated in the survey. Any child with cavitated lesions on three of the four maxillary incisors was diagnosed as having "baby bottle tooth decay". The results of this study, which was published in 1987, showed that the prevalence of "baby bottle tooth decay" was 66.8%, 41.8%, and 53.1%, respectively, for the Alaskan Native children, American Indian children, and combined. The authors suggested that the increased prevalence of "baby bottle tooth decay" among the Alaskan children might be due to barriers to access to care and cultural and socioeconomic differences.

Similar results were obtained by Broderick et al.[47], who screened 1,607 3- to 5-year-old Navajo (n=1,463) and Cherokee (n=144) children. These children were recruited from 105 Head Start centers in Arizona and Oklahoma. The results of this study, which were published in 1989, showed wide variation at different centers in the prevalence of nursing caries, which was defined as the presence of at least 2 carious maxillary incisor tooth surfaces. In one center, no one was diagnosed as having nursing caries, while in ten other centers, the prevalence of nursing caries was 100%. In total, 72% of the Navajo children in Arizona were diagnosed as having nursing caries and 55% of the Cherokee children in Oklahoma were diagnosed as having nursing caries.

Barnes et al.[48] designed a study in five southwestern States (Arkansas, Louisiana, New Mexico, Oklahoma, and Texas) to compare the prevalence of nursing caries among four different ethnic groups: White-Americans (n=221), African-Americans (n=409), Hispanic-Americans (n=449), and Native Americans (n=151). Nursing caries was defined as caries affecting two or more maxillary primary incisors. The results of this study, which were published in 1967, showed that the prevalence of nursing caries was 22.2%, 20.5%, 23.8%, and 35.1% in White-Americans, African-Americans, Hispanic-Americans, and Native-Americans, respectively. When nursing caries was defined as the presence of carious lesions in 3 or more maxillary incisors, the prevalence was 14.5%, 13.2%, 14.7%, and 23.2% in White-Americans, African-Americans, Hispanic-Americans, and Native-Americans, respectively.

In Canada, Derkson et al.[49] examined 594 children aged 9 months to 6 years who were randomly selected from public health clinics and community centers in Vancouver. They assessed the prevalence of nursing caries, which was defined as the presence of smooth-surface caries on labial/lingual surface of the maxillary incisors, and the results were published 1982 and showed that the prevalence was 3.2%. Using the same definition of nursing caries, Budowski[50] assessed the prevalence of nursing caries in 302 children aged 9 months to 5 years who were recruited from day care centers in Toronto. The results, which were published in 1989, showed that the prevalence of nursing caries was 7.4%, which was slightly greater than the Derkson et al.[49] study. In contrast, the prevalence of nursing caries was very high (65%) in Albert et al.[51] study, which published in 1988. In their study, Albert et al.[51] screened 260 preschool Inuit children aged 3 months to 5 years from 8 communities in the Keewatin District in the North-West

Territories, using the same definition of nursing caries as used by Derkson et al.[49] and Budowski[50].

Other studies from North America have been conducted to estimate the prevalence of nursing caries. For example, Katz et al.[52] studied the prevalence of nursing caries in 375 3- to 5-year-old Head Start children. They defined nursing caries as the presence of a ring-like pattern of decay affecting 1 or more maxillary incisors or 1 or more maxillary incisors that were decayed to the gum line. The results, which were published in 1992, showed that the prevalence of nursing caries was 12.0%. Another study was conducted in Antigua by Vignarajah et al.[53] who screened 482 children 3 to 4 years of age children who were randomly selected from preschool programs. The results, which were published in 1992, showed that the prevalence of nursing caries, which was defined as the presence of carious lesions on the labial or palatal surfaces of at least 2 maxillary incisors, was 4.6%.

Broadly speaking, the most common design of the prevalence studies was cross-sectional. For example, Tsai et al.[54] conducted a study to assess the prevalence of dental caries in pre-school aged children in Taiwan, and to provide a “more descriptive caries index, such as pit and fissure caries, facial/lingual caries, molar proximal caries, and facial/lingual molar proximal lesions”. Moreover, the study was designed to assess the effects of some risk factors, such as lack of proper tooth brushing, high consumption of sweets, and living in an area of low urbanization.

This study[54], with results published in 2006, included 981 children who were living in 1,681 houses from a total of 5,625 houses, which were selected randomly from throughout Taiwan. This study showed that there was high ECC prevalence of over 56% among the 0- to 6-year-old children, following the World Health Organization (WHO) criteria and definitions[55]. Limitations of this survey were those associated with the “nature of field examinations”, such as selection criteria and generalizability. In other words, there was a problem of ensuring the representation of the whole population at large. In addition, the reported caries levels were most likely underestimations of the true prevalence due to two factors. First, the oral examinations did not include radiographs. Second, the criteria used for survey examinations were “conservative” and did not include non-cavitated lesions. Moreover, most of the children did not participate in the second examination, although the authors did offer it. However, this study had many advantages, since it provided the first nation-wide report on caries prevalence of pre-school Taiwanese children.

In Iowa, the prevalence of cavitated and non-cavitated dental lesions was assessed by Warren et al.[12] in the longitudinal Iowa Fluoride Study [56, 57, 58, and 59]. Three thousand and four hundred children from post-partum wards of eight Iowa hospitals were invited to participate, 1,882 did so, and 1,368 provided subsequent information. When the children were 2 to 5 years of age (mean age=4.63), 800 children remained in the study, and 698 were examined by two trained examiners. Although this study differentiated between cavitated and non-cavitated lesions, it did not distinguish between cavitated enamel and cavitated dentin lesions.

The results of this study[12], which were published in 2002, showed that the prevalence of dental caries was 37%, including non-cavitated lesions (63% of the children were caries-free). However, the prevalence of dental caries was 27% when non-cavitated lesions were excluded (73% of the children were caries-free). Also, the mean number of teeth with caries experience including non-cavitated lesions was 1.30 among all children, while it was 0.78, when non-cavitated lesions were excluded. In addition, the study showed that the mean number of surfaces with caries experience among all children, including non-cavitated lesions, was 2.02, while it was 1.08, when non-cavitated lesions were excluded.

The authors stated some limitations of this study, such as the problem of generalizability, since most of the children who remained in this study were from high SES families, so they were not representative of the total population[12]. Furthermore, there was weak inter-examiner reliability for non-cavitated lesions at the tooth level (Kappa=0.24).

Kolker et al.[60] conducted a study during the time period 2002-2003, to assess the relationships between dietary patterns and dental caries in a cohort of 436 low-income African-American children aged 3 to 5 years in Detroit, Michigan. A “2-stage area probability sample” of 39 census tracts included households with an annual income 250% of the U.S. federal poverty level or less, selecting only African-American children less than 6 years of age. From 1,386 eligible children, 1,021 parent-child pairs were recruited and 517 completed food questionnaires. However, 81 children were excluded because the parents reported that they did not have a typical diet in the week prior to the examination (n=40) and due to errors in reporting their responses (n=41). Thus, a total of 436 children were included in the analyses. Children’s dietary intake data were collected by using The Block Kids Food Frequency Questionnaires[R] from Block Dietary Data System of Berkely, California[R]. Trained dentists did the dental examinations using the International Caries Detection and Assessment System (ICDAS)[61]. The overall prevalence of dental caries was 75.0%. However, the prevalence of dental caries was 64.2%, 74.2%, and 86.6% among 3-, 4-, 5-year-old children, respectively.

Similarly, Mitsugi et al[62] recruited 60 3- to 5-year-old Japanese children from the Hiroshima University Dental Hospital and followed up for one year. This study, which was published in 2005, was designed to assess the relationships between the incidence of dental caries and the presence of *Streptococcus mutans* and *Streptococcus sobrinus* bacteria. At baseline, all the children were examined by two trained pedodontists using the WHO caries diagnostic criteria for determining dmft[55]. Children who received antibiotics three months before the study were excluded.

The overall prevalence of caries experience among the study subjects was 76.6%, while the prevalence of caries experience was 92.3%, 95.6%, 50.0%, and 46.2% in children who were *S. mutans* positive and S. sobrinus negative, *S. mutans* positive and *S.*

sobrinus positive, *S. mutans* negative and *S. sobrinus* positive, and *S. mutans* negative and *S. sobrinus* negative, respectively.

Summary of Prevalence Studies

In short, the prevalence of ECC is different in various places and populations around the world and over time. Although the prevalence of ECC has decreased since the middle of the last century in the developed countries, a high percentage of children throughout the world and in the U.S. still suffer from ECC and its consequences. For example, two series of studies (discussed in detail previously in this section) were conducted in the United Kingdom to show the general trends in the prevalence of ECC among different samples with the same characteristics[24-29]. The first series of studies showed a decreased prevalence of ECC among 12- to 60-month-old children from 1971 through 1988[42-44]. The second series of studies showed a decrease in prevalence of ECC among 3-year-old children from 1973 through 1989[24,27,28].

Most of the prevalence studies were cross-sectional studies that assessed the prevalence of ECC among specific risk groups, such as low SES children from specific racial and ethnic background. However, some of these studies assessed the prevalence of ECC at the national level, such as Tsai et al.[45] study (discussed in detail previously in this chapter), which was designed to assess the prevalence of ECC in Taiwan among Taiwanese children who were less than 6 years of age.

As has been mentioned, there seems to be wide variation in the prevalence of ECC, in part due to different definitions and criteria used. Thus, not all of the studies are comparable. For example, Silver et al.[25,26] defined nursing caries as the presence of dental caries on the labial or palatal surfaces of two or more maxillary incisors, while Wendt et al.[30,31] defined nursing caries as the presence of initial demineralization and cavitation on any of the maxillary incisors. Furthermore, some authors followed the WHO criteria, such as Tsai et al.[54], while others followed the International Caries Detection and Assessment System (ICDAS), such as Kolker et al.[60]

A number of studies have been conducted in the United States to assess the prevalence of ECC. For instance, Warren et al.[12] assessed the prevalence of cavitated and non-cavitated dental caries among 698 children aged 2 to 5 years in Iowa as part of the longitudinal Iowa Fluoride Study[56-59]. The study showed that the prevalence of ECC was 37% when non-cavitated lesions were included and 27% when non-cavitated lesions were excluded.

Broadly speaking, a relatively limited number of studies has been conducted to assess the prevalence of ECC among specific high-risk groups, such as African-Americans. Kolker et al.[60] assessed the prevalence of ECC among 436 low income African-American children aged 3 to 5 years in Detroit, Michigan, using the International Caries Detection and Assessment System (ICDAS). This study[60] showed that the prevalence of ECC was 64.2%, 74.2%, and 86.6% among 3-, 4-, and 5-year-old children, respectively.

Incidence of ECC

Incidence Definitions and Types

Incidence is defined as the proportion of individuals in a group free of a specific disease at baseline that develops the disease during a determined period of time[19]. The term incidence is used broadly, since there can be different definitions of its numerator and denominator. In some cases, the persons with the disease at baseline are excluded from the denominator, since they are not at risk of getting the disease again. An example of this is the situation when patients become edentulous, so they are not at risk of developing dental caries. In determining incidence of dental caries, on the other hand, persons who have incipient or frank dental lesions on specific surfaces of specific teeth at baseline might have incipient lesions progress to frank lesions, or develop other dental lesions on other surfaces of the same teeth or other teeth. So, when assessing the incidence of dental caries at the person level or tooth level, the denominator usually remains the same as it was at baseline. However, if the incidence of dental caries is considered at the surface-level, the carious surfaces may be excluded from the denominator, although filled surfaces are not, since they might get recurrent caries. Generally, the term "increment" is used to indicate the number of cases with new disease during a specific period of time[19]. In dental studies, investigators widely use the term "mean caries increment" to indicate the mean increase in dfs/dft and/or DFS/DFT during a specific period of time[19].

There are three types of caries increment. The first is "Crude Caries Increment" (CCI), which could be defined as the total number of sound teeth at baseline that become decayed or filled at the follow-up[63]. Crude Caries Increment is not a precise measure of caries increment, because it does not account for the "reversals" in the caries scoring that can occur, as described as follows: True biological reversals occur when initially decayed teeth at baseline are remineralized at the follow-up. On the other hand, other reversals may be due to raters' misclassification. For example, the investigator may record a tooth surface as decayed at baseline, but mistakenly record it as sound at follow-up.

When reversals are considered, the term "Net Caries Increment" (NCI) is used. Net Caries Increment is equal to the Crude Caries Increment minus the number of reversals (NCI=CCI-Reversals)[63]. For example, if at baseline there were five surfaces with dental caries (incipient and frank lesions) and at follow-up, two of these surfaces were recorded as sound (reversals) and four other surfaces, which were sound as baseline, were recorded as decayed (CCI=4), the NCI= 4-2=2. Generally, there are some assumptions associated with NCI, such as that the probabilities of false negative and false positive errors, which are made by the examiner, are considered to be equal both at baseline and follow-up. Furthermore, in using NCI, all the reversals are assumed to be due to remineralization. In other words, all reversals are considered to be true biological ones.

In order to bridge the gap between Crude Caries Increment and Net Caries Increment, an adjusted form of measuring caries increment was developed. This adjusted form is known as "Adjusted Caries Increment" (ADJCI). The basic idea of the ADJCI is that it gives less weight to the reversals than the NCI does[63]. The formula for ADJCI is:

$ADJCI = Y2\ (1-(Y3/\ (Y3+Y_4)))$, where:

Y2= number of teeth or surfaces that went from sound status to decayed/filled status.

Y3= number of teeth or surfaces that went from decayed/filled status to sound status, and Y4= number of decayed, recurrent, filled, filled unsatisfactory at baseline that changed to decayed, recurrent, filled, filled unsatisfactory at the follow-up[63].

Some researchers have not reported incidence, but only the increase in the prevalence of dental caries or the increase in the mean dft/dfs with time to indicate the progressiveness of the incidence of dental caries in special populations. They do that by simply subtracting the percentage of the children who had dental caries at baseline from that at follow-up or subtracting the mean dmft/dmfs at baseline from that at follow-up. In these studies, age has been shown to be directly associated with the incidence of ECC. In other words, the prevalence of ECC tends to increase with time, but at different rates, as shown by different studies which will be addressed in a later section of this chapter.

Studies Focused on Incidence of ECC

Broadly speaking, there are relatively few studies that assessed the incidence of ECC for young children. However, there are several other longitudinal studies that have incidence data, but were not designed primarily to assess the incidence of ECC. A longitudinal study by Grindefjord et al.[64] and published in 1995 involved 1,095 Swedish one-year-old children who were examined at baseline. After 18 months, 832 were re-examined at the first follow-up (children age=2.5 years) to assess the prevalence of acquisition of mutans streptococci among them. From the first follow-up (children age=2.5) to the second follow-up (children age=3.5), caries progression was assessed. At the second follow-up, 692 were re-examined, while the other 140 children dropped-out of the study for different reasons, such as moving, missing the examinations, and unwillingness to continue to participate. Two dentists examined the children at both follow-up assessments at ages 2.5 years and 3.5 years and they examined the surfaces of all the teeth. Surfaces were recorded as being sound, having initial decay, or having frank decay. Chi-square, Student's t test, and Wilcoxon Mann-Whitney U test were used to analyze the data.

At the first follow-up, 78 (10.3%) of the children were diagnosed as having dental caries and the other 754 were diagnosed as caries-free[64]. However, the number of children with dental caries at baseline was not mentioned. Although it is not specifically stated, it appears that, at the second follow-up, two of the 78 children who were previously diagnosed with dental caries at the first follow-up were recorded as caries-free, and the other 76 were diagnosed as having new dental lesions (72) or restorations(4). On the other hand, for the caries-free group (754), 178 developed new caries lesions and the others (576) remained caries-free. So, the prevalence of dental caries at the second follow-up was equal to (76+178)/692=36.7% and there was an increase of 26.4% in the prevalence of dental caries after a 1-year follow-up (36.7%-10.3%). The difference was statistically significant ($p<0.001$) between the first follow-up and the second follow-up. Furthermore, the percentage of children who had new caries lesions for the period from the first follow-up to the second follow-up was 36.7% ((4+72+178)/692). It should be noted that 28.9% ((178/614)*100%) of the caries-free children developed dental caries during this period, while 92.3% ((72/78)*100%) of the children who were diagnosed with caries at the first follow-up developed new caries lesions during the same period.

In Table 3, which was reproduced from the study, Grindefjord at al.[64] reported caries progression at the surface level from age 2.5-3.5 years. They stated that 0.7% and 1.6% of the intact tooth surfaces (56,978) developed initial and frank caries lesions, respectively. During this period, 3,460 new surfaces erupted, and although it is not mentioned, it was obvious that the newly erupted teeth were not included in the analysis.

Table 3.[64] Caries progression (%) in children who were examined at the first follow-up and the second follow-up (n=692 people and 57,285 surfaces)

First follow-up, 18 months after baseline (children aged 2.5 years) Surfaces	Second follow-up, 2.5 years after baseline (children aged 3.5years) Surfaces Intact	Initial	Manifest	Treated
Intact(n=56,978)	97.7%	0.7%	1.6%	0%
Initial lesions (n=166)	17%	19%	64%	0%
Manifest lesions(n=141)	0%	0%	92%	8%
Newly erupted surfaces between 1st follow-up to the 2nd follow-up. N=3,460 All the surfaces of all the teeth were examined				

In addition, although it is not stated by the authors, we can calculate the total surface Crude Caries Increment from Table 3[64], which equals the sum of sound surfaces that developed carious lesions: ((0.7+1.6)/100)* 56,978=~ 1,310 surfaces for the whole study group. So the mean surface CCI was ~1.89 per person (1,310/692). In addition, we can calculate the surface total Net Caries Increment, which equals the surface CCI minus the total reversals (initial carious surfaces at baseline that were scored as intact at follow-up). Since 17% of the decayed surfaces at the first follow-up (166) were scored as intact at the second follow-up, the total number of reversals is 17/100 * 166=~28. So the total surface NCI was equal to 1,310-28=1,282 surfaces for all study subjects, and the mean surface NCI was 1.85 per person (1,282/692).

The authors[64] stated that 64% of the initial carious surfaces at follow-up one became manifest lesions at follow-up two, and they concluded that children who had caries at follow-up one developed more new carious lesions than those who were caries-free at follow-up one.

In Finland, Karjalainen et al.[65] followed 148 three-year old Finnish children for three years in a study that will be discussed in detail later in this chapter (in the risk factors section). At follow-up, 135 children remained (mean age=73.7 months), and oral examinations were done by a pediatric dentist at baseline and the 3-year follow-up. The results of this study, which was published in 2001, showed that the prevalence of carious lesions and fillings (only dentin lesions) at baseline was 8.0% (11/135), while it was about 28.1% (38/135) at follow-up. Thus, the prevalence of dental caries had increased by about 20.1% during the 3-year follow-up. Furthermore, the prevalence of carious lesions and fillings, when defined with enamel lesions included, was not different at baseline from that at the 3-year follow-up (40.0% for both time periods (54/135)). The mean dmft among all children at baseline and at the follow-up were 0.19 and 0.94, respectively, so the mean increment in dmft was about 0.75 (0.94-0.19).

In Japan, Sakuma et al.[66] conducted a study to predict dental caries development in 1.5-year-old Japanese children. In this study, which was published in 2007, the authors conducted secondary data analysis using data from the Japanese Public Health Services, and 5,107 1.5-year-old children were selected out of 6,738 according to specific inclusion criteria. The inclusion criteria were that the children had to have had oral examinations at 1.5 and 3 years of age, questionnaires completed by their parents, and the required information about the presence of plaque on the labial surfaces of upper incisors at baseline was recorded. The children were subsequently assigned to four groups: N-city, F-city, Y-city (they are urban areas ranging from middle to small cities), and other municipalities (rural area), according to whether they lived in a rural or urban setting and according to the size of the population. General practitioners conducted the oral examinations, yet the authors mentioned nothing about examiner training or intra- or inter-examiner reliability. The results showed that the percentage of children with new carious lesions at follow-up ranged from 28.99% (Y-city) to 35.00% (other municipalities). Furthermore, higher dft at baseline was significantly associated with an increment of more than one carious tooth from the age of 1.5 to 3 years in N-city (P-value<0.01) and other municipalities (P-value<0.05). Moreover, the mean carious tooth increments were 1.30, 1.17, 1.24, and 1.39 for N-city, F-city, Y-city, and other municipalities, respectively. The authors did not mention whether they assessed CCI, NCI, or ADJCI.

Another study by Warren et al.[67] published in 2009 followed very young (6-24 months of age), low SES children prospectively for 18 months and reported important incidence data. Although the main purpose of the study was to assess baseline risk factors associated with 18-month caries prevalence in these children, information about the increased prevalence of caries was included. In this study, 212 WIC-enrolled children were recruited (volunteers), and a questionnaire was designed to collect baseline information from the mothers, such as child's race and ethnic group, family income, mother's education level and mother's beverage consumption. Other information, such as child's beverage consumption (type of beverage, frequency, and amount), night bottle feeding practices, and fluoride exposures was collected at baseline, and after 4-5 months, 9 months, 13-14 months, and 18 months. Caries (cavitated d2-3 lesions and non-cavitated d1 lesions), visible plaque examinations, and MS level (categorized as none, <10, 10-100, 100-200, too many to count) were recorded at baseline, after 9 months, and after 18 months. At follow-up, 128 remained in the study and were re-examined.

This study[67] showed that the prevalence of d1, d2 or filled surfaces increased from 8.6% at baseline to 77.0%. In other words, there was an increase of 68.4% in prevalence of d1d2-3f during the 18-month follow-up (P<0.05, McNemar's test). McNemar's test is a non-parametric statistical technique used to assess whether the difference in a dichotomous traits between two matched pairs is significant[68]. In addition, the prevalence of d2 or filled surfaces increased from 2.3% to 19.5%, an increase of 17.2% (P<0.05, McNemar's test), and the prevalence of d1 surfaces increased from 7.8% to 76.0%, an increase of 68.2% (P-value not mentioned). It is worth noting that the authors mentioned nothing about reversals, so it is not possible to determine the NCI or the ADJCI.

The results of the study[67] showed that the prevalence of cavitated caries was associated with age (P=0.006, Chi-square). The authors mentioned that, among those who were < 12 months at baseline, 11% developed caries (cavitated or non-cavitated); among those who were 12-17 months, 18% developed caries (cavitated or non-cavitated), and among those 18-24 months, 39% developed caries (cavitated or non-cavitated).

As previously mentioned in the prevalence section, Mitsugi et al.[62] designed a study to assess the relationships between the one-year incidence of dental caries and the presence of *S. mutans* and *S. sobrinus* bacteria in a cohort of 60 children who were 3 to 5 years of age at baseline. At the follow-up (1-year after recruitment), 27 children (45%) had new carious lesions, while the incidence of dental caries was 38.5%, 66.7%, 40.0%, and 15.4% in children who were *S. mutans* positive and *S. sobrinus* negative, *S. mutans* positive and *S. sobrinus* positive, *S. mutans* negative and *S. sobrinus* positive, and *S. mutans* negative and *S. sobrinus* negative, respectively. The authors mentioned that inter-examiner reliability was 90%. Although the authors did not mention whether or not there was loss to follow-up or the number of persons who were examined at the follow-up, they mentioned that 11 (18.3%) of the children were caries-free at the follow-up. Since 11 children represents 18.3% of the total children, then the total number of children at the follow-up was 60 (n=11*100/18.3), which is the same as that at baseline. That means there was no loss to follow-up.

Litt et al[68] conducted a study, which was published in 1995, to create a complete model of caries development among low income predominantly African-American children who were 3 to 4 years of age (mean age =3.86) at baseline. The children and their caregivers, who were mostly their mothers, were recruited from Head Start Programs in Hartford City, New London County, Connecticut. At baseline, 460 children had dental examinations by two trained dentists with a mirror, explorer, and flashlight at the surface level. Also, samples of saliva were collected by a sterile wooden tongue depressor in order to assess salivary levels of MS, which were reported at three levels: 0= no detectable CFU; 1=1 to 50 CFUs; 2= more than 50 CFUs. Three hundred and fifty-five of the caregiver-child pairs completed questionnaires about social and behavioral characteristics, dental self-efficacy, dental knowledge, and perceived life stress with the help of research assistants. At one-year follow-up, 184 were re-examined for dental caries and had saliva samples collected, while the caregivers were re-interviewed. Dental caries was identified according to the criteria developed by Radike[69]. Each tooth surface was recorded as decayed, missing, filled, sealed, or sound, while missing teeth were recorded as five missing surfaces if they were lost for reasons other than decay, and teeth with stainless steel crowns were recorded as five filled surfaces. Since the authors used Radike criteria, only cavitated lesions were recorded. However, the authors did not mention it clearly.

The findings of this study[68] showed that the mean dmfs for the 184 children who remained in the study was 2.8 at baseline and 4.6 at 1-year follow-up. This represented an increase of 1.8 (4.6-2.8) in the mean dmfs during this period. On the other hand, the prevalence of dental caries was 44% and 58% at baseline and 1-year follow-up, respectively.

Studies Focused on Comparison of Incidence Rates of ECC between Two or More Groups

Longitudinal studies of older preschool children have shown that the incidence of ECC among children who were caries-free at baseline was less than that of children who had caries at baseline. For example, a study by O'Sullivan et al.[70] published in 1996 assessed the association of the Early Childhood Caries patterns with caries incidence in preschool children. This was part of a prospective study designed to address the biological and psychological risk factors associated with caries incidence[71]. The participants were 481 Connecticut Head Start children, with mean age of 3.8 years at baseline. The children were followed for 2 years, and mean dmfs was determined at baseline and at the two-year follow-up, with 147 children examined both times. They were categorized at baseline into three groups as either “caries-free, having pit or fissure caries without maxillary anterior caries, or having maxillary anterior caries alone or with posterior caries[70] (p. 81).” Five children were excluded from the analysis, since they could not be categorized into any of the previously mentioned categories.

At baseline, 86 of the children were caries-free, 41 had pit and fissure caries pattern, and 15 had maxillary anterior caries pattern[70]. At two-year follow-up, the mean caries increment for the pit and fissure group was 2.1 times as much as that of the caries-free group, with mean caries increments of 2.86 and 1.37, respectively (P-value=0.05). Furthermore, the mean caries increment (Δdmfs) for the maxillary anterior teeth group was about 3.7 times as much as that of caries-free group, with mean caries increments of 5.13 and 1.37, respectively.

In addition to the determination of the mean caries increments for the three groups, the authors also reported the distribution of the new carious lesions among different surfaces: PF=pit and fissure surfaces, PP=posterior proximal surfaces and BL=buccal/lingual smooth surfaces[70]. The information was reported in a figure and did not include exact values, so approximations (all the asterisk values are approximations) of the results showed that, at two-year follow-up, the mean caries increment of PF among those in the pit and fissure caries group was about 1.5*, which was about 1.5 times as much as that of the caries-free group (Δdmfs =1*) and the difference was statistically significant ($P<0.05$). Also the mean Δdmfs of the PF among maxillary anterior caries group was about 2.5*, which is about 2.5 times that of the caries-free group. The mean

Δdmfs of PP among pit and fissure caries group and the anterior maxillary caries group were approximately 0.8* and 1.5*, respectively, which are about 4 and 7.5 times that of the caries-free children (mean Δdmfs=0.2*). Moreover, the mean Δdmfs of BL were about 0.45*and 0.9* for the pit and fissure caries group and the anterior maxillary caries group, respectively, which were about 3 and 6 times that of the caries-free children (Δdmfs=0.15*).

Finally, the authors mentioned that at baseline, 86 (59%) were caries-free and 56 (38%) had caries experience, while 5(3%) dropped out[70]. At follow-up, only 39% of the children were caries-free, and 61% had caries experience. So the difference in the prevalence of dental caries between follow-up and baseline was 22% (61%-39%).

In addition to the O'Sullivan et al.[70] study, Sclavos et al.[72] stated that the incidence of Early Childhood Caries among children who were diagnosed as having Nursing Bottle Caries (cases) was greater than that among children who had not had Nursing Bottle Caries (control). In their study, which was published in 1988, Sclavos et al.[27] defined children with Nursing Bottle Caries as those who had caries on the labial or palatal surfaces of incisors and a history of long term exposure to the nursing bottle. Sixty-nine cases (mean age 3.9 years at baseline) and 66 controls (mean age 3.5 years at baseline) were recruited from the dental school at the University of Queensland in Australia. The recruited children were examined every 6-9 months at three recall visits. For the cases, the mean increments of dmft/dmfs since the previous exam at recall 1, recall 2 and recall 3 were 5.4/8.0, 3.3/4.6, and 2.7/4.6, respectively. Among the controls, the mean increments of dmft/dmfs since the previous exam at recall 1 and recall 2 and recall 3 were 0.7/0.9, 0.9/1.4, and 0.5/0.9, respectively. The differences in the mean dmft and dmfs at all the recall visits between the cases and controls were statistically significant (P-value<0.001). The authors concluded that children with Nursing Bottle Caries were more susceptible to developing new carious lesions than those who did not have Nursing Bottle Caries.

Incidence Data from Preventive Intervention Studies

Several studies that reported on the incidence of Early Childhood Caries over a specific period of time during clinical trials of caries prevention procedures included incidence data. These data, especially for the control group, are useful measures of incidence. Weintraub et al.[73] conducted a longitudinal, randomized clinical trial study to assess the effect of application of fluoride varnish on the development of ECC among low SES children in San Francisco. In their study, 376 children aged 6-44 months were recruited from two public health centers between October 2000 and August 2002. The inclusion criteria were that all the participants were caries-free and had four erupted maxillary incisors. These children were assigned to three groups: consultation and fluoride varnish application twice a year, consultation and fluoride varnish application once a year, or only consultation. At one-year follow-up, 280 remained, and 261 were examined for caries incidence. At two-year follow-up, 202 remained, and 183 were examined for caries increment.

One of the most important limitations of this study was that placebo varnish was unintentionally applied instead of the active fluoride varnish during a 10-month period to some children[73]. Seventy-five percent of those who were assigned to receive two applications of fluoride varnish during the whole study received only one active fluoride varnish. On the other hand, about 49% of those who were assigned to receive four applications of fluoride varnish during the whole study received only two. However, this “protocol deviation” was taken into consideration during data analysis.

The results of this study showed that the incidence of ECC in the control group was significantly greater than for other groups[73]. In the control group, there were 126 caries-free children at baseline, and 100 of them underwent at least one of the two follow-up oral exams. At the 1-year follow-up, 92 children were examined and 27 (29.3%) had dental caries incidence, while 65 (70.7%) were diagnosed as remaining caries-free. At the 2-year follow-up exam, 63 children who were caries-free at 1-year follow-up participated, and 15 of them (23.8%) had dental caries, while 48 (76.2%) were diagnosed as caries-free.

In the group that received consultation and one fluoride varnish application annually, there were 124 caries-free children and 93 of them were examined during at least one of two follow-up exams[73]. At 1-year follow-up, 86 children were examined, and 13 (15%) had dental caries incidence, while 73 (85%) were diagnosed as remaining caries-free. At 2-year follow-up, 69 children who were caries free at 1-year follow-up, were examined and 10 (14.5%) had dental caries incidence, while 59 (85.5%) were diagnosed as caries-free. In total, for those 93 children, who received parental consultation and one application of fluoride annually and had at least one follow-up examination, 23 (24.7%) had dental caries incidence.

In the group that received parental consultation and two applications of fluoride varnish annually, 87 caries-free children at baseline underwent at least one of the two follow-up exams[73]. At the 1-year follow-up exam, 83 children were examined and 11 (13.2%) had dental caries incidence, while 72 (86.8%) were diagnosed as remaining caries-free. At 2-year follow-up, 70 children who were caries-free at 1-year follow-up, were examined. Only 3 (4.5%) had dental caries incidence, while 67 (95.5%) were diagnosed as remaining caries-free. Overall, only 14 children (16%) out of 87 children who had one or both examinations had new carious lesions during the 2-year follow-up.

The analysis of these data showed that the incidence of ECC in both of the intervention groups was significantly lower than that of the control group ($P<0.001$)[73]. Also, the study showed a reduced incidence of ECC with an increase in the number of fluoride applications ($P<0.001$).

In contrast, some authors reported that there was no effect of application of fluoride varnish on decreasing incidence and increment of ECC. For example, an article published in 1982 by Grodzac et al.[73] showed that 401 children (mean age=3.5 years at baseline) who were enrolled in 18 preparatory schools in Warsaw, Poland, had dental examinations at baseline, after one year, and after 2-year follow-up. Dental examinations were done by two trained dentists in a preparatory school clinic using mirror and fine, sharp explorer under perfect lighting condition. The schools were assigned randomly to two groups: the control schools group that did not receive any form of fluoride application and the treatment group that received topical fluoride varnish application twice annually. After the 2-year follow-up, 80 children had dropped out because they changed their place of residence, leaving a total of 321 children. Radiographic examinations were performed at baseline and the second follow-up, and radiographs were evaluated by two trained radiologists as an auxiliary method to detect caries on the distal surfaces of the primary canines, mesial and distal surfaces of the primary first molars and the mesial surfaces of the primary second molars. However, many children did not have both sets of radiographs, mainly due to children's refusal to cooperate and parents' refusal to expose their children to radiation. Of the 321 remaining children, there were 194 children in the test group, 148 of whom had incomplete radiographs and 46 children had complete radiographs. In the control group, there were 127 children, 100 of whom did not have complete radiographs, while 27 had complete radiographs.

The authors determined separately the mean 2-year increments of dmfs1/dmft1 (initial caries lesions were included) and dmfs2/dmft2 (only cavitated caries lesions were included) for those who had clinical dental examinations without complete radiographs and those who had clinical dental examinations with complete radiographs[74]. The mean caries increments of dmfs1, dmft1, dmfs2, and dmft2 in the control group among those who had incomplete radiographs were 6.89, 2.51, 6.71, and 2.46, respectively, and in the test group among those who had incomplete radiographs, the mean values were 6.24, 1.91, 6.35, and 2.04, respectively. None of the differences in the mean caries increment between the control and the test group were statistically significant, except for dmft1 (P= 0.027).

For those who had complete radiographs, the mean caries increments of dmfs1, dmft1, dmfs2, and dmft2 in the control group were 5.89, 1.93, 6.33, and 2.19, respectively, and in the test group, the mean values were 5.55 (however, in the text, it is 5.44), 1.94, 5.59, and 1.91, respectively[74]. None of the differences in the mean caries increments between the control and the test group were statistically significant. The authors concluded that the application of fluoride varnish twice a year to the teeth of high risk children was not an effective prophylactic measure.

Other studies have assessed the effect of home visits and telephone contact as a preventive measure that could be useful in decreasing the incidence of ECC. For example, Plonka et al.[75] recruited 325 low SES children (mean age=42 days at baseline) between January 2007 and June 2008 from the public health clinics of the Logan-Beaudesert area in the state of Queensland, Australia, and followed them for 24 months. The study children were randomly assigned to two groups, yet because of some ethical considerations, their mothers were given the chance to change their group at the start. However, 91% decided to keep their children in their assigned groups. In the first group, 236 children (mean age=44 days) were assigned at baseline to receive home visits with preventive care at 6, 12, and 18 months. At the 24-month follow-up, 188 children received dental examinations with instruction to brush the children's teeth two times a day with low fluoride toothpaste and advice regarding healthy diets. In the second group, 89 children (mean age 10 days) were assigned to receive telephone contacts with preventive instruction at 6, 12, and 18 months. At the 24-month follow-up, 58 received dental examinations. The preventive measures in the group that got telephone contacts included only oral hygiene instructions. In addition, 40 other children (mean age 27 months) were recruited two months before the end of the study, and served as a reference group, since they did not receive home visits nor telephone contacts.

The results of this study[75] showed that, at the 24-month follow-up, three children (1.5%) in the home visits group developed dental caries, and had mean dmft (for those who developed dental caries) equal to 1.3, while four children (6.8%) in the telephone contact group developed dental caries, and with dmft equal to 3.0. The difference in the percentage of children with dental caries between the two groups was statistically significant (P=0.05). On the other hand, nine children (22.5%) in the reference group had dental caries at the 24-month follow-up, with mean dmft equal to 3.0, and the differences in the percentages of children with ECC in the reference versus home visits and telephone contacts groups were both statistically significant (P-values <0.0001 and 0.03, respectively). Thus, the authors concluded that the incidence of ECC could be reduced significantly by using home visits or telephone contacts as preventive strategies.

A randomized clinical trial study (RCT) by Isokangas et al.[76] and published in 2000 assessed the occurrence of dental decay in children from age 0 to 5 years after maternal consumption of xylitol chewing gum, use of chlorhexidine, and fluoride treatment. Three-hundred and thirty-eight pregnant women were recruited and had an oral screening to assess their salivary levels of MS. One-hundred and ninety-five pregnant women fit the inclusion criteria of having high salivary levels of MS (CFU>=100,000/ml). One-hundred and twenty of them were randomly assigned to the xylitol gum group, 32 to the chlorhexidine treatment group and 36 to the fluoride group, while the remaining seven pregnant women were put in the xylitol group because they reported using xylitol chewing gum daily. However, if most of them were randomly assigned, it is not clear why there were 120 pregnant women in the xylitol group.

The women in the xylitol treatment group started using xylitol chewing gum four times a day, with a total daily dose of six to seven grams three months after the delivery of the child[76], but there is no statement for how long. The women in the chlorhexidine treatment group and fluoride treatment group received three chlorhexidine varnish and three fluoride varnish treatments, respectively. These were done at 6, 12, 18 months after the delivery of the child. However, it is not clear what dose of fluoride varnish was applied. The women in the chlorhexidine and fluoride group were instructed not to use xylitol chewing gum during the study period. In the xylitol group, 103, 97, 92, and 90 children had clinical examinations (only cavitated lesions were recorded) at age 2, 3, 4, and 5 years, respectively, while 28, 26, 21, and 23 children in the chlorhexidine treatment group had their clinical examinations (only cavitated lesions were recorded) at age 2, 3, 4, and 5 years, respectively. In the fluoride treatment group, 33, 33, 32, and 30 children had clinical examinations (only cavitated lesions were recorded) at 2, 3, 4, and 5 years of age. In addition, all the children had their microbiological examinations only at baseline when the children were 2 years of age.

The results of this study showed that the mean annual dmf values for children in the xylitol group were 0.02, 0.17, 0.41, and 0.83 at 2, 3, 4, and 5 years of age[76]. On the other hand, the mean annual dmf values for children in the chlorhexidine group were 0.21, 0.77, 1.91, and 3.22 at 2, 3, 4, and 5 years of age, while they were 0.21, 0.55, 1.69, and 2.87 for the children in the fluoride treatment group at age 2, 3, 4, and 5 years, respectively. The authors mentioned that the difference in the mean annual dmf value for the children in the xylitol group at age 5 years (0.83) was statistically significantly lower than that in the chlorhexidine treatment group (3.22) and fluoride treatment group (2.87).

Summary of the Incidence Studies

In short, studying the incidence of ECC is important, since it gives an indication about the occurrence of ECC. However, there are a limited number of studies that have reported on the incidence of ECC, because incidence studies are prospective and thus, time-consuming, expensive, and subject to attrition. Broadly speaking, the incidence of ECC increases with age and different studies reported different incidence rates and increments. There are many risk factors associated with higher incidence of ECC, such as having more carious teeth at baseline. For example, O'Sullivan et al.[70] stated that children with pit and fissure caries and smooth surface caries had greater caries increment during 2-year follow-up compared to caries-free children.

In addition, the incidence of ECC can be modified by applying different preventive care programs. For example, many studies stated that the application of fluoride varnish decreases the incidence of ECC when compared with a control group. Weintraub et al.[73] conducted a RCT study to assess the efficacy of professional application of fluoride varnish and the results showed that the incidence of ECC in the control group was statistically significantly higher in the control group (did not get fluoride varnish application) compared to the treatment group (get professional fluoride varnish application). Furthermore, other preventive care programs showed significant impact on decreasing the incidence of ECC, such as home visits and telephone contacts.

Risk Factors

Introduction

As has been mentioned previously in this chapter, dental caries is a chronic, transmissible, and multifactorial disease that affects a large percentage of the population worldwide. Generally, there are four important factors that play major roles in the development of dental caries. These are the host, microbes, diet, and time. In 1940, Keyes[77] explained the interaction of host, microbes, and diet, and made the well-known Venn diagram that shows these interactions. Later on, the time element was added to indicate that the process of progression of dental caries requires time.

Many risk factors have been reported in the dental literature. Risk factors can be categorized as: demographic (sometimes called risk indicators, since they are not modifiable), such as age, gender, race/ethnicity, and socioeconomic status; dietary, such as sugar consumption, bottle-feeding, infant formula; microbiological, such as the presence of streptococcus mutans; and behavioral, such as tooth brushing, presence of visible plaque, and the use of fluoridated toothpaste.

Demographic Risk Factors

Age

Child's age is considered one of the most important factors that affects the prevalence of ECC. In their study, Nair et al.79 showed that WIC-enrolled children in Linn County, Iowa had 0.13 times greater odds of having ECC (OR=1.13, 95% CI= 1.04-1.26) for each one month increase in age. Age in months was also associated with ECC in the Ramos-Gomez et al.[80] study of the relationships between different bacterial, behavioral, and environmental factors and ECC (odds ratio=1.06, 95% CI=1.02-1.10). Furthermore, a prospective study on sucrose consumption, visible plaque and caries in children from 3 to 6 years of age conducted by Karjalainen et al.[81] showed that the mean dmft for children increased from 0.19 to 0.94 during the three-year follow-up. In addition, this study showed that the proportion of children who had dentin lesions or fillings increased from 8% to 28% during the same follow-up period.

Other longitudinal studies showed that the prevalence of ECC increases with time. For instance, Warren et al.[67] conducted a prospective study to assess baseline risk factors associated with caries prevalence among 212 WIC-enrolled children aged 6-24 months at baseline. This study showed that the prevalence of children who had caries experience (non-cavitated lesions included) increased from 8.6% at baseline to 77.0% at the 18-month follow-up. Also, the prevalence of children who had caries experience (non-cavitated lesions excluded) increased from 2.3% at baseline to 19.5% at the 18-month follow-up. Also, Litt at al.[68] developed a model to study caries development among 460 low income predominantly African-American children aged 3 to 4 years at baseline in New London County, Connecticut. The findings of this study showed that the mean dmfs of the 184 children who had dental examinations at both baseline and 1-year follow-up, was 2.8 and 4.6 at baseline and 1-year follow-up, respectively. In addition, the prevalence of dental caries was 44% and 58% at baseline and the 1-year follow-up, respectively.

Gender

Many national surveys showed that the prevalence rates of ECC among males and females were almost the same and the differences were not statistically significant. For example, in NHANES III, 1988-1994, 1,177 children aged 12-23 months were examined for dental caries (581 males and 596 females)[15]. The results showed that the prevalence rates of ECC among males and females were 0.70% and 1.30%, respectively, when questionable carious lesions were excluded. However, the prevalence rates of ECC among males and females were 1.70% and 1.60%, respectively, when questionable carious lesions were included. Nevertheless, the differences in the prevalence rates of ECC between males and females were not statistically significant (Chi-square P-value=0.22)[15]. Also, data from NHANES III, 1988-1994, showed that there was not a statistically significant difference in the prevalence of dental caries between males (39.50%) and females (40.24%) in a sample of children (number not mentioned) aged 2-11 years (P-value>0.05, exact P-value not mentioned)[82]. NHANES,1999-2004, showed that there was not a statistically significant difference in prevalence rates of dental caries among males (44.43%) and females (39.80%) aged 2-11 years (number of children not mentioned) of age (P-value>0.05, exact P-value not mentioned)[82].

In addition, many studies have been conducted to assess the associations of risk indicators, including gender, with ECC. For example, Tsai et al.[54] conducted a study in Taiwan (discussed in detail previously) to assess the effect of different risk indicators for ECC among 981 children (526 boys and 455 girls) aged less than 6 years. The results published in 2006 showed that the prevalence rates of ECC (defined according to the World Health Organization (WHO) criteria[55]) were 56.70% and 57.22% among the girls and boys, respectively, and the difference in the prevalence rates among the girls and the boys was statistically significant (CI=0.96-2.05)[54]. (P-value was not mentioned)

Hallett el al.[83] conducted a study in Queensland, Australia, to investigate the relationships between some demographic and behavioral risk factors and ECC. A cross-sectional sample of 2,515 4- to 5-year-old children (number of boys vs. girls in the total population not mentioned) was recruited in 2000 from all the state-run preschool children in the north Brisbane health region (they did not mention the number of the preschools). The sampling was based on selecting the first 42% of the available children from each participating pre-school. The children had dental examinations and dental caries experience was recorded using dmft/dmfs indices. ECC was defined as having 1-5 surfaces with caries experience, while severe-ECC (S-ECC) was defined as having six or more surfaces with caries experience.

The results of this study[83] showed that 460 boys and 386 girls (cannot tell percentages—see the paragraph above) had dental caries experience (total=846), and the difference in prevalence of dental caries among boys and girls was not statistically significant at α=0.05. Further breakdown of the data showed that 321 boys and 289 girls (total=610) had ECC (dmfs=1-5) and the difference in prevalence of ECC among boys and girls was not statistically significant at α=0.05, while 139 boys and 97 girls (total=236) had S-ECC (dmfs>=6) and the difference in prevalence of S-ECC among boys and girls was not statistically significant at the significance level of α=0.05 as well.

Vachirarojpisan et al.[84] conducted a study in a rural area in Thailand to assess some of the demographic risk indicators associated with ECC among 6- to 19-month-old children born between March, 2000 and April, 2001. Five-hundred and twenty children were recruited (272 boys and 248 girls) and categorized into four age groups: 6-8 months (n=15), 9-10 months (n=48), 11-14 months (n=167), and 15-19 months (n=157). Since there were not enough teeth in the first and the second groups, statistical analyses were conducted for the children in the third and fourth groups and only for those who had at least one erupted tooth. In the third group, 140 children (70 boys and 70 girls) were included, and in the fourth group, 132 children (63 boys and 69 girls) were included. The "Intensity of Early Childhood Caries" (I-ECC), which was defined as the proportion of teeth with caries experience/erupted teeth, was 0.32 and 0.26, respectively, among boys and girls in the third group, and the difference was not statistically significant (P-value=0.38). In the fourth group, I-ECC was 0.48 and 0.43 among boys and girls, respectively, and the difference was not statistically significant (P value= 0.35).

Ramos-Gomez et al.[80] designed a cross-sectional study to assess the relationships between ECC and several risk factors, such as demographic, behavioral, and environmental factors, in 146 children (45% female and 55% male) aged 3 to 55 months. The volunteer subjects were recruited from San Francisco General Hospital, and the caregivers were asked to complete a questionnaire about behavioral and demographic factors. The study published in 2006 showed that there was statistically significant difference in the prevalence of ECC among boys and girls (CI=0.55-2.53, P-value not mentioned)

Kolker et al.[60] conducted a study in 2002-2003, to assess the effects of dietary patterns on dental caries among 436 low-income (annual income 250% of the U.S. federal poverty level or less) African-American children aged 3 to 5 years in Detroit, Michigan. From 1,386 eligible children, 1,021 parent-child pairs were recruited and 517 completed food questionnaires. However, 81 children were excluded, because of the absence of a typical diet in the week prior to the examination (n=40) and errors in reporting their responses (n=41). Thus, a total of 436 children (198 boys and 238 girls) were included in the analyses. There were 148, 139, and 149 children aged 3, 4, and 5 years, respectively. Among children who were 3 years of age, 66 were boys and 82 were girls and the prevalence of ECC was 30% and 31%, respectively. Among children who were 4 years of age, 68 were boys and 71 were girls and the prevalence of ECC was 35% and 32%, respectively. Among children who were 5 years of age, 64 were boys and 85 were girls and the prevalence of ECC was 35% and 37%, respectively. At each age, the differences in the prevalence rates of ECC between girls and boys were not statistically significant (P-values not mentioned).

Race/Ethnicity

National surveys showed that the prevalence of caries experience varied among different racial and ethnic groups. SES-adjusted data from NHANES III, 1988-1994, showed that the prevalence rates of caries experience among poor (0–99% of the Federal Poverty Line (FPL)) Mexican-American, non-Hispanic Black, and non-Hispanic White children ages 2 to 5 years were 45.6%, 32.5%, and 29.7%, respectively; among the near-poor (100–199% FPL), the prevalence rates were 42.8%, 32.6%, and 24.3%, respectively; and among the non-poor (200% FPL or higher), the prevalence rates were 24.3%, 18.7% and 12.6%, respectively[85]. However, this report did not show whether the differences in the prevalence rates of ECC among different racial and ethnical groups were statistically significant[85].

In addition, SES-adjusted data from NHANES, 1999-2004, showed that the prevalence rates of caries experience among poor (0–99%FPL) Mexican-American, non-Hispanic Black, and non-Hispanic White children aged 2- to 5 years were 50.2%, 37.4%, and 41.7%, respectively, among the near-poor (100–199% FPL), the prevalence rates were 41.8%, 27.5% and 26.6%, respectively[85]. Among the non-poor (200% FPL or higher), the prevalence rates were 26.5%, 23.3%, and 17.1%, respectively. However, this report did not show whether the differences in the prevalence rates of ECC among different racial and ethnical groups were statistically significant[85].

On the other hand, many studies have been conducted to assess the relationships between different risk indicators and ECC. For example, a study by Warren et al.[86] published in 2008 assessed the relationships between different risk indicators and ECC in 1-year-old children. Two-hundred and twelve WIC-enrolled children participated and their caregivers completed a questionnaire that contained demographic and behavioral information. Caries was recorded at the cavitated (d2-3) and non-cavitated (d1) levels. Bivariate analyses of factors associated with caries experience (d1, d2-3, or filled) among children who had at least one erupted teeth (n=187 children) showed that there were no statistically significant differences in the prevalence rates of ECC among Caucasian (11%), Hispanic (21%), and African-Americans or others (6%), with P-value=0.32. Although the results were not statistically significant, there was a substantial difference in the prevalence of ECC among the different racial and ethnic groups and that might be due to the limited sample size which affected the power of detection the differences in the prevalence rates and that the majority of the study subjects were White-American children (66.33%) compared to Hispanic children (23.91%) and African-Americans (9.76%).

Psoter at al.[87] conducted a study to assess the relationships between race/ethnicity and ECC among a cohort of 5,171 Arizona pre-school children from February, 1994 to September, 1995. The children were recruited from Head Start and WIC programs, day care centers and health fairs. Demographic information was collected from the children's caregivers. Dental caries was defined at the cavitated levels and recorded as a dichotomous variable (yes/no). The authors mentioned that 85% of the examined children were non-Hispanic White and Hispanic, but they did not mention the percentage of each ethnic and racial group. SES-, educational level-, gender- and age-adjusted logistic regression odds ratios for ECC were 3.57 (95% CI 2.50-5.09), 1.41 (95% CI 1.02-1.96), 1.87 (95% CI 1.53-2.28) and 1.94 (95% CI 1.19-3.18) among Native Americans, Blacks, Hispanics, and others, respectively, using Whites as the reference group.

SES

National surveys showed that the prevalence rates of ECC differ among different SES groups. Data from NHANES, 1988-1994[85], showed that the prevalence rates of ECC among 2- to 5-year-old children were 35.5%, 29.1%, and 14.0% among the poor (0–99%FPL), near-poor(100–199% FPL) and non-poor (200% FPL or higher), respectively. Among males, the prevalence rates were 34.6%, 30.5%, and 12.9%, respectively, while among females, the prevalence rates of ECC were 36.2%, 27.5%, and 15.3%, respectively. However, this report did not show whether the differences in the prevalence rates of ECC among the poor, near-poor, and non-poor were statistically significant. Also, stratifying the data by race/ethnicity showed that the SES-specific prevalence rates of ECC followed the same pattern. The prevalence rates of ECC among Mexican-Americans were 45.6%, 42.8% and 24.3%, respectively; among non-Hispanic Blacks, the prevalence rates were 32.5%, 32.6% and 18.7%, respectively; and among non-Hispanic Whites, the prevalence rates were 29.7%, 24.3%, and 12.6%[85].

Edelstein et al.[82] also used data from NHANES, 1988-1994, and tested the statistical significance of the differences in the prevalence of dental caries experience among poor (51.2%), near-poor (44.5%), and non-poor (31.1%) children aged 2 to 12 years. The results showed that the differences were not statistically significant (P-value not mentioned).

In addition, data from NHANES, 1999-2004[85], showed that the prevalence rates of ECC among 2- to 5-year-old children were 41.8%, 30.4%, and 17.8% among the poor (0–99%FPL), near-poor (100–199% FPL) and non-poor (200% FPL or higher), respectively. Among males, the prevalence rates of ECC were 42.0%, 31.8%, and 21.1%, respectively, while among females, the prevalence rates of ECC were 41.6%, 29.1%, and 14.5%, respectively. Also, stratifying the data by race/ethnicity showed that the SES-specific prevalence rates of ECC followed the same pattern. The prevalence rates of ECC among Mexican-Americans were 50.2%, 41.8% and 26.5%, respectively; among non-Hispanic Blacks; the prevalence rates were 37.4%, 27.5% and 23.3%, respectively; and among non-Hispanic Whites, the prevalence rates of ECC were 41.7%, 26.6%, and 17.1%, respectively[85].

Edelstein et al.[82] also used data from NHANES, 1999-2004, and tested the statistical significance of the differences in the prevalence of dental caries experience among poor (54.3%), near-poor (48.8%), and non-poor (32.3%) children aged 2 to 12 years. The results showed that the differences were not statistically different.

On the other hand, many studies have been conducted to assess the relationship between SES and the prevalence of ECC. Some of these studies showed that there were statistically significant relationships between SES and the prevalence of ECC. For example, Warren et al.[86] assessed the relationships between different demographic information and ECC in 212 WIC-enrolled children aged 1-year old. Caries was defined at the cavitated (d2-3) and non-cavitated (d1) levels. The results published in 2008 showed that there was a statistically significant difference in the prevalence of ECC among children of families with annual incomes of less than $25,000 (15.0%) and children of families with annual income of $25,000 or more (0.0%), with p-value of 0.04.

In contrast, other studies showed that there was no statistically significant relationship between the prevalence of ECC and SES. For instance, Kumarihamy et al.[88] conducted a cross-sectional study to assess the prevalence of ECC in 422 1- to 2-year-old children from vaccination and weighing centers in semi-urban areas in the district of Colombo, Sri Lanka. The relationships between different socio-demographic factors and means of dmft, which was defined at the cavitated (d1) and non-cavitated (d2 and d3) levels, were assessed. One-way ANOVA showed that there was no evidence that means of dmft in the low-income group (1.52), middle-income group (2.28), and high-income group (1.50) were statistically significantly different (P-value=0.10).

Dietary Risk Factors

Sugar Consumption

Many studies have reported statistically significant relationships between sugar consumption and ECC; however, other studies suggested no such relationship. For example, Rugg-Gunn et al.[89] assessed the relationship between dietary habits and caries increments prospectively during a 2-year follow-up from autumn 1979 to autumn 1981 among 405 English adolescents (mean age=11.5 years) and showed that these relationships were not statistically significant. Burt et al.[90], in a Michigan study, showed that there was a weak association between total sugar consumption and dental caries incidence during a three-year follow-up between 1982 and 1985 among 499, 10- to 15-year-old children. In addition, Gibson et al.[91] conducted a study to examine the relationship between dietary sugar and the presence of dental caries experience, which was defined as the presence of active cavitation that included dentine and pulp, filled teeth due to caries, and missing teeth due to caries. Data from 1,450 British children aged 1.5-4.5 years, who participated in the “National Diet and Nutrition Survey”, were collected in 1992 and 1993 and analyzed. Four-day diaries were collected, providing information about food intake (g/day). Average daily energy and nutrient intakes (KJ) were assessed for each type of food by dividing the overall energy intakes of each food item during the four days of information collection (two week days and two weekend days) by four. There were six food item categories: 1) Biscuits and cakes, 2) Sugar confectionery, 3) Chocolate confectionery, 4) Total biscuits, cakes and confectionery, 5) Soft drinks, and 6) Non-milk extrinsic sugar. Then, percent energy consumption of the mentioned food item categories was categorized into three groups: low, medium and high consumer for each of the mentioned food item categories.

The results showed that there was no statistically significant relationship at the significance level of 0.05 between the presence of caries experience and the percent energy consumption of any of the six food item categories included in the analysis, which were assessed categorically as mentioned above[91]. However, in low SES children who brushed their teeth less than twice per day, there were significant relationships between the presence of dental caries experience and the consumption of non-milk extrinsic sugar and sugar confectionery, with P-values of 0.03 and 0.009, respectively. In contrast, the overall results for all children together (high and low SES), and the results for children from high socioeconomic status showed that there was not any significant relationship at the significance level of 0.05 between the presence of caries experience and any of the six food item categories.

On the other hand, a longitudinal study by Persson et al.[92] was designed to study 12-month old children and gather diet diaries during a 2-year study period in 1979 and 1980. At baseline, 312 children were enrolled, and 275 (88%) were examined at the 2-year follow-up, when they were 3 years old. Analysis of frequency of consumption was done and the results showed that the frequency of consumption per month for some sucrose-rich foods was higher among children with caries experience. For instance, the means of the monthly frequency of consumption of cakes and sweet soups were 7.1 and 3.9, respectively, among children with no caries experience, while they were 10.2 and 8.7, respectively, among children with caries experience (P-value was not mentioned).

Freeman et al.[93] conducted a cross-sectional study which assessed the dental health status using the dmft index of five-year-old low-income children in north and west Belfast. Two-hundred and forty children were selected randomly from 2,666 children in 58 primary schools, and 163 children were examined. This study[93], which was published in 1997, showed that 68% of the examined children had caries experience and that there were not a statistically significant relationships between consumption of sugared carbonated drinks and sugar-containing bedtime drinks combined, and the prevalence of dental caries (CI 1.01-1.67). The authors mentioned that consumption of sugared carbonated drinks and sugar-containing bedtime drinks combined was a categorical variable, without mentioning more information about how it was measured.

As previously mentioned in the incidence section, a study by Warren et al.[67] published in 2009 was designed to examine the relationship between sugar consumption and ECC incidence in very young children who were recruited from WIC clinics in Iowa. Two hundred and twelve 6- to 24-month-old children were enrolled to be followed for 18 months. The mothers were asked to complete questionnaires about their child's sweetened-beverage consumption at baseline, after 4-5 months, after 9 months, after 13-14 months, and after 18 months. For the sweetened-beverages questionnaires, the mothers were asked about the frequency of consumption, and the average amount of consumption. Sugar-sweetened beverages included soda pop, sport drinks, powder concentrate beverages made with sugar, or juice-based drinks with added sugar.

At the 18-month follow-up, 128 children remained and the association between caries experience after 18 months and the consumption of sugar-sweetened beverages was assessed adjusting for age[67]. The results, which were published in 2009, showed that the odds of caries among children who consumed sugar-sweetened beverages were 3.04 as high as the odds for those who did not consume sugar-sweetened beverages at baseline, adjusted for age (odds ratio=3.04 and P=0.04). Also, logistic regression analysis of the baseline sugar-sweetened consumption relationship to d2-3f caries after 18 months was statistically significant (P=0.001), as were age and presence of MS.

Warren et al.[86] also used data from the previously mentioned longitudinal study[67], and analyzed them cross-sectionally to assess factors associated with ECC at baseline when children were from 6 to 24 months old. Out of the 268 children who were recruited in the study, 212 children were examined at baseline (mean age=13 months). However, 25 children were predentate, so they were excluded from the analysis. The bivariate analysis of the remaining 187 children (mean age=14 months) showed that child's mean quantity of consumption of sugared beverages per week was associated with caries experience (P=0.02). However, mean quantity of consumption of sugared beverages per week was not significantly associated with ECC experience in age-adjusted logistic regression (P-value=0.17). Since the consumption of sugared beverage was associated with caries in bivariate analyses and was not associated with caries in the age-adjusted logistic regression analyses, the consumption of sugared beverage was suggested to be associated with older age. Furthermore, because the participants were very young at baseline (mean age=14 months), the duration of sugared beverage consumption for most children in this analysis may not have been long enough to yield a statistically significant association.

Also, the findings showed that the mean quantities of the consumption of regular soda pop, sugar-added powdered beverages and juice drinks per week were significantly higher among children with ECC than that of the caries-free children with P-values of <0.01, <0.01, 0.04, respectively, for each of these beverage categories[86]. The most striking finding was that the mean quantity of the consumption of 100% juice per week was significantly higher among children with ECC than that of the caries-free children (P<0.01). The most important drawback of this study was its cross-sectional design, which did not allow for establishment of causality.

A secondary data analysis published in 2009 by Nair et al.[79] based on an interventional study designed to assess the efficacy of psychoeducational methods to prevent ECC among toddlers[94]. The inclusion criteria were: children were from 18 to 36 months of age; mothers were able to communicate in English and were at least 18 years old; and the caregiver had lived with the child for at least 1 year before the beginning of the study.

At baseline, mothers (115) were asked to complete questionnaires, which included information about snacking (four common snacks), beverage consumption (three common drinks), and snack and beverage combined contents[79]. The United States Department of Agriculture's National Nutrient Database for Standard Reference was used to identify the component of different kind of food and drinks. The authors assigned three categories for the cumulative cariogenicity of snack and beverages: 0, 1, or 2 for non-cariogenic, moderately cariogenic and highly cariogenic, respectively. The bivariate analysis between the presence of ECC and cumulative cariogenicity (0, 1, or 2) for most common drinks showed that there was a statistically significant relationship (P=0.02). However, the authors mentioned some limitations of this study, such as the limited sample size, convenience sampling, and the use of secondary cross-sectional data. It is worth noting that the beverage data did not include the frequency of consumption.

The effect of the frequency of consumption of sugar-containing food or drinks on the development of dental caries is also controversial. A case control study in Australia by Seow et al.[95] which was published in 2009 to assess some of the risk indicators associated with ECC among children in non-fluoridated area in Australia. Six hundred and seventeen children (0-4 years old at baseline) were enrolled in this study. Sixty-five of them were enrolled from free public hospitals, 29 from three private specialist pediatric dental clinics, and 62 from childcare facilities in the state of Queensland, Australia. In addition, 461 controls were recruited from childcare facilities. The mothers were asked to complete questionnaires, including some information about bottle feeding, types of fluids and solid food consumed, and frequency of the consumption of the foods and the drinks. All of the outcome measures were recorded as binary variables (yes/no), and the results of this study showed that there was a statistically significant relationship between caries and added sweeteners to bottles (yes/no), sugar added to weaning solids (yes/no), sugar in fluids (more than two times daily) (yes/no), sugary snacks (yes/no), and sweeteners added to bottle (yes/no), with P-values of <0.001, 0.021, 0.002, 0.047, and <0.001, respectively. The previously mentioned results were not adjusted for age.

On the other hand, a multivariable model, including all the risk indicators after mutual adjustment for age of the child and the age of the mother, showed that, for those with consumption of sweetened-fluids more than once per day, the odds were 4.04 times as high among the cases in the childcare facilities as those among the control group (OR= 4.04 and P=0.07)[95]. There was not a significant relationship between the consumption of the sugary snacks and ECC (P=0.34).

The authors mentioned some of the strengths of this study, which included its high external validity for the Australians because the children were recruited from so many different private and public clinics, and the consistency of the results of this study with the previous ones[95]. However, there were some limitations, such as the possibility of the mothers' recall bias.

In addition, Karjalainen et al.[65] conducted a prospective study, which was published in 2001, in order to assess the relationship between daily sugar consumption in children with low caries prevalence. All the children were part of a prospective STRIP (Special Turku Coronary Risk Factor Intervention Project) infant randomized clinical trial study. The STRIP study was designed to decrease exposure to known environmental risk factors of atherosclerosis. The study included 1,062 7-month-old infants who had important dietary information collected as part of the study. Karjalainen et al.[65] invited every fifth child of the STRIP study to have a dental examination when they were 3 years of age (baseline). A total of 148 families participated and the mean age of the participants was 37.4 months at baseline. These children were followed for three years. At the 3-year follow-up, 135 children (mean age=73.7 months) remained and were re-examined.

The results of this study showed that the differences in the sucrose consumption expressed by grams and percentage of energy from sugar were significantly lower in the caries-free children than that for the children with caries (enamel lesions included) at both baseline (P-values= 0.03 and 0.026, respectively) and follow-up (P-values=0.004 and 0.001, respectively)[65]. In contrast, the results showed that there was not a significant relationship between increased frequency of sweet intake at baseline and caries development at follow-up (CI 0.8-2.5). One of the most important limitations of this study was that the subjects who agreed to participate in the long-term STRIP study might have had more positive health behaviors than people in general society the society. Thus, the generalizability of the results might be limited.

As previously mentioned in the prevalence section of this chapter, Kolker et al.[60] conducted a study, which was published in 2007, to assess the relationship between dietary patterns and dental caries in a cohort of 436 low-income African-American children in Detroit aged 3 to 5 years, Michigan. From 1,386 eligible children, 1,021 pairs of children and their parents agreed to participate and 517 completed food questionnaires. However, 81 children were excluded because they reported that they did not have a typical diet in the week prior to the examination (n=40) and due to errors in reporting their responses (n=41). Thus, a total of 436 children were included in the analyses. Children's dietary intake data were collected by using The Block Kids Food Frequency Questionnaires from Block Dietary Data System of Berkeley, California. Trained dentists examined the children using the ICDAS[61].

Soda consumption accounted for 11% of total sugar intake and was significantly associated with higher levels (n=12-52 surfaces) of dmfs (P=0.049)[60]. Since the authors used the "backward elimination procedure" in model selection, a significance level of 0.1 was used. At this significance level, dmfs level was associated with intakes of powdered/sports drinks, real orange juice, and real juice not including orange (P-values= 0.90, 0.90, and 0.0001, respectively).

In summary, sugar consumption is considered one of the most important risk factors for the development ECC. Thus, many studies have been conducted to assess the relationships between the frequency and the amount of sugar consumption and ECC. Some of these studies showed statistically significant associations between increased frequency and amount of sugar consumption and ECC. For example, as mentioned before in this section, Warren et al.[67] stated that the consumption of sugar-sweetened beverages (yes/no) was associated with ECC in 128 18-month-old children. On the other hand, other studies showed that there was a weak or no relationship between the frequency and the amount of sugar consumption and ECC. For instance, Gibson et al.[91] examined the relationship between sugared contained diet and drinks and caries experience in 1,450 British children aged 1.5 to 4.5 years. The study[91] showed that there were no statistically significant associations between caries experience and percent energy consumption from each of the following sugar-containing diet: biscuits and cakes combined, sugar confectionery, chocolate confectionery, total biscuits, or cakes and confectionery combined which were categorized as low, medium, and high daily percent energy consumption. Also, the study showed that there were no statistically significant relationships between some sugared beverages, such as soft drinks and non-milk extrinsic sugar, and caries experience.

Breast Feeding, Night Feeding and Infant Formula

Results from the 1991 National Health Interview Survey's Child Supplement showed that the majority (95.0%) of children who were 6 months to 5 years old had a history of use of a baby bottle[96], and about 16.6% of the children surveyed had a history of use of a night bottle with contents other than water[96]. These dietary practices, along with the use of infant formula, were associated with early and high levels of *S. mutans* colonization, which is a risk factor for ECC[71].

Many studies have been conducted to assess the relationships between different dietary practices in children with ECC. In Nigeria[97], a study was conducted to assess the relationships between dental caries and infant feeding practices in preschool children. Three of 20 areas were randomly selected near Lagos State, Nigeria. Children were invited to participate in this study and dental examinations (dmft) were given by one single dental examiner, following the WHO Oral Health Surface Method[19]. Dental mirrors and natural light were used in the dental examinations, without the use of radiographs. Intra-examiner reliability was assessed on 23 primary school students (kappa and percent agreement not mentioned). Also, dental caries experience was categorized as rampant caries, caries, and no caries. Rampant caries was defined as the presence of dental caries on one or more maxillary incisors, with or without the involvement of the primary molars at the cavitated levels only. There were 396 6- to 71-month-olds in the study. The results published in 2010 showed that 19 children (4.8%) had rampant caries, 22 (5.5%) had caries experience and 355 (89.2%) had no caries experience, while 2 (0.5%) others had no teeth. Linear regression modeling with dmft counts as the outcome measure showed that mean dmft values increased significantly with every one-month increase in the duration of breastfeeding (P-value=0.002).

However, there were no statistically significant relationships between either night feeding or the duration of bottle feeding (months) and mean dmft (P-values 0.58 and 0.51, respectively).

A study was conducted in Sweden by Forsman et al.[98] to assess the relationships between breastfeeding and consumption of infant formula and dental caries. The children were recruited from Gothenburg, Sweden, a non-fluoridated city with fluoride concentration in tap water of about 0.2 ppm. One thousand children aged 4 years of age, who were born in 1964, were randomly selected from the records of Children's Welfare Center (WEC) in Gothenburg. The authors then selected all the children who were exclusively breastfed for at least the first 5 months of life (B-children) from "the records of the 4-year check-up programs" of WEC, because these records had this information. On the other hand, the authors selected all children who were exclusively fed infant formula for at least the first 5 months of life (F-children), depending on pre-obtained questionnaires asked about feeding patterns of the children when they were infants and completed by the parents. Children who had fluoride tablets or topical fluoride treatments were excluded from the study. Overall, there were 77 B-children and 44 F-children. Caries was recorded by a pediatric dentist when the children were 4 years of age. However, it was not mentioned how caries was recorded and whether it was at the cavitated level only or at the cavitated/non-cavitated levels. There was no statistically significant difference in the mean deft between B-children (mean deft=3.85) and F-children (mean deft=4.30) (P>0.05, specific P-value not mentioned).

Hallett el al.[83] conducted a cross-sectional study in Queensland, Australia, to assess the relationships between some demographic and behavioral risk factors and ECC. All the state-run preschool children in the north Brisbane health region were invited to participate in 2000, and 2,515 preschool children participated and had dental examinations. The dmft/dmfs indices were recorded, but they did not mention whether they were at the cavitated levels only or the cavitated/non-cavitated levels. Children who had dmfs of 1 to 5 were defined as children with ECC, while children with dmfs six or higher were dedined as children with S-ECC. Sampling was based on selecting the first 42% of the available children from each participating pre-school. The results showed that there were statistically significant relationships between the prevalence of ECC and sleeping with bottle (P-value<0.001), and sips from bottle (P-value<0.001)[83].

Seow et al.[95] conducted a case-control study to assess risk factors for ECC, including medical histories and feeding practices. Six-hundred and seventeen children aged 0 to 4 years were recruited from childcare facilities, public and private general anesthesia (GA) dental clinics. About three quarters of the children were ECC-free (n=461) and the rest of the children had ECC (n=156), with 62 in childcare, 65 in a public GA clinic, and 29 in a private GA clinic. The children were examined for the presence of ECC by one examiner using a head-lamp, and following WHO criteria, 1987[6]. ECC was defined according to the AAPD definition[10] and caries was defined at the cavitated level only. A multinomial logistic modeling approach was used to analyze the data. The results showed that there were statistically significant relationships between ECC and both sleeping with the bottle (P-value=0.007) and adding sweetener to bottle (P-value<0.001). However, there was no statistically significant relationship between ECC and breastfeeding (P-value=0.10). Mother's recall bias was mentioned by the authors to be the most important limitation of this study[95].

Another study by Warren et al.[67] published in 2009 followed very young (6-24 months of age), low SES children prospectively for 18 months to assess baseline risk factors associated with 18-month follow-up caries prevalence in these children. In this study, 212 WIC-enrolled children were recruited (volunteers), and a questionnaire was designed to collect baseline demographic and behavioral information from the mothers, including night bottle feeding practices. Caries was defined at the cavitated (d2-3) and non-cavitated (d1) levels. Age-adjusted logistic regression analyses of the relationships between baseline risk factors and ECC at the cavitated level only showed that children who used nighttime bottles had about 30% greater odds for having ECC (Odds ratio=1.34). However, the result was not statistically significant (P-value=0.60). Also, there was no statistically significant relationship between night bottle feeding at baseline and d2-3f caries after 18 months (n=128) (P-value of 0.60).

Microbiological Risk Factors

Mutans Streptococci Bacteria

There are four species (seven serotypes) of the mutans streptococci (MS) group, only two grow in the oral cavity. *Streptococcus mutans* of human is the most predominant species isolated from human saliva and is believed to be the primary causative factor of dental caries[99].

Salivary level of MS is another important factor in the development of ECC. Ramos-Gomez et al.[80] designed a cross-sectional study to assess the relationship between different risk factors and ECC in 146 children aged 3 to 55 months, including different demographic, behavioral, and environmental factors. Most importantly, this study[80] characterized cariogenic bacterial in saliva, such as MS and Lactobacilli and chemical markers for calcium, phosphate, and fluoride.

The study subjects (volunteers) were recruited from San Francisco General Hospital. The parents or caregivers were asked to complete a questionnaire about behavioral and demographic factors[80]. Then 3 ml of saliva was collected from each child to assess the salivary level of MS and Lactobacilli. In this study, several methods were used to analyze the results, including logistic regression and non-parametric (median) ANOVA analysis. The main results of this study[80] showed that, in younger children, lower levels of salivary MS were statistically significantly associated with ECC compared to that of older children. However, the authors did not mention the P-value nor the confidence interval. Another finding was that uninsured children had 1.31 times the odds of having ECC (OR=1.31), when the results were adjusted for age (P-value was not mentioned). One limitation of this study was that the authors mentioned that they couldn't get stimulated saliva, since it was really difficult to get stimulated saliva from very young children[80]. In addition, the authors mentioned that the collected information regarding different behavioral and environmental factors, such as frequency of tooth brushing per day and other risk factors which were mentioned previously in this section, might be inaccurate, so the validity of the study might be questioned.

In addition, O'Sullivan et al.[100] conducted a longitudinal study, which was published in 1996, to assess the effect of the presence of ECC, which was defined according to the definition of the American Academy of Pediatric Dentistry and salivary levels of MS at baseline on the incidence of ECC. One hundred and forty-eight children (mean baseline age=3.8 years) were enrolled and examined for ECC and salivary levels of MS at baseline, and after 1 and 2 years. The results of this study showed that ECC incidence was associated with the level of MS at each of the follow-up exams (CFU>50). Two of the limitations of this study were that the caries status was dichotomized and recorded as caries-free versus caries-positive, and the authors did not differentiate between cavitated and non-cavitated lesions.

Another study which was conducted by Nair et al.[79] and published in 2009 examined 115 WIC-enrolled children in Linn County, Iowa, who were from 18 to 36 months old and who had mothers that were at least 18 years old, could communicate in English, and lived with the child for at least 1 year before the beginning of the study.

At baseline, mothers were asked to complete questionnaires and unstimulated saliva was collected using a sterile wooden tongue blade[79]. These samples of saliva were then transferred to a Replicate Organism Detection and Counting (RODAC) plate filled with mitis salivarius agar containing sorbitol and bacitracin (MSB agar) which were incubated at 37 C for 48 hours[79]. A single trained examiner assessed all of the children for the presence of visible plaque on the labial surfaces of the maxillary incisors. The results of this study showed that the presence of plaque on any maxillary incisor was associated with ECC (odds ratio= 3.78, 95%CI= 1.41-10.16), and was also associated with the presence of MS levels greater than 50 CFUs (OR=3.39, 95% CI= 1.28-8.96). Thus, the presence of 50 CFUs of salivary MS was indirectly associated with ECC. The authors mentioned some limitations of this study, including the limited sample size and convenience sample, and the use of secondary cross-sectional data.

Moreover, Alaluusua et al.[101] assessed the relationship between the time of acquisition of *S. mutans* in the plaque and saliva of 39 children longitudinally, when they were 2, 3, and 4 years old, and caries experience. The results, which were published in 1983, showed that children who were *S. mutans* positive at age 2 had statistically significantly greater dmfs at age 4 (dmfs=10.6) than children who were *S. mutans* negative at age 2 and became positive at either age 3 or age 4 (dmfs=3.4) with P-value<0.005. Also, children who were *S. mutans* positive at age 2 had statistically significantly greater dmfs at age 4 (dmfs=10.6) than children who remained *S. mutans* negative throughout the study (dmfs=0.3) with p-value<0.0003. The authors concluded that children whose saliva and plaque colonize early in the life with *S. mutans* had higher dmfs than those whose saliva and plaque colonize later in the life with *S. mutans*.

As a follow-up to the prospective Finnish mother-child study by Isokangas et al.[76], which was published in 2000 and discussed earlier in this chapter in the incidence section, Laitala et al.[102] conducted a cohort study, which was published in 2012 to assess the relationships between early acquisition of MS from dental plaque and ECC. In the Finnish mother-child study, pregnant mothers were recruited and randomly assigned into three groups. The intervention group was treated with xylitol and two control groups which were treated with fluoride and chlorhexidine, respectively. After the delivery of the children, Isokangas et al.[76] followed the children from age 2 to age 5 years, and Laitala et al.[102] followed the remaining children (n=164) until age 10; 147 children fulfilled the inclusion criteria of having comprehensive information about dental visits and treatments when they were 10 years of age. Among those who remained at age 10, 29 had been MS+

at age 2, while the remaining 118 had been MS-. Dental examinations were done by an experienced dentist who recorded only cavitated lesions, fillings and extractions and was blinded to the MS colonization at baseline when the children were 2 years of age.

Survival analysis was used to compare the survival time of the caries-free children with regard to MS colonization at baseline (children age=2 years) and Cox regression models were used to assess the effect of MS on the survival time[76]. The results of this study showed that the children who were MS- had longer caries-free survival time and lower caries experience compared to the children who were MS+. More precisely, the median age of first recording of dmft>0 or DMFT>0 was 8.0 years for the MS- children and 4.6 years for MS+ children, and the difference was statistically significant (P-value<0.001).

In summary, dental caries is a bacterial disease that is caused mainly by mutans streptococci (MS) bacteria in the oral cavity. Many studies have been conducted to assess the relationships between the presence of MS in saliva and plaque (yes/no), salivary levels of MS (mostly measured in CFUs) and time of acquisition of MS and ECC. Most of these studies showed that MS was associated directly or indirectly with ECC. For example, Nair et al.[79] studied 115 WIC-enrolled children in Iowa and stated that the presence of plaque on any maxillary incisors was associated with the presence of greater than 50 CFUs of MS (95% CI=1.28-8.96) and ECC (CI=1.41-10.16), so salivary MS levels was associated indirectly with ECC. Furthermore, Alaluusua et al.[101] conducted a longitudinal study and examined 39 children at age 2, 3, and 4 years. The study showed that children who were diagnosed as *S. mutans* + at age 2 years had statistically significantly greater dmfs than those who were diagnosed as *S. mutans*+ at age 3 and 4, and those who remained *S. mutan* – with P-values of <0.005 and <0.0003, respectively.

Behavioral Risk Factors

Presence of Visible Plaque

Another important risk factor for ECC is the presence of visible plaque. The presence of visible plaque on any of the maxillary incisors was associated with ECC in the Nair et al.[79] study discussed previously. Moreover, visible plaque has been shown to have a stronger association with caries than was previously thought, as evidenced by the Finnish Family Competence study by Paunio et al[32]. Another study which was conducted in a non-fluoridated region in Australia by Seow et al.[95] showed that the percentage of children with visible plaque on any maxillary incisors' labial surfaces was significantly higher among children with ECC than that among caries-free children ($p<0.001$). This relationship remained significant after adjustment for the child's age and the age of the mother ($P=0.003$). However, other studies have reported that there was no statistically significant correlation between visible plaque and dental caries. For example, a prospective study on visible plaque and ECC by Karjalainen et al.[65] stated that there was no statistically significant association between the presence of visible plaque and ECC in cross-sectional analyses. Moreover, longitudinal analysis conducted as part of the same study showed that the presence of visible plaque at the age of three was not associated with the dental health at 6 years of age (OR= 0.9 / 95% CI 0.5-1.8)[65]. The presence of visible plaque, however, was associated with increased caries risk when combined with sugar intake frequency (P-value not mentioned).

Toothbrushing and the Use of Fluoridated Toothpaste

Many studies assessed the effect of toothbrushing and the use of fluoridated toothpaste on the prevalence and the incidence of ECC. Also, many of these studies have shown that toothbrushing was an important confounding factor, affecting the significance of other risk factors associated with ECC. A study by Tsai et al.[54] published in 2006 assessed the prevalence of ECC among children younger than 6 years of age. Nine-hundred and eighty-one children younger than 6 years of age were recruited from different regions in Taiwan and had dental examinations according to the World Health Organization (WHO) criteria[55] by three pediatric dentists (not mentioned whether they were trained or calibrated). Multiple logistic regression analyses with caries as an outcome showed that there was a statistically significant difference in the prevalence of ECC between children whose teeth had been brushed every night before bedtime and those whose had not (P-value<0.05, exact P-value not mentioned).

A case-control study of ECC in Australia by Seow et al.[95] published in 2009 assessed risk factors for ECC, including maternal psychological influences, in 617 children aged 0-4 years. The children were recruited from childcare facilities, public and private dental clinics. There were 461 ECC-free children and 156 children with ECC (childcare (n=62), public GA clinic (n=65), and private GA clinic (n=29)). The children were examined for the presence of ECC, enamel hypoplasia, and MS. Meanwhile, the mothers of the children were asked to complete dental questionnaires and psychological questionnaires. In addition, medical, dental, feeding and toothbrushing histories were collected. A multinomial logistic modeling approach was used to analyze the data statistically. The results showed that ECC-free children had fewer troubles with brushing than children with dental caries experience and the difference was statistically significant (P-value= 0.01). Exact definition of the variable “troubles with brushing” was not mentioned, and the authors stated that recall bias was one of the most important limitations of this study.

On the other hand, a cross-sectional study by Kumarihamy et al.[88] published in 2011 assessed the prevalence of ECC in 1- to 2-year-old children in semi-urban areas in the district of Colombo, Sri Lanka. Four-hundred and twenty-two children were selected from vaccination and weighing centers by selecting every third visiting child, and consents were obtained from the caregivers. Recruiting continued until the required sample size (n=422) was met. The study showed that there was no statistically significant difference in the mean dmft, assessed at the cavitated and non-cavitated levels following WHO criteria[55], between children who had their teeth brushed once or less per day and those who had their teeth brushed more than twice per day (P-value=0.18). Also, the study showed that there was no statistically significant difference in the mean dmft among those who used fluoridated dentifrice and those who did not (P-value=0.22).

A study by Warren et al.[86] published in 2008 assessed risk factors associated with dental caries experience in 1-year-old children. Two-hundred and twelve WIC-enrolled children were recruited (volunteers), and a questionnaire was designed to collect baseline information from the mothers, such as a child's race and ethnic group, family income, mother's education and mother's beverages consumption. In addition, information about toothbrushing and the use of fluoridated toothpaste was collected. Caries was recorded at the cavitated (d2-3) and non-cavitated (d1) levels. Also, the presence of visible plaque was assessed, and MS level assessments (none, <10, 10-100- 100-200, too many to count) were conducted.

Bivariate analyses of factors associated with caries experience (d1, d2-3, or filled) showed that there was no statistically significant difference in the prevalence of ECC between children who had their teeth brushed daily and those who had not (P-value=0.26)[86]. However, there was a statistically significant relationship between higher prevalence rates of ECC and the use of fluoridated toothpaste (P-value= 0.02). The authors explained this finding by mentioning that the children were so young and many of them had few teeth erupted. So, those who brushed their teeth using fluoridated toothpaste were older in age, and since there was a positive relationship between age of the child and the prevalence of ECC, older children who used fluoridated toothpaste had higher prevalence of ECC. After adjusting for age in logistic regression, the results showed that there was no statistically significant relationship between the use of fluoridated toothpaste and ECC (P-value=0.28)[86].

Vachirarojpisan et al.[84] conducted a study in October, 2001 to assess associations of demographic risk indicators with ECC among 6- to 19-month-old children who were born between March, 2000 and April, 2001 in a rural area in Thailand. Five-hundred and twenty children were recruited and then categorized into four age groups: 6 to 8 months (n=15); 9 to 10 months (n=48); 11 to 14 months (n=167); and 15 to 19 months (n=157). Only children in the third and the fourth groups were included in the analyses, because the children in the first and the second group had very few teeth. I-ECC was assessed, which was defined as the number of teeth that had dental caries experience divided by the total number of erupted teeth. The results showed that there was no statistically significant difference in I-ECC values in the third and the fourth groups between the children who used fluoridated toothpaste and those who did not, with P-value of 0.93 and 0.99, respectively.

Kolker et al.[60] conducted a study in 2002-2003 to assess the effect of dietary patterns on dental caries among 436 3- to 5-year-old low SES African-American children in Detroit, Michigan. One-thousand twenty-one parent-child pairs were recruited from 1,386 eligible children, and 517 completed food questionnaires. Eighty-one children were excluded, because of the absence of enough information about diet in the week prior to the examination (n=40) and errors in recording their responses (n=41). Thus, a total of 436 children were involved in the analyses (148, 139, and 149 children aged 3, 4, and 5 years, respectively). Full multinomial logistic regression model with 4 levels of dmfs (level 1=0 dmfs; level 2=1-4 dmfs; level 3=5-11 dmfs; and level 4=12-52 dmfs) showed that there was no difference in the levels of dmfs among children who had different frequencies of tooth brushing (P-value=0.55).

Overall Summary

The study of the prevalence of ECC is relatively easy, since it depends on a cross-sectional analysis of collected data at one time without the need for any follow-up.

Because of this, there is a relatively large number of studies that assessed the prevalence of ECC in different populations. However, different definitions and criteria were used by the investigators to assess the prevalence of ECC. These different definitions partly contributed to obvious variations in the prevalence of ECC in different samples. Thus, not all of the prevalence studies are comparable. The study of the prevalence of ECC over time in the developed countries showed a dramatic decrease in the number of children with dental caries. For example, a series of studies were conducted in the UK in which 263, 252, 230 children aged 3 years were examined in 1973, 1981, and 1989[25,26,29] for the presence of "rampant caries", which was defined as the presence of dental caries on the palatal or the labial surfaces of two or more maxillary incisors. The results showed that the prevalence of ECC was 8%, 1%, and 4%, respectively. In the United States, many studies have been conducted to assess the prevalence of ECC. For example, Warren et al.[12] assessed the prevalence of cavitated and non-cavitated dental lesions in 698 children aged 2 to 5 years as part of the longitudinal Iowa Fluoride Study[56-59]. The results of this study[12] showed that the prevalence of ECC was 37% when non-cavitated lesions were included, while the prevalence of ECC was 27% when non-cavitated lesions were excluded. Although many prevalence studies of ECC have been conducted, there are few studies which have assessed the prevalence of ECC among high-risk groups, such as African-Americans. Kolker et al.[60] examined 436 low-income African-American children aged 3 to 5 years in Detroit, Michigan. The prevalence of ECC was 64.2%, 74.2%, and 86.6% among 3-, 4-, and 5-year-old children, respectively, using the International Caries Detection and Assessment System (ICDAS). Also, Litt et al.[68] conducted a study of the prevalence of ECC among low income African-American children with mean age of 3.86 years. The prevalence of dental caries was 44% among 184 children recruited from Head Start Programs in Hartford City, New London County, Connecticut.

The number of studies that assessed the incidence of ECC, on the other hand, is limited because the study of incidence requires longitudinal follow-up of the study subjects, which is time-consuming and expensive. Most investigators use the term “mean caries increment” in order to refer to the mean increase in dfs/dft and/or DFS/DFT during a specific period of time[19]. There are three types of caries increment: Crude Caries Increment which does not account for the “reversals”, Net Caries Increment which equals Crude Caries Increment minus reversals, and Adjusted Caries Increment which gives less weight to the reversals. Some studies assessed dental caries incidence as a primary focus. For example, Grindefjord et al.[64] designed a study to assess the incidence of ECC in a cohort of 1,095 Swedish children aged 1 year at baseline and followed them for 2.5 years. The children had dental examinations at 2.5 (n=832) and 3.5 (n=692) years of age. The results of the study showed that the Crude Caries Increment for a period of one year (between age 2.5 and 3.5 years) was ~1.89 surfaces per person and the Net Caries increment was 1.85 surfaces per person.

Other studies compared the incidence of ECC among two or more groups. For instance, O’Sullivan et al.[70] compared the incidence of ECC in three groups of children in a 2-year follow-up study in Connecticut. The first group was composed of caries-free children (n=86 at the follow-up exam.); the second group was composed of children who had pit and fissure caries at baseline (n=41 at the follow-up exam); and the third group was composed of children who had maxillary anterior caries at baseline (n=15 at the follow-up exam). The results showed that the mean caries increment for pit and fissure caries was 2.1 times as great as that of the caries-free group (P-value=0.05) and the mean caries increment for the maxillary anterior caries group was about 3.7 times as much as that of caries-free group (P-value=0.05).

Other incidence studies were preventive intervention studies. For example, Weintraub et .[73]assessed the incidence of ECC in a longitudinal randomized clinical trial study in San Francisco among 376 children who were 6 to 44 months old at baseline and were assigned randomly to three groups. The first group was intended to have fluoride varnish application and consultation twice a year; the second group was intended to have fluoride varnish application and consultation once a year; and the third group had consultation only (control group). However, 49% and 75% of the children in the first group and the second group, respectively, received placebo varnish for the first 10 months of the study instead of active fluoride varnish. At one-year follow-up, 261 remained and had dental examinations and, at two-year follow-up, 183 remained and had dental examinations. The results of this study showed that the incidence of ECC in the control group over the 2-year period was significantly higher than that in the first and the second group (P-value<0.001).

Broadly speaking, there are many risk factors for the development of ECC, such as sugar consumption, the presence of higher levels of salivary MS, the presence of visible plaque, and age. Many studies have been conducted to assess the relationship between the frequency and the amount of sugar consumption and dental caries in children. Some of these studies showed a statistically significant relationship between the frequency and the quantity of sugar consumption and ECC. For example, Kolker et al.[60] assessed the relationships between different kinds of diets and dental caries in a cohort of 436 low-income children aged 3 to 5 years in Detroit. The study showed that the consumption of sugared soda and 100% natural juice (not including orange) were significantly associated with higher levels of dmfs, with p-values of 0.049 and 0.0001, respectively.

Other studies showed that there was a weak or no relationship between the frequency and the amount of sugar consumption and ECC. For instance, Burt et al.[90] in the Michigan study assessed the relationships between sugar consumption and dental caries in a cohort of 499 children aged 10 to 15 years. The results showed that, during the three-year follow-up, there was a weak relationship between total daily amount of sugar consumed, in between meal intake of sugar, sugar consumption as proportion of total energy, and frequency of consumption and dental caries.

Other risk factors, such as high levels and early acquisition of MS, the presence of visible plaque and age, were shown to be associated with the development of ECC. For instance, Nair et al.[79] examined 115 WIC-enrolled children aged 18-36 months and stated that the presence of plaque on any maxillary incisor and the presence of MS levels greater than 50 CFUs were statistically significantly associated with ECC with 95% CI= 1.41-10.16 and 1.28-8.96, respectively. This study[79] also showed that these children had 0.13 greater odds of having ECC for each one month increase in age (OR=1.13, 95% CI = 1.04-1.26).

In short, many studies have been conducted to assess the prevalence and risk factors for ECC in different populations. The results of these studies showed large variation in the prevalence of ECC and controversial findings regarding the relationships between different risk factors and ECC. However, few studies have been conducted to assess prevalence of ECC and its relationships to different risk factors among very young high-risk children, such as African-American children, partly because of the logistic difficulties in recruiting young children. Very few studies have been conducted to assess the incidence of ECC among children. Most of the information that is available about the incidence of ECC comes from longitudinal studies which were conducted basically to assess some risk factors associated with ECC or from randomized clinical trial studies that were designed to assess the efficacy of different preventive treatment and measures. So, it is rational and important to conduct new studies that assess the prevalence, incidence and risk factors associated with ECC to increase the understanding. It is essential to design and conduct studies that assess Crude Caries Incidence and Net Caries Incidence, especially among very young high-risk children, as this will help us understand the development of ECC and shed the light on the teeth and surfaces that develop caries first.

CHAPTER 3

MATERIAL AND METHODS

Overall Study Aspects

Introduction

Data for this project were obtained from an ongoing prospective study which was conducted at the University of Alabama at Birmingham (UAB). The original study was designed to increase the understanding of the process of acquisition of bacteria, especially mutans streptococci bacteria, by children during the time of the eruption of the primary molars and permanent molars. Also, the original study was designed to "investigate the host/parasite relationship for the prognosis and epidemiology of dental caries and its effect on the pathogenesis of dental caries during a 2-year follow-up"[103] (page1). Many bacteria were included in this study, such as the group of bacteria known as the mutans streptococci, including *Streptococcus mutans*, because of its importance in the initiation of the dental caries process. This project involves secondary data analysis of the UAB project data.

There were three aims for the original study. The first was to understand the natural history of the acquisition and colonization of mutans streptococci bacteria in plaque, on the tongue, and in saliva, and to assess the relationship between salivary IgA, which is induced by the presence of MS, and the incidence of dental caries. The second aim was to "establish the number and stability of MS genotype" longitudinally in the study subjects, while the third aim was to determine the similarity in the genotypes of MS among the caregivers and their associations with caries incidence[103].

Study Children Recruitment[103]

Two cohorts of high caries risk, low socioeconomic status, African-American children and mostly with a single parent were invited to participate from Perry County, Alabama. In the first cohort, 75 5-6 year old children were intended to be recruited at baseline. However, 91 children were recruited from the kindergarten at Uniontown Elementary School from March, 2007 to January, 2008.

In the second cohort, 75 African-American children 8-18 months of age were intended to be recruited to participate at baseline by word of mouth in Perry County. However, 97 children aged 3-22 months were recruited from July, 2008 to December, 2009, which was one year after the recruitment of children in the first cohort[103]. The children in the second cohort were not recruited from Head Start programs because the intended ages of these children were too young to be enrolled in this program, which is usually 3 to 5 years of age. The recruitment was facilitated by the Baptist and African Methodist Episcopal (A.M.E.) churches and the “Community Health Advisors,” who were local workers and had a role in educating people in previous programs about risk factors associated with coronary heart diseases and cancer prevention. The Community Health Advisors were not affiliated with the University of Alabama at Birmingham and the direct work with them in this project was through sub-contract with the city. Broadly speaking, the recruitment process for cohort 1 and cohort 2 was terminated when sufficient children who were not “screen-failures” were recruited to participate. Screen-failures were reported when the recruited children had all first permanent molars (cohort 1) or all first primary molars (cohort 2) erupted at the first dental examination, so they were excluded. However, very few children were excluded in the second cohort.

The inclusion criteria for the study were that all the school-age children in the first cohort had to not have the first permanent molars erupted, while the infants in the second cohort could not have had their first primary molars erupted. Furthermore, all the children had to live with their biological mothers and have at least one biological sibling, and the parents had to plan to remain in the area for at least 3 years. Also, the children had to be free from any systemic diseases, such as genetic diseases and birth defects, bleeding disorders, heart conditions, kidney disorders, endocrinal disorders, bone disorders, hepatitis, epilepsy, HIV disease, cancer, immune-deficiencies, and could not have a history of immunosuppressive drug treatment. In both cohorts, index children could not have lived at the same place nor had the same household.

Institutional Review Board (IRB) Approval and Informed Consent

An application was submitted to the Institutional Review Board at the University of Alabama at Birmingham by the Principal Investigator in August 2006 for approval to launch the recruitment of the index children and their family members. The request contained all the information regarding the purpose of the study, number and ages of individuals and specimens intended to be recruited, lack of conflict of interest, and methods of managing and storing the data records, including any and all research-related electronic files and paper documents, was approved in October, 2006. Thirty amendments of the initial IRB approval were submitted and the biggest amendment was approved in November, 2011, when an extension for another 3 years of the original project was submitted.

In addition, the informed consent documents were attached with the IRB request, as shown in the Appendix. The informed consent documents included information about the purpose of this study, and general information about the study, such as duration, frequency of oral examinations, and the importance of completing the diaries and sending them to the investigators. Furthermore, the informed consent documents included information about possible risks and discomforts, which were limited to discomforts related to dental examination of the children. The informed consent documents also included information about benefits, such as receipt of fluoride varnish application and oral hygiene instructions (discussed more later in this chapter), confidentiality and subjects’ right to withdraw at any time without penalty. Informed consent was obtained from all caregivers who were willing to participate.

Information Collection[103]

Baseline Questionnaires (Appendix)

At baseline, the parents were asked to provide detailed demographic, medical, dietary and dental information about their children and other family members who agreed to participate. Demographic information included name, date of birth, and sex (male or female). Medical information included information about the delivery type (standard, C-section, forceps, or others), whether the child was full term (37 to 42 weeks), and child's birth weight (pounds and ounces). Also, the medical history of the index child included information about the presence of allergies and chronic systemic medical conditions (yes/no) such as heart diseases, rheumatic fever, diabetes, hepatitis, epilepsy, bones and kidney disease, having HIV positive, birth defects, and other medical conditions. Information about acute illnesses in the previous 6 months, such as ear, sinus, skin, urinary tract infections, sore throat, and chest cold, was also collected.

Detailed data were gathered about antibiotic use, including total duration of antibiotics taken in the previous six months (none, 1-2 weeks, 3-6 weeks, 7-12 weeks, or more than 12 weeks), time since taking last antibiotic (days, weeks, or months), types of antibiotics (open question), and ways of taking antibiotics (liquid, pill, or other). Furthermore, heights (inches) and weights (pounds and ounces) were obtained starting from the 2nd follow-up (about 24 months after baseline examination).

Dietary and nutritional information involved questions about whether the index child was breastfed (yes/no), types (brand names, not applicable for breast milk), frequency (times per day), timing (throughout the day/at meal and snack times), method of drinking liquids other than water (throughout the day or at meal/snack times), and amounts (cups per day) of beverages (breast milk, infant formula, milk, juice, water, and others) consumed, and history of bed-time (night and nap) bottle use (yes/no). The participants had the chance to record more than one brand name for each of the beverages, so we will deal with them separately in the analyses. For example, if a caregiver reported the use of city water and bottled water, this will be recorded separately as using city water and bottled-water. In other words, the number of people who used city water, bottled-water, well water, etc. will not be mutually exclusive.

Also, juices included 100% natural juice with no sugar added, such as orange juice, tomato juice, and apple juice, while “other beverages” includes all the soft drinks such as, Coke^{TM} and other carbonated beverages, lemonade, Hi-C^{TM}, Hawaiian Punch^{TM}, Caprisun^{TM}, and Koolaid^{TM}, but not 100% natural juice. Drinks like Gatorade^{TM}, Vitamin Water^{TM}, and Red Bull^{TM} also were defined as soft drinks, although they were sometimes called "sport drinks". Also, “other beverages” included tea, milk, and coffee.

There was no question about infant formula in the baseline questionnaire. However, the parents provided brand names which were categorized into either milk or infant formula. For example, if the parents reported “Enfamil AR Lipil^{TM}”, “Isomil^{TM}”, “Similac^{TM}”, etc., it was recorded as infant formula. If the parents reported “Dairy Fresh milk^{TM}”, plain milk, “Piggly Wiggly milk^{TM}”, etc. it was recorded as milk.

Furthermore, the questionnaire asked about oral hygiene information concerning toothbrushing (yes/no), frequency of toothbrushing (times per day), the use of toothpaste (yes/no), and type of toothpaste (brand name). For the brand name of toothpaste, the question was open-ended. So, according to the brand names of the toothpastes provided by the participants, they were reclassified as fluoride-free toothpaste, fluoride toothpaste and unknown. Online sources of information were used in order to support determinations about the fluoride contents of toothpastes.

The dental history included questions about the sources of drinking water (the participants had the opportunity to record more than one source of drinking water from four categories: bottled water, well water, city water, and others), the use of vitamin drops or tablets with fluoride (yes/no), history of a dental problem (yes/no), reason for last visit to the dentist (regular check/other), and the presence of a regular dentist (yes/no) at the time the questionnaires were completed. It should be noted that, in developing questionnaires, they were first sent to a number of “Community Health Advisors” to ask for their ideas and advice. Also, the questionnaires were pilot-tested at UAB with 10 parents who had children older or younger than the ages needed for participation in the study. Thus, they were automatically excluded from the study.

During the study, the parents were given the questionnaires to complete and the coordinators in Uniontown reviewed any questions that came up. Less than 50% of the questionnaires were filled out with the direct help of the coordinators (i.e., the coordinators reviewed the form with them question by question). Furthermore, at the time of this study, the parents of the children in the first cohort were the only ones to complete the four-day diet diaries, shown in the Appendix. The diet diaries covered two weekdays and two weekend days. These diet diaries were similar to ones that had been used previously at the University of Alabama at Birmingham School of Dentistry Pediatric Dental Clinics, since these instruments had been shown to be effective in collecting dietary information to assess the relationships between different food items and ECC. For the study, the parents were provided with self-addressed stamped envelopes to return the diaries. These diaries were collected every six months during the course of the study.

Follow-up Questionnaire (Appendix)

As mentioned before, the caregivers were asked to complete follow-up questionnaires every six months after the baseline questionnaire during the first two years of the study and annually after that. So, caregivers completed the follow-up questionnaires when the mean ages of the children were approximately 1.5, 2.0, 2.5, 3.0 and 4.0 years, respectively. The study codes for these questionnaires were visit 10, visit 40, visit 50, visit 60, visit 70 and visit 90, respectively (Table 4).

Table 4. Dental examinations and questionnaires with study codes and mean ages of the children*

Code	Dental examinations	Questionnaires	Mean age of the children (years)
Visit 10	--	Recruitment	0.9
Visit 20	Baseline	--	1.0
Visit 40	--	Intermediate	1.5
Visit 50	1st follow-up	1st follow-up	1.9
Visit 60	--	Intermediate	2.5
Visit 70	2nd follow-up	2nd follow-up	3.1
Visit 90	3rd follow-up	3rd follow-up	4.1

*There was no visit 30 or visit 80.

* Only 23 children reported at visit 90 for the questionnaire.

Follow-up questionnaires had several additional relevant dietary questions compared to the previously mentioned baseline questionnaire. For example, questions about consumption of sweets such as candy and gum (yes/no), frequency of consumption of sweets such as candy and gum (times/day, times/week and times/month), consumption of sweetened foods such as Pop Tarts™ and sugared cereals, and the frequency of consumption of sweetened food (times/day, times/week and times/month), whether plain sugar was added to any food or drinks (yes/no) and the amount of plain sugar added to any food or drinks (tesspoons/day) also was added. The frequency of consumption of sweets and sweetened foods was converted to times per day (times per week/7 and times per month/30).

Logistics[103]

In Uniontown, there were 18 “trained Community Health Advisors” whose main role was to educate people about the risk factors associated with cardiovascular diseases and cancer prevention program. Thirteen of these “Community Health Advisors” had special training in preventive oral health as an integral part of "The Center for Health Promotion's Special Interest Project", so they were also involved in this study. In addition, the Baptist and African Methodist Episcopal (A.M.E.) churches supported the project, as did various community-based health programs which helped ensure wide participation. Moreover, the "Community Advisory Board", which was comprised of "a former mayor, two retired teachers, a city council person, a former member of the Alabama Legislature, a pastor, the Medical Director of the Andrew Hayden Clinic, two Community Health Advisors, and a business owner", helped support the logistics related to the project and assured the population that the project was designed to benefit the community.

Examiner Training, Calibration and Reliability[103]

There are four dentists involved in this study who serve as dental examiners for the children in the two cohorts and/or their families and they were the same examiners all throughout the study until the present (August, 2012). Three of the examiners who were faculty members at the School of Dentistry, University of Alabama at Birmingham (two pediatric dentists and one general dentist) have examined the index children in both cohorts. The fourth examiner, who is a local dentist, had the responsibility for examining households' family members' teeth.

All the examiners attended a 4-hour educational session before the beginning of the baseline examinations. Prior to examining subjects participating in a pilot study, they reviewed pictures of dental lesions of various severity, including cavitated and non-cavitated lesions, hypoplastic lesions, and sealed and filled teeth. This was the only educational session held from the beginning of the study in 2007 until the present (August, 2012). For examiner calibration at the initial educational session and at least the annual examinations, Dr. Noel Childers, who is the principal investigator of this project, was the gold standard examiner.

From the beginning of the study, calibration of the dental examination of about ten children (~10%) was done annually. In the first three years of the project, all the children, whose dental exams were part of calibration, were from the first cohort (5-9 years of age) and, since the fourth year of the project, the results of the dental exams of about ten children (~10%) from the second cohort were used in calibration instead of from the first cohort. However, there was no training on exams of families (adults, older children) or calibration on them.

The results of the dental examinations by the three examiners, other than the gold standard, were compared to the gold standard's results (inter-examiner reliability) and the results of the gold standard were compared to the gold standard himself in order to assess intra-examiner reliability. Only kappa was used to assess inter- and intra-examiner reliability. The UAB team mentioned that there were not statistically significantly differences among the examiners. Kappa values for inter- and intra-examiner reliability were not made available to this investigator, so they cannot be presented.

Oral Examinations and Sample Collection[103]

The study subjects had thorough oral examinations annually at baseline, 1st follow-up (12 months from baseline), 2nd follow-up (24-months from baseline), 3rd follow-up (36 months from baseline) and 4th follow-up (48 months from baseline) by four trained and calibrated examiners using portable equipment with mirrors, light source and compressed air, without the use of radiographs. Dental explorers were used only occasionally. Dental examinations included DMFT/DMFS/dmft/dmfs at the cavitated level only, without differentiating between d2 and d3 lesions, and non-cavitated lesions (d1) were not recorded.

Saliva and plaque samples were collected at baseline, which will be discussed in detail later in this chapter. At alternate six months follow-up (i.e., 6 and 18 month follow-up), only the eruption of the first permanent molars in the first cohort and of the first primary molar in the second cohort were recorded (i.e., limited examination documentation at these visits). For caries diagnosis, the examiners followed the criteria from WHO criteria[55]. However, there was no training manual for the WHO criteria; instead, photos of molar teeth with various presentations were used by the examiners. The International Caries Detection and Assessment (ICDAS) workshop materials (Baltimore, MD March 2005) were not used as planned initially, because of the difficulty in obtaining examiner agreement. Toothbrush or rubber-cup prophylaxes were completed after sample collection (plaque, and saliva as described in a later section) and before the oral examinations for cohort 1. A technician recorded the examination data on a paper form initially, then by computer when a data entry system was developed. The selected age groups and the frequency of the examinations were designed to correspond with the eruption of primary and permanent molar teeth, and so the incidence of dental caries during the 2 years of follow-up could be assessed.

Benefits to the Study Subjects and Subject Compensation

Following the oral examination, a professional topical fluoride application was provided for every index child every visit, using trays initially (four minutes- 1.23% acidulated phosphate fluoride gel) for cohort 1, and then fluoride varnish for the rest of the visits, as a decision was made by the investigators to do both cohort's fluoride treatment by the same method as planning for recruitment of cohort 2 began. Also, referrals to dentists were made when cavitated (d2 and d3) lesions were observed. Furthermore, oral hygiene instructions were provided by dental personnel, including the examiners, dental hygienist, and dental residents in the Department of Pediatric Dentistry, according to the anticipatory guidance in the textbooks Primary Preventive Dentistry, 6th edition[104] and Pediatric Dentistry: Infancy through Adolescence, 4th edition[105]. However, the dental personnel were not calibrated or standardized.

In addition, monetary compensation of $20 and a preventive oral kit, containing a toothbrush was provided to all the participants at each visit. Additional supplies such as toothpaste, dental flossers, dental floss, dental mirrors, and extra toothbrushes for family members were provided as available.

Family Members Recruitment and Information Collection[103]

The families were asked to participate in this study, and the average number of the recruited family members for each child was 5, so there were 375 (75*5) family members in cohort 1 and another 375 (75*5) in cohort 2. Demographic information for the family members was collected, including name, date of birth, sex, and race. Each primary caregiver's relationship to the index child was recorded as "mother, grandmother, nanny, older sibling, or others". A secondary caregiver was defined as the person who spent the second greatest amount of and time with the index child after the primary caregiver. All the family members had an oral examination and plaque and saliva samples collected. As was done with the children, families of the children in the first cohort were recruited first and, after one year, family members of the second cohort were recruited.

Background about Population of Children by Sex and Race in Perry County in 2000

According to the Institute for Rural Health Research in 2000[8], the number of male children was less than female and the number of white children was less than African-American/others in Perry County, as shown in Table 5. Based on these numbers, 45.6% of the children in Perry County, Alabama, were males and 54.4% were females. Among the 0-4 year old age group, 48.7% of the children were male and 51.3% were female, while in the 5-9 year old age group, 48.8% were male and 51.2% were female.

For the racial distribution, 30.8% of the children were white and 69.2% were African-American or other (Table 5). However, in the 0-4 year old age group, 18.8% of the children were White-Americans and 81.2% were African-Americans or other, while in the 5-9 year old age group, 17.6% were White-Americans and 82.4% were African Americans or other. One of the shortcomings of this report[8] was that a breakdown of the African-American/other category was not provided.

Data Collection and Management[103]

Data collection occurred in the field, at the University of Alabama at Birmingham laboratories, and at other University of Alabama at Birmingham sites. Five full-time equivalent personnel were responsible for field data collection, participant tracking, central database development and management, and laboratory information management. Also, these personnel were responsible for primary data collection and quality control. Data management was categorized into six tasks

Data Collection[103]

The principal investigator and nine other co-investigators (pediatric dentists, general dentists, a statistician, and two epidemiologists) designed paper forms for the purpose of data collection. These forms had instructions that helped to increase data reliability and validity. The investigators recorded personal identifying information of the participants and the date and time of examinations and sample collection that facilitated "HIPAA compliant data transfer[103]."

Table 5. Background about population of children by sex and race in Perry County in 2000[8]

Age	Total			White			African-American/Other		
	Total	Male	Female	Total	Male	Female	Total	Male	Female
Total	11,861	5,410	6,451	3,660	1,735	1,925	8,201	3,675	4,526
0-4	903	440	463	170	76	94	733	364	369
5-9	982	479	503	173	75	98	809	404	405

Data Entry System (DES)

A novel pen tablet based data entry system was developed for this study. This data entry system is comprised of 3 subsystems; the underlying database, the data entry system, and the alert system. The database system is built on a distributed database where each tablet can work independently without network connection and synchronize when a network becomes available, facilitating electronic data acquisition in the field. The main data repository is maintained at UAB's School of Public Health. Physical access to the database is monitored, protected and available to authorized personnel only.

The data entry system provides a means to enroll, collect participant information, collect clinic information, record and track data samples, record clinic information, and provide participant scheduling information to project personnel.

The alert system is a programmable rules-based notification system that continually provides data-monitoring functions, with email reports, phone text messages, and web based data query forms being sent in real time. The system is also capable of tracking whether or not an alert has been responded to and can be programmed to send out additional notifications based upon programmable escalation rules.

Data Storage[103]

All the data were stored in a "networked computerized database[9]" using Microsoft Access™. The reasons behind selecting Microsoft Access™ were its multiple levels of "security and validity checks[103]", characteristics that allow more than one source to enter data at the same time, and the possibility of the centralized control over the database. Also, MS Access™ is very flexible and allowed new data to be added at any time.

Design of the Database[103]

The database was designed in a systematic way that allowed accurate and efficient transfer to SAS program for analysis.

Data Dissemination[103]

Data were disseminated in the form of reports in order to guarantee quality assurance and assessment. Data dissemination included individual identifiers, and the types and the numbers of biological specimens.

Data Analysis[103]

Data were merged for analysis using SAS and the principal investigator had full access to all the parts of the data for data analysis. Also, statisticians and others who had the permission from the principal investigator had access to the data.

Current Study

Introduction

In this report, the prevalence and the three-year incidence of dental caries among children in the second cohort were assessed. In addition, the study assessed the relationships between prevalence and incidence of dental caries and different demographic risk indicators and dietary and dental behaviors

Research Questions:

1. What is the prevalence of ECC among low-SES, African-American Alabama children aged 3-22 months at baseline at each dental exam (baseline and the three follow-up examinations?
2. What are the prevalence counts of ECC among low-SES, African-American Alabama children aged 3-22 months at baseline at each dental exam (baseline and three follow-up examinations)?
3. What are the caries incidence rates among low-SES, African-American children in Alabama who were 3-22 months of age at baseline for all six possible time periods (baseline to 1st follow-up, baseline to 2nd follow-up, baseline to 3rd follow-

up, 1st follow-up to 2nd follow-up, 1st follow-up to 3rd follow-up and 2nd follow-up to 3rd follow-up) ?

4. What are the crude and net caries increments among low SES, African-American children in Alabama who were 3-22 months of age at baseline for all six possible time periods (baseline to 1st follow-up, baseline to 2nd follow-up, baseline to 3rd follow-up, 1st follow-up to 2nd follow-up, 1st follow-up to 3rd follow-up and 2nd follow-up to 3rd follow-up)?
5. What are the bivariate associations between caries prevalence (dichotomous and count) at the follow-up examinations and individual baseline behavioral, birth-related and demographic risk factors, such as frequency of drinking 100% natural juice (times per day), quantity of drinking 100% natural juice (cups per week), frequency of drinking sugar-added beverages (times per day), quantity of drinking sugar-added beverages (cups per week), frequency of drinking of infant formula or milk (times per day), quantity of consumption of infant formula or milk (cups per day), toothbrushing (yes/no), frequency of toothbrushing (times per day), and brushing with toothpaste (yes/no), in African-American children aged 3-22 months at baseline?
6. What are the bivariate associations between caries incidence (dichotomous and count) during all six possible time periods and individual baseline behavioral, birth-related, demographic risk factors, such as, frequency of drinking 100% natural juice (times per day), quantity of drinking 100% natural juice (cups per week), frequency of drinking sugar-added beverages (times per day), quantity of drinking sugar-added beverages (cups per week), frequency of drinking infant formula or milk (times per day), quantity of consumption of infant formula or milk (cups per day), toothbrushing (yes/no), frequency of toothbrushing (times per day) and brushing with toothpaste (yes/no), in African-American children aged 3-22 months at baseline?

7. What are the bivariate associations between three-year caries incidence (dichotomous and count) and different behavioral risk factors at the 2nd follow-up examination and area-under-the-curve (AUC) across visits 40, 50, 60 and 70, such as, frequency of drinking 100% natural juice (times per day), quantity of drinking 100% natural juice (cups per week), frequency of drinking sugar-added beverages (times per day), quantity of drinking sugar-added beverages (cups per week), frequency of drinking infant formula or milk (times per day), quantity of consumption of infant formula or milk (cups per day), toothbrushing (yes/no), frequency of toothbrushing (times per day) and brushing with toothpaste (yes/no), in African-American children aged 3-22 months at baseline?
8. What are the bivariate associations between caries incidence from 1st follow-up to 2nd follow-up (dichotomous and count) and different behavioral risk factors at the 2nd follow-up and area-under-the-curve (AUC) of these risk factors assessed across visits 40, 50, 60 and 70, such as, frequency of drinking 100% natural juice (times per day), quantity of drinking 100% natural juice (cups per week), frequency of drinking sugar-added beverages (times per day), quantity of drinking sugar-added beverages (cups per week), frequency of drinking infant formula or milk (times per day), quantity of consumption of infant formula or milk (cups per day), toothbrushing (yes/no), frequency of toothbrushing (times per day) and brushing with toothpaste (yes/no), in African-American children aged 3-22 months at baseline?
9. What are the multivariable associations between three-year caries incidence (dichotomous and count) and different behavioral risk factors and demographic risk indicators in African-American children aged 3-22 months at baseline?
10. What are the multivariable associations between caries incidence from 1st follow-up to 2nd follow-up (dichotomous and count) and different behavioral risk factors

and demographic risk indicators in African-American children aged 3-22 months at baseline?

Study Hypotheses

The following are the hypotheses for the study:

Demographics

Prevalence

1. Greater age of the child is associated with greater prevalence of ECC at the follow-up examinations.
2. There is no difference in prevalence of ECC at the follow-up examinations between males and females.
3. Higher numbers of household members of the index child are associated with higher prevalence of ECC at the follow-up examinations.
4. Higher parent DMFS scores are associated with greater prevalence of ECC at the follow-up examinations.

Incidence

1. Greater age of the child is associated with greater incidence of ECC.
2. There is no difference in the incidence rate of ECC between males and females.
3. Higher numbers of household members are associated with greater incidence of ECC.
4. Higher parent DMFS scores are associated with greater incidence of ECC.

Risk Factors

Prevalence

1. Increased frequency of consumption of 100% natural juice (times per day) is associated with lower prevalence of ECC at the follow-up examinations.

2. Increased quantity of consumption of 100% natural juice (cups per day) is associated with lower prevalence of ECC at the follow-up examinations.
3. Increased frequency of consumption of sugar-added beverages (times per day) is associated with greater prevalence of ECC at the follow-up examinations.
4. Increased quantity of consumption of sugar-added beverages (cups per day) is associated with greater prevalence of ECC at the follow-up examinations.
5. Increased frequency of consumption of infant formula or milk (times per day) is associated with greater prevalence of ECC at the follow-up examinations.
6. Increased quantity of consumption of infant formula or milk (cups per day) is associated with greater prevalence of ECC at the follow-up examinations.
7. Increased frequency of consumption of milk (times per day) is associated with lower prevalence of ECC at the follow-up examinations.
8. Increased quantity of the consumption of milk (cups per day) is associated with lower prevalence of ECC at the follow-up examinations.
9. Increased frequency of consumption of water (times per day) is associated with lower prevalence of ECC at the follow-up examinations.
10. Increased quantity of consumption of water (cups per day) is associated with lower prevalence of ECC at the follow-up examinations.
11. Consumption of sweets (yes/no) is associated with greater prevalence of ECC at the follow-up examinations.
12. Increased frequency of consumption of sweets (times per day) is associated with greater prevalence of ECC at the follow-up examinations.
13. Consumption of sweetened foods (yes/no) is associated with greater prevalence of ECC at the follow-up examinations.
14. Increased frequency of consumption of sweetened food (times per day) is associated with greater prevalence of ECC at the follow-up examinations.

15. Consumption of food and drinks with added plain sugar (yes/no) is associated with greater prevalence of ECC at the follow-up examinations.
16. Increased quantity of plain sugar (teaspoons per day) added to children's food and drinks is associated with greater prevalence of ECC at the follow-up examinations.
17. Toothbrushing is associated with lower prevalence of ECC at the follow-up examinations.
18. Increased frequency of toothbrushing is associated with lower prevalence of ECC at the follow-up examinations.
19. Use of toothpaste is associated with lower prevalence of ECC at the follow-up examinations.
20. Use of fluoride toothpaste is associated with lower prevalence of ECC at the follow-up examinations.
21. Having a regular dentist is associated with lower prevalence of ECC at the follow-up examinations.
22. Having a previous visit to a dentist is associated with higher prevalence of ECC at the follow-up examinations.
23. The use of vitamin drops or tablets with fluoride is associated with lower prevalence of ECC at the follow-up examinations.

Incidence

1. Increased frequency of consumption of 100% natural juice (times per day) is associated with greater incidence of ECC.
2. Increased quantity of the consumption of 100% natural juice (cups per day) is associated with greater incidence of ECC.
3. Increased frequency of consumption of sugar-added beverages (times per day) is associated with greater incidence of ECC.

4. Increased quantity of consumption of sugar-added beverages (cups per day) is associated with greater incidence of ECC.
5. The increased frequency of consumption of infant formula or milk (times per day) is associated with greater incidence of ECC.
6. The increased quantity of the consumption of infant formula or milk is associated with increased incidence of ECC.
7. Increased frequency of consumption of milk (times per day) is associated with lower incidence of ECC.
8. Increased quantity of consumption of milk (cups per day) is associated with lower incidence of ECC.
9. Increased frequency of consumption of water (times per day) is associated with lower incidence of ECC.
10. Increased quantity of consumption of water (cups per day) is associated with lower incidence of ECC.
11. Consumption of sweets (yes/no) is associated with greater incidence of ECC at the follow-up examinations.
12. Increased frequency of consumption of sweets (times per day) is associated with greater incidence of ECC at the follow-up examinations.
13. Consumption of sweetened foods (yes/no) is associated with greater incidence of ECC.
14. Increased frequency of consumption of sweetened foods (times per day) is associated with greater incidence of ECC.
15. Consumption of food and drinks with added plain sugar (yes/no) is associated with greater incidence of ECC.
16. Increased amount of plain sugar (teaspoons per day) added to children's foods and drinks are associated with greater incidence of ECC.
17. Toothbrushing is associated with lower incidence of ECC.

18. Increased frequency of toothbrushing is associated with lower incidence of ECC.
19. Using toothpaste is associated with lower incidence of ECC.
20. Using fluoridated toothpaste is associated with lower incidence of ECC.
21. Having a regular dentist is associated with lower incidence of ECC.
22. The use of vitamin drops or tablets with fluoride is associated with lower incidence of ECC.

Variables' Definitions and Bivariate and Multivariable Statistical Analyses

Definitions of the Dependent Variables

Table 6 shows the names of different dependent variables and their types, whether they are dichotomous (yes/no), categorical, or continuous at different ages.

Bivariate Analysis and Definitions of the Independent Variables with Dichotomous Dependent Variables

Tables 7 to 14 show the definitions of different independent variables and their types whether they were dichotomous (yes/no), categorical, or continuous. Also, it contains the bivariate analyses when these independent variables were used with dichotomous dependent variables. For the variables that were assessed at the follow-up examinations, area-under-the curve (AUC) of these variables in the follow-up visits was assessed and then the bivariate relationships between AUC of these variables and different dependent variables were assessed. AUC will be assessed for every child with both visit 40 or 50, and visit 70.

A SAS 9.3 macro was used to assess the AUC for each subject and independent variable. Also, the bivariate relationships between the variables that were assessed at the 2nd follow-up examination (visit 70) and different dependent variables will be assessed.

Table 6. Definitions of the dependent variables

Variable Name	Type	Details
Presence of ECC (prevalence)	Dichotomous at each age	Baseline (yes/no) 1st follow-up (12 months after baseline) (yes/no) 2nd follow-up (24 month after baseline) (yes/no) 3rd follow-up (36 months after baseline) (yes/no)
Prevalence of ECC (counts)	Discrete count	Baseline: dmfs (value depends on the number of erupted teeth) 1st follow-up: dmfs (value depends on the number of erupted teeth) 2nd follow-up: dmfs (value depends on the number of erupted teeth 3rd follow-up: dmfs (value depends on the number of erupted teeth)
Crude Incidence of ECC	Dichotomous	From baseline to 1st follow-up (yes/no) From baseline to 2nd follow-up (yes/no) From baseline to 3rd follow-up (yes/no) From 1st follow-up to 2nd follow-up (yes/no) From 1st follow-up to 3rd follow-up (yes/no) From 2nd follow-up to 3rd follow-up (yes/no)
Net incidence of ECC	Dichotomous	From baseline to 1st follow-up (yes/no) From baseline to 2nd follow-up (yes/no) From baseline to 3rd follow-up (yes/no) From 1st follow-up to 2nd follow-up (yes/no) From 1st follow-up to 3rd follow-up (yes/no) From 2nd follow-up to 3rd follow-up (yes/no)
Crude caries increment (CCI)	Discrete count	From baseline to 1st follow-up (dmfs) From baseline to 2nd follow-up (dmfs) From baseline to 3rd follow-up (dmfs) From 1st follow-up to 2nd follow-up (dmfs) From 1st follow-up to 3rd follow-up (dmfs) From 2nd follow-up to 3rd follow-up (dmfs)
Net increment of ECC	Discrete count	From baseline to 1st follow-up (dmfs) From baseline to 2nd follow-up (dmfs) From baseline to 3rd follow-up (dmfs) From 1st follow-up to 2nd follow-up (dmfs) From 1st follow-up to 3rd follow-up (dmfs) From 2nd follow-up to 3rd follow-up (dmfs)

Table 7. Bivariate analyses and definitions of the demographic independent variables with dichotomous dependent variables

Dichotomous Dependent Variable (yes/no)	Independent Variable	Type of Independent Variable	Bivariate Statistical Analysis Technique
NCI	Age	Continuous (months with two decimal places)	Logistic Regression
	Sex	Dichotomous (male/female)	Logistic Regression

Table 8. Bivariate analyses and definitions of the birth-related independent variables with dichotomous dependent variables

Dichotomous Dependent Variable (yes/no)	Independent Variable	Type of Independent Variable	Bivariate Statistical Analysis Technique
NCI	Full-term delivery (37 to 42 weeks)*	Dichotomous (yes/no)	Logistic Regression
	Delivery process	Dichotomous (Standard, C-section**)	Logistic Regression
	Low birth weight (<5 lbs. 8 oz)***	Dichotomous (yes/no)	Logistic Regression

*Children who were born before 37 week were reported as preterm children or premature children.

**C-section referred to cesarean delivery.

***Children who were less than 5 pounds and 8 ounces were reported as low birth weight children

Table 9. Bivariate analyses and definitions of the medical independent variables with dichotomous dependent variables

Dichotomous Dependent Variable (yes/no)	Independent Variable	Type of Independent Variable	Bivariate Statistical Analysis Technique
NCI	Presence of systemic diseases*	Dichotomous (yes/no)	Logistic regression
	Presence of acute illness in the previous six months**	Dichotomous (yes/no)	Logistic regression
	Use of antibiotics in the previous six months	Dichotomous (yes/no)	Logistic regression
	Amoxicillin use (ever)	Dichotomous (yes/no)	Logistic regression
	Antibiotic use other than amoxicillin (ever)	Dichotomous (yes/no)	Logistic regression

*Systemic diseases include genetic diseases and birth defects, bleeding disorders, heart conditions, kidney disorders, endocrinal disorders, bone disorders, hepatitis, epilepsy, HIV disease, cancer and immune-deficiencies.

**Acute illnesses include ear infection, sinus infection, sore throat, skin infection, urinary tract infection and chest cold.

Table 10. Bivariate analyses and definitions of the feeding-practices independent variables with dichotomous dependent variables

Dichotomous Dependent Variable (yes/no)	Independent Variable	Type of Independent Variable	Bivariate Statistical Analysis Technique
NCI	Breastfeeding	Dichotomous (yes/no)	Logistic regression
	Time of consumption of liquids except water	Dichotomous (Throughout the day/At meal/snack times)	Logistic regression
	Night or nap bottle feeding history (ever)	Dichotomous (yes/no)	Logistic regression

Table 11. Bivariate analyses and definitions of the beverages independent variables with dichotomous dependent variables

Dichotomous Dependent Variable (yes/no)	Independent Variable	Type of Independent Variable	Bivariate Statistical Analysis Technique
NCI	Frequency of drinking milk*	Continuous (Times per day)	Logistic Regression
	Amount of drinking milk*	Continuous (Cups per day)	Logistic Regression
	Frequency of drinking water*	Continuous (Times per day)	Logistic Regression
	Amount of drinking water*	Continuous (Cups per day)	Logistic Regression
	Frequency of drinking 100% natural juice*	Continuous (Times per day)	Logistic Regression
	Amount of drinking 100% natural juice*	Continuous (Cups per day)	Logistic Regression
	Frequency of drinking sugar-added beverages*	Continuous (Times per day)	Logistic Regression
	Amount of drinking sugar-added beverages**	Continuous (Cups per day)	Logistic Regression
	Frequency of drinking infant formula	Continuous (Times per day)	Logistic Regression
	Amount of drinking infant formula	Continuous (Cups per day)	Logistic Regression

*Variables that were assessed at the follow-up questionnaires.

**Sugar-added beverages include all beverages, except milk, water, 100% juice.

Table 12. Bivariate analyses and definitions of dietary independent variables with dichotomous dependent variables

Dichotomous Dependent Variable (yes/no)	Independent Variable*	Type of Independent Variable	Bivariate Statistical Analysis Technique
NCI	Consumption of sweets**	Dichotomous (yes/no)	Logistic Regression
	Frequency of consumption of sweets**	Count (times per day)	Logistic Regression
	Consumption of sweetened food***	Dichotomous (yes/no)	Logistic Regression
	Frequency of consumption of sweetened food***	Count (times/day)	Logistic Regression
	Consumption of food or drinks with added plain sugar	Dichotomous (yes/no)	Logistic Regression
	Amount of plain sugar added to food or drinks	Count (teaspoons per day)	Logistic Regression

*All the variables in this table were assessed in the follow-up examinations.

**Sweets include candy, gum, etc.

***Sweetened foods include Pop Tarts™, sugared cereals, etc.

Table 13 shows bivariate analyses and definitions of dental independent variables (toothbrushing, frequency of toothbrushing, use of toothpaste, source of drinking water, presence of a regular dentist, history of a visit to a dentist, reason of last visit to a dentist) with dichotomous dependent variables. Table 14 shows bivariate analyses and definitions of miscellaneous independent variables (mother's DMFS and children's body mass index) with dichotomous dependent variables.

Table 13. Bivariate analyses and definitions of the dental independent variables with dichotomous dependent variables

Dichotomous Dependent Variable (yes/no)	Independent Variable*	Type of Independent Variable	Bivariate Statistical Analysis Technique
NCI	Toothbrushing	Dichotomous (yes/no)	Logistic regression
	Frequency of toothbrushing	Ordinal (To be identified later)	Logistic regression
	Use of toothpaste	Dichotomous (yes/no)	Logistic regression
	Source of drinking water	Dichotomous (city water/others)	Logistic regression
	Ever use of vitamin drops or tablets with fluoride	Dichotomous (yes/no)	Logistic regression
	Presence of regular dentist	Dichotomous (yes/no)	Logistic regression
	History of a visit to a dentist	Dichotomous (yes/no)	Logistic regression
	Reason for last visit to dentist	Dichotomous (regular check/other)	Logistic regression

*All the variables were assessed at follow-up visits.

Table 14. Bivariate analyses and definitions of miscellaneous independent variables with dichotomous dependent variables

Dichotomous Dependent Variable (yes/no)	Independent Variable	Type of Independent Variable	Bivariate Statistical Analysis Technique
NCI	Parent’s caries status	DMFS count	Logistic regression
	Body mass index (BMI)	Count	Logistic regression

Bivariate Analysis and Definitions of the Independent

Variables with Continuous Dependent Variables

Tables 15 to 22 show the definitions of different independent variables and their types, whether they are dichotomous (yes/no), categorical, or count. Also, they contain the bivariate analyses when these independent variables are used with count dependent variables.

Table 15. Bivariate analyses and definitions of the demographic independent variables with count dependent variables

Count Dependent Variable (dmfs)	Independent Variable	Type of Independent Variable	Bivariate Statistical Analysis Technique
Net Caries Incidence (NCI)	Age	Continuous	Negative Binomial Regression Model
	Sex	Dichotomous (Male/Female)	Negative Binomial Regression Model

Table 16. Bivariate analyses and definitions of the birth-related independent variables with count dependent variables

Count Dependent Variable (dmfs)	Independent Variable	Type of Independent Variable	Bivariate Statistical Analysis Technique
NCI	Full-term delivery (37 to 42 weeks)	Dichotomous (yes/no)	Negative Binomial Regression Model
	Delivery process	Dichotomous (Standard, C-section)	Negative Binomial Regression Model
	low birth weight (<5 lbs. 8 oz)	Dichotomous (yes/no)	Negative Binomial Regression Model

Table 17. Bivariate analyses and definitions of the medical independent variables with count dependent variables

Count Dependent Variable (dmfs)	Independent Variable	Type of Independent Variable	Bivariate Statistical Analysis Technique
NCI	Presence of systemic diseases*	Dichotomous (yes/no)	Negative Binomial Regression Model
	Presence of acute illness in the previous six months**	Dichotomous (yes/no)	\Negative Binomial Regression Model
	Use of antibiotics in the previous six months	Dichotomous (yes/no)	Negative Binomial Regression Model
	Amoxicillin use (ever)	Dichotomous (yes/no)	Logistic regression
	Antibiotic use other than amoxicillin (ever)	Dichotomous (yes/no)	Logistic regression

*****systemic diseases include genetic diseases and birth defects, bleeding disorders, heart conditions, kidney disorders, endocrinal disorders, bone disorders, hepatitis, epilepsy, HIV disease, cancer and immune-deficiencies.

**Acute illnesses include ear infection, sinus infection, sore throat, skin infection, urinary tract infection and chest cold.

Table 18. Bivariate analyses and definitions of the feeding-practices independent variables with count dependent variables

Count Dependent Variable (dmfs)	Independent Variable	Type of Independent Variable	Bivariate Statistical Analysis Technique
NCI	Breastfeeding	Dichotomous (yes/no)	Negative Binomial Regression Model
	Time of consumption of non-water beverage	Dichotomous (Throughout the day/At meal/snack times)	Negative Binomial Regression Model
	Night or nap bottle feeding history (ever)	Dichotomous (yes/no)	Negative Binomial Regression Model

Table 19. Bivariate analyses and definitions of the independent beverage variables with count dependent variables

Count Dependent Variable (dmfs)	Independent Variable	Type of Independent Variable	Bivariate Statistical Analysis Technique
NCI	Frequency of drinking milk*	Continuous (Times per day)	Negative Binomial Regression Model
	Amount of milk*	Continuous (Cups per day)	Negative Binomial Regression Model
	Frequency of drinking water*	Continuous (Times per day)	Negative Binomial Regression Model
	Amount of water*	Continuous (Cups per day)	Negative Binomial Regression Model
	Frequency of drinking 100% natural juice*	Continuous (Times per day)	Negative Binomial Regression Model
	Amount of 100% natural juice*	Continuous (Cups per day)	Negative Binomial Regression Model
	Frequency of drinking sugar-added beverages*,**	Continuous (Times per day)	Negative Binomial Regression Model
	Amount of sugar-added beverages*	Continuous (Cups per day)	Negative Binomial Regression Model

*Variables that were assessed at the follow-up visits.

**Sugar-added beverages include all beverages except: milk, water, 100% juice.

Table 20 shows bivariate analyses and definitions of dietary independent variables (consumption of sweets, frequency of consumption of sweets, consumption of sweetened food, frequency of consumption of sweetened food, consumption of food or drinks with added plain sugar and amount of plain sugar added to foods or drinks) with count dependent variables.

Table 20. Bivariate analyses and definitions of independent dietary variables with count dependent variables

Continuous dependent variable (dmfs)	Independent Variable*	Type of Independent Variable	Bivariate Statistical Analysis Technique
NCI	Consumption of sweets**	Dichotomous (yes/no)	Negative Binomial Regression Model
	Frequency of consumption of sweets**	Count (times per day)	Negative Binomial Regression Model
	Consumption of sweetened food***	Dichotomous (yes/no)	Negative Binomial Regression Model
	Frequency of consumption of sweetened food***	Count (times/day)	Negative Binomial Regression Model
	Consumption of food or drinks with added plain sugar	Dichotomous (yes/no)	Negative Binomial Regression Model
	Amount of plain sugar added to food or drinks	Count (teaspoons per day)	Negative Binomial Regression Model

*All the variables in this table were assessed in the follow-up examinations.

**Sweets include candy, gum, etc.

***Sweetened foods include Pop Tarts™, sugared cereals, etc.

Table 21 shows bivariate analyses and definitions of dental independent variables (toothbrushing, frequency of toothbrushing, use of toothpaste, source of drinking water, presence of a regular dentist, history of a visit to a dentist, reason of last visit to a dentist) with count dependent variables. In addition, Table 22 shows bivariate analyses and definitions of miscellaneous independent variables (mother's DMFS) with count dependent variables.

Table 21. Bivariate analyses and definitions of the dental independent variables with count dependent variables

Count Dependent Variable (dmfs)	Independent Variable*	Type of Independent Variable	Bivariate Statistical Analysis Technique
NCI	Toothbrushing	Dichotomous (yes/no)	Negative Binomial Regression Model
	Frequency of toothbrushing	Times per day	Negative Binomial Regression Model
	Use of toothpaste	Dichotomous (yes/no)	Negative Binomial Regression Model
	Source of drinking water	Dichotomous (city water/others)	Negative Binomial Regression Model
	Ever used fluoride vitamin drops or tablets	Dichotomous (yes/no)	Negative Binomial Regression Model
	Presence of regular dentist	Dichotomous (yes/no)	Negative Binomial Regression Model
	History of a previous visit to a dentist	Dichotomous (yes/no)	Negative Binomial Regression Model
	Reason for last visit to dentist	Dichotomous (regular check/other)	Negative Binomial Regression Model

*All the variables were assessed at the follow-up visits.

Table 22. Bivariate analyses and definitions of miscellaneous independent variables with count dependent variables

Count Dependent Variable (dmfs)	Independent Variable	Type of Independent Variable	Bivariate Statistical Analysis Technique
NCI	Mother's caries status	Count (DMFS)	Negative Binomial Regression Model

Multivariable Analyses

Multivariable Analyses with Dichotomous Dependent Variables

For the multivariable analyses with dichotomous dependent variables, we included all the variables with P-values of 0.15 or less in the bivariate analyses mentioned above. Logistic regression was used to analyze the data.

Multivariable Analyses with Count Dependent Variables

For the multivariable analyses with count dependent variables, we included all the variables with P-values of 0.15 or less in the bivariate analyses mentioned above. Negative binomial regression was used to analyze the data.

Prioritization and Selection of Dependent Variables for Multivariable Analyses

Generally speaking, there were two main categories of dependent variables: prevalence and incidence. Each of them was either dichotomous or a count. Also, there were four time points in the study for prevalence: baseline, 1st follow-up (after 12 months from baseline), 2nd follow-up (after 24 months from baseline), and 3rd follow-up (after 36 months from baseline). Thus, there were a total of eight models with prevalence as a dependent variable (four time points times two types of prevalence (dichotomous/count)), since prevalence was assessed cross-sectionally. However, there were six possible time periods to assess for incidence: baseline to 1st follow-up, baseline to 2nd follow-up, baseline to 3rd follow-up, 1st follow-up to 2nd follow-up, 1st follow-up to 3rd follow-up, and 2nd follow-up to 3rd follow-up. So, there were 12 possible models with incidence as a dependent variable (six possible time periods times two types of incidence (dichotomous/count)). Thus, taken together, we had 20 models at the person-level. Also, there were many other possible caries outcomes that could have been considered, such as prevalence and/or incidence by arch (maxillary vs. mandibular), tooth-type (incisors vs. canine vs.1st molars vs. 2nd molars), side (right vs. left), gender (males vs. females), caries experience status (decayed, filled, missing, decayed or filled, decayed or missing or filled), person-level or tooth-level or surface-level experience, and type of incidence (CCI, NCI, or ADJCI).

One question was “Did we need to include all of the 20 possible person-level models in the project?” The answer probably was “No.” After running some preliminary descriptive analyses for prevalence rates at the four time points, we found that the prevalence rates for ECC were 1.10% (n=91), 12.79% (n=86), 39.29% (n=84), and 65.75% (n=73), for baseline, 1st, 2nd and 3rd follow up, respectively. Since the prevalence was so low at baseline (one person had dental caries experience out of 91), we would not be able to build a model with the prevalence of ECC at baseline as an outcome. According to Petrie et al.[106], we should not over-fit the model by including too many independent variables, because the model will be of “little use for predicting future outcomes”. Also, Petrie et al.[106] stated that a general rule in building a “sensible model”, was to include no more than “n/10” independent variables. A more specific rule is to include no more than “number of cases/10” independent variables. Also, since the prevalence of ECC at baseline was very low (1.1%), the incidence from baseline to 1st follow-up, baseline to 2nd follow-up and baseline to 3rd follow-up was almost the same as the prevalence at 1st follow-up, 2nd follow-up and 3rd follow-up, respectively. Thus, prevalence of ECC at baseline and the three follow-up examinations as a dependent variable were not analyzed in order to prevent redundancy.

For the incidence modeling, we used NCI instead of CCI, because it accounts for the reversals, some of which are due to the examiner “error” and are unavoidable in clinical examinations and expected because of the fluoride varnish treatment the children received. Our main priority was to build a model with the incidence of ECC for the whole period (baseline to 3rd follow-up) as an outcome, so we had two models (dichotomous/count). For the incidence from baseline to the 3^{rd} follow-up, we had the smallest sample size, because we had fewer children at the 3^{rd} follow-up examination than all other exams, due to attrition. Thus, we chose another incidence period that gave us a relatively high incidence of ECC and a larger sample size, so we had a greater power to detect differences in the incidence of ECC, depending on different dietary and dental practices. After running some initial univariate analyses, we found that the incidence from the 1^{st} follow-up to the 2^{nd} follow was relatively high (38.6%), with a relatively larger sample (n=83). Thus, we decided to build four main multivariable models (two incidence periods times two types of incidence (dichotomous and count)). Also, since there was a difference in the ages of the children at recruitment (from 3-22 months), we decided to consider adjustment for age in each of the four multivariable models. In short, we decided to develop the following multivariable models:

Model 1-A: Three-year incidence from baseline to 3^{rd} follow-up (dichotomous)-not adjusted for age.

Model 1-B: Three-year incidence from baseline to 3^{rd} follow-up (dichotomous)-adjusted for age.

Model 2-A: Three-year incidence from baseline to 3^{rd} follow-up (dmfs count)-not adjusted for age.

Model 2-B: Three-year incidence from baseline to 3^{rd} follow-up (dmfs count)-adjusted for age.

Model 3-A: Incidence from the 1^{st} follow-up to the 2^{nd} follow-up (dichotomous)-not adjusted for age.

Model 3-B: Incidence from the 1^{st} follow-up to the 2^{nd} follow-up (dichotomous)-adjusted for age

Model 4-A: Incidence from the 1^{st} follow-up to the 2^{nd} follow-up (count)-not adjusted for age.

Model 4-B: Incidence from the 1st follow-up to the 2nd follow-up (count)-adjusted for age.

For every incidence variable, we had three age options: age at the beginning of the time period, age at the end of the time period and change in age interval (age difference). The change in age between the baseline exam and the 3rd follow-up exam was relatively uniform (90% of the children who were examined at baseline were reexamined after 2.7-3.1 years). Thus, change in age was not included as an independent variable, because it was not a likely predictor of incidence of ECC. On the other hand, age at baseline depended on having at least one unerupted 1st molar (inclusion criteria), and 90% of the children were from approximately age 0.6 to 1.6 years at baseline. However, when we assessed the relationship between the age at baseline and the incidence of dental caries, age was not statistically significantly associated with ECC. Thus, we decided to include the age at the 3rd follow-up as a predictor of incidence instead (90% of the children were approximately from age 3.5 to 4.6 years at the 3rd follow-up).

Model Selection and Multivariable Model Building

Each behavioral risk factor and each risk indicator first was modeled separately, using univariable logistic regression with dichotomous dependent variables, or negative binomial models with count dependent variables. We used the significance level of 0.15 as a screening cut-off for inclusion of the independent variables in the development of the multivariable models related to risk for ECC (incidence). For the logistic regression, odds ratios were used to assess the magnitudes and the directions of the associations among the dichotomous dependent variables and different independent risk factors assessed at baseline (visit 10), AUC (visits 40 to 70) and the 2nd follow-up (visit 70). However, for the negative binomial models, the incidence rate ratios (IRR) were assessed. IRR is obtained from the exponentiation of the estimates of the negative binomial model regression equation, since the coefficient estimates are assessed on the log scale.

Multivariable logistic regression and multivariable negative binomial models were used to assess risk associated with the independent risk factors and risk indicators that had screening P-values $<= 0.15$ from the bivariate analyses. A manual backward elimination procedure was performed to choose the best model. Changes in Akaike Information Criterion (AIC) were not considered when evaluating the fitted model generated from the preceding step, because AIC is sensitive to changes in sample size associated with deleting variables from the models. Thus, the P-values determined what variables remained in the multivariable models.Independent variables that had the highest P-values were eliminated sequentially, so our final model had variables with statistically significant parameter estimators at an α level of 0.10. The reason behind choosing α level of 0.10 instead of 0.05 was the limited sample size and exploratory nature of the analysis, so we did not have high power to detect differences in the incidence of ECC among children with different behavioral risk factors, and thus did not want to exclude variables of possible importance. Nevertheless, we also explored models at α=0.05.

Also, two-way interactions between the different independent variables which remained in the final models were assessed. In addition, we developed alternative adjusted models for age by forcing it into all the models, even if it was not significantly associated with different dependent variables at the bivariate level. SAS 9.3 (SAS Institute Inc., Cary, NC, USA) was used to assess all the data, using “PROC COUNTREG” when the dependent variables were counts and “PROC LOGISTIC” when the independent variables were dichotomous.

CHAPTER 4

RESULTS

Descriptive Analyses

Introduction

Data from the University of Alabama at Birmingham School of Dentistry were analyzed using SAS 9.2 (SAS Institute Inc., Cary, NC, USA). In this chapter, the results of univariate statistics on prevalence and incidence of ECC, descriptive analyses for behavioral and demographic variables, as well as bivariate and multivariable analyses on associations of ECC prevalence and incidence with different behavioral and demographic risk factors will be presented in detail.

Study Subjects

At baseline, 97 children had dental examinations. However, 86, 84, and 73 children had dental examinations at the 1st, 2nd, and 3rd follow-up, respectively. There were more boys than girls at all dental examinations (Table 23), constituting approximately 60% of the total number of children.

Table 23. Study children at baseline and the three follow-up examinations-by gender

Gender	Number of children (%) by gender			
	Baseline	1st follow-up	2nd follow-up	3rd follow-up
Male	55 (56.7%)	49 (57.0%)	49 (58.3%)	44 (60.3%)
Female	42 (43.3%)	37 (43.0%)	35 (41.7%)	29 (39.7%)
Total	97	86	84	73

The mean ages of the study children were 1.05, 2.01, 3.06, and 4.02 years at baseline and the three follow-up examinations, respectively, with medians slightly lower, and the age ranges were 0.3-1.9, 1.1-3.2, 2.1-4.3, and 3.1-4.9 years at baseline and the three follow-up examinations, respectively (Table 24).

Prevalence

Person-level results from the dental caries examinations in Table 25 show an increase in the prevalence of dental caries experience with age. Approximately 1.1%, 12.8%, 39.3%, and 65.8% of the children had caries experience at baseline and the three follow-up examinations, respectively, excluding the six children at baseline who did not have any erupted teeth. At baseline, there was only one child with untreated dental caries (d=1.1%), which increased to 10.5%, 21.4% and 28.8% at the three follow-up examinations, respectively. Nevertheless, there were no filled teeth at baseline (f=0%), and the prevalence of children who had filled teeth increased dramatically to 4.7%, 22.6% and 46.6% at the three follow-up examinations, respectively. There were no children with extracted teeth due to dental caries at baseline and the 1st follow-up. However, there were two (2.4%) and four (5.5%) children with extracted teeth at the 2nd follow-up and the 3rd follow-up, respectively.

Table 26 shows surface-level caries experience, including only erupted surfaces. Approximately 0.1%, 1.4%, 6.2%, and 10.4% of the surfaces had caries experience at baseline and the three follow-up examinations, respectively. The percentages of decayed surfaces increased from approximately 0.1% to 1.0% during the three-year follow-up, while the percentages of filled surfaces increased from 0% to 8.6% during the same period. There were no missing surfaces due to caries at baseline and the 1st follow-up. However, 0.3% and 0.8% of the surfaces were missing due to caries at the 2nd and the 3rd follow-up examinations, respectively.

Table 24. Age distribution of subjects by examination

Dental exam	Mean	SD	Lowest	5%	10%	25%	50%	75%	90%	95%	Highest
Baseline	1.05	0.34	0.27	0.63	0.70	0.79	0.96	1.27	1.55	1.73	1.87
1st follow-up	2.01	0.35	1.12	1.54	1.61	1.75	1.96	2.21	2.48	2.55	3.21
2nd follow-up	3.06	0.41	2.13	2.53	2.59	2.75	3.02	3.36	3.57	3.75	4.33
3rd follow-up	4.02	0.35	3.13	3.55	3.62	3.79	3.95	4.24	4.53	4.71	4.90

Table 25. Prevalence of ECC at the person-level at baseline and the three follow-up examinations*

Dental exam	Number of children	Person-level prevalence			
		d(%)	m(%)	f(%)	dmf(%)
Baseline	91	1.10	0.00	0.00	1.10
1st follow-up	86	10.47	0.00	4.65	12.79
2nd follow-up	84	21.43	2.38	22.62	39.29
3rd follow-up	73	28.77	5.48	46.58	65.75

*Six children were excluded at baseline, since no teeth were erupted.

Table 26. Percentage of surfaces with caries experience at baseline and the three follow-up examinations*

Dental examination	Number of children	Number of erupted surfaces	Percentages of surfaces			
			d(%)	m(%)	f(%)	dmf(%)
Baseline	91	3,045	0.07	0.00	0.00	0.07
1st follow-up	86	7,430	0.43	0.00	0.94	1.37
2nd follow-up	83	8,350	0.77	0.30	5.17	6.24
3rd follow-up	73	7,255	1.01	0.76	8.61	10.38

*Only erupted surfaces included.

Table 27 shows tooth-level caries experience by tooth type. The percentage of incisors that had caries experience increased from approximately 0.4% to 11.1% during the three-year follow-up. Also, the percentages of teeth that had caries experience increased from 0.0% to 4.1%, 0.0% to 15.8% and 0.0% to 31.2% for canines, 1st molars, and 2nd molars, respectively, during the three-year follow-up. The percentages of decayed incisors, canines, 1st molars and 2nd molars increased from 0.4% to 1.6%, 0% to 1.7%, 0% to 2.1% and 0% to 7.53%, respectively, during the three-year follow-up. The percentages of filled incisors, canines, 1st molars and 2nd molars increased from 0% to 7.7%, 0% to 2.4%, 0% to 13.7% and 0% to 23.6%, respectively, during the three-year follow-up. There were no missing incisors due to caries at both baseline and the 1st follow-up, but 0.7% and 1.9% of the incisors were missing due to caries at the 2nd and the 3rd follow-up examinations, respectively. In contrast, there were no missing teeth due to caries among canines, 1st molars and 2nd molars at any of the dental examinations.

Table 27. Percentages of teeth with caries experience by tooth type at baseline and the three follow-up examinations*

Dental examination	Tooth type	Number of children	Teeth number	d(%)	m(%)	f(%)	dmf(%)
Baseline	Incisors	97	481	0.41	0.00	0.00	0.41
	Canines	97	45	0.00	0.00	0.00	0.00
	1st Molars	97	79	0.00	0.00	0.00	0.00
	2nd Molars	97	4	0.00	0.00	0.00	0.00
1st follow-up	Incisors	86	687	1.9	0.00	1.75	3.64
	Canines	86	318	0.31	0.00	0.00	0.31
	1st Molars	86	344	2.03	0.00	1.17	3.29
	2nd Molars	86	137	0.00	0.00	1.46	1.46
2nd follow-up	Incisors	83	672	2.53	0.74	6.55	9.82
	Canines	83	336	1.19	0.00	0.56	1.79
	1st Molars	83	336	2.68	0.00	7.44	10.12
	2nd Molars	83	326	2.45	0.00	6.75	9.20
3rd follow-up	Incisors	73	575	1.56	1.91	7.65	11.13
	Canines	73	292	1.71	0.00	2.39	4.11
	1st Molars	73	292	2.05	0.00	13.70	15.75
	2nd Molars	73	292	7.53	0.00	23.63	31.16

*Only erupted teeth included.

Table 28 shows surface-level caries experience by tooth type. The percentages of surfaces that had caries experience by tooth type increased from 0.1% to 10.2%, 0% to 2.2%, 0% to 12.6% and 0% to 16.7% for incisors, canines, 1st molars and 2nd molars, respectively, during the three-year follow-up. The percentages of decayed surfaces among incisors, canines, 1st molars and 2nd molars increased from 0.1% to 0.6%, 0% to 0.3%, 0% to 1.0% and 0 to 2.4%, respectively, during the three-year follow-up. The percentages of filled surfaces among incisors, canines, 1st molars and 2nd molars increased from 0% to 7.7%, 0% to 1.9%, 0% to 11.6% and 0% to 14.3%, respectively, during the three-year follow-up. There were no missing surfaces among incisors due to caries at both baseline and the 1st follow-up, but 0.7% and 1.9% were missing due to caries at the 2nd and the 3rd follow-up examinations, respectively. In contrast, there were no missing surfaces due to caries among canines, 1st molars and 2nd molars at any of the dental examinations.

Tables 29 and 30 show more detailed descriptive analyses of 1st molars (B, I, L and S) and 2nd molars (A, J, K and T), respectively, by surface type (buccal, distal, mesial, lingual and occlusal) starting from the 1st follow-up, since there were few 1st molars (n=79) and 2nd molars (n=4) erupted at baseline. Tables 29 and 30 both show that occlusal surfaces had the highest caries experience compared to other surfaces, due to the presence of pits and fissures that make these surfaces more vulnerable to dental caries. The percentages of occlusal surfaces with caries experience increased from approximately 3.0% to 15.4% and 1.5% to 29.1% for 1st and 2nd molars, respectively, from the 1st follow-up to the 3rd follow-up (two-year period). Caries experience increased from 0.0% to 11.6%, 0.3% to 12.3%, 0.6% to 11.6% and 0.0% to 12.0% on the buccal, distal, mesial and lingual surfaces of the 1st molars, respectively, from the 1st follow-up to the 3rd follow-up (two-year period), as shown in Table 29. On the 2nd molars (Table 30), caries experience increased from 1.5% to 13.7%, 0.0% to 11.7%, 0.0% to 12.0% and

0.0% to 17.1% on the buccal, distal, mesial and lingual surfaces, respectively, from the 1st follow-up to the 3rd follow-up (two-year period). Caries experience increased from 3.4% to15.1%, 2.3% to 12.3%, 4.7% to 16.4%, 2.3% to 19.2% on teeth B, I, L and S (primary first molars), respectively, from the 1st follow-up to the 3rd follow-up (Table 29). On the primary second molars, caries experience increased from 0.0 %to 31.5%, 0.0% to 27.4%, 2.6% to 30.1% and 2.6% to 35.6% on teeth A, J, K and T, respectively.(Table 30).

Table 28. Percentages of surfaces with caries experience by tooth type at baseline and the three follow-up examinations*

Dental examination	Tooth type	Number of children	Erupted surfaces number	d(%)	m(%)	f(%)	dmf(%)
Baseline	Incisors	97	2,405	0.08	0.00	0.00	0.08
	Canines	97	225	0.00	0.00	0.00	0.00
	1st Molars	97	395	0.00	0.00	0.00	0.00
	2nd Molars	97	20	0.00	0.00	0.00	0.00
1st follow-up	Incisors	86	3,435	0.70	0.00	1.75	2.45
	Canines	86	1,590	0.06	0.00	0.00	0.06
	1st Molars	86	1,720	0.41	0.00	0.35	0.76
	2nd Molars	86	685	0.00	0.00	0.58	0.58
2nd follow-up	Incisors	83	3,360	0.95	0.74	6.55	8.24
	Canines	83	1,680	0.24	0.00	0.60	0.83
	1st Molars	83	1,680	1.07	0.00	6.49	7.56
	2nd Molars	83	1,630	0.61	0.00	5.71	6.32
3rd follow-up	Incisors	73	2,857	0.63	1.91	7.65	10.19
	Canines	73	1,460	0.34	0.00	1.85	2.19
	1st Molars	73	1,460	1.03	0.00	11.58	12.60
	2nd Molars	73	1,460	2.40	0.00	14.32	16.71

Table 29. Tooth-specific and surface-specific caries experience of 1st molars at the three follow-up examinations*

Dental examination**	Tooth level				Surface level		
	Tooth	Number of teeth	%dmfs		Surface	Number of surfaces	%dmfs
1st follow-up	B	86	3.44%		Buccal	344	0.00%
	I	86	2.33%		Distal	344	0.29%
	L	86	4.65%		Mesial	344	0.59%
	S	86	2.33%		Lingual	344	0.00%
					Occlusal	344	2.99%
2nd follow-up	B	84	11.9%		Buccal	336	6.89%
	I	84	9.52%		Distal	336	7.44%
	L	84	8.33%		Mesial	336	6.89%
	S	84	10.71%		Lingual	336	6.55%
					Occlusal	336	10.12%
3rd follow-up	B	73	15.05%		Buccal	292	11.64%
	I	73	12.33%		Distal	292	12.33%
	L	73	16.44%		Mesial	292	11.64%
	S	73	19.18%		Lingual	292	11.99%
					Occlusal	292	15.42%

*Only erupted teeth and surfaces included

**Baseline examination was excluded, because there were a few children with erupted 1st molars.

Table 30. Tooth-specific and surface-specific caries experience of 2nd molars at the three follow-up examinations*

Dental exam	Tooth level				Surface level		
	Tooth	Number of teeth	%dmfs		Surface	Number of surfaces	%dmfs
1st follow-up	A	28	0.00%		Buccal	137	1.46%
	J	31	0.00%		Distal	137	0.00%
	K	39	2.56%		Mesial	137	0.00%
	T	39	2.56%		Lingual	137	0.00%
					Occlusal	137	1.46%
2nd follow-up	A	81	7.41%		Buccal	326	6.13%
	J	79	7.59%		Distal	326	5.21%
	K	84	8.30%		Mesial	326	5.52%
	T	82	13.41%		Lingual	326	6.13%
					Occlusal	326	8.59%
3rd follow-up	A	73	31.51%		Buccal	292	13.70%
	J	73	27.40%		Distal	292	11.65%
	K	73	30.14%		Mesial	292	11.99%
	T	73	35.62%		Lingual	292	17.12%
					Occlusal	292	29.11%

*Only erupted teeth and surfaces included

Incidence

Ten of the children had only one dental examination, so it was not possible to include them in the assessment of incidence of dental caries, while 72 children had all dental examinations (baseline and the three follow-up exams), so they were included in the assessment of caries incidence at all study time periods (baseline to 1st follow-up, baseline to 2nd follow-up, baseline to 3rd follow-up, 1st follow-up to 2nd follow-up, 1st follow-up to 3rd follow-up and 2nd follow-up to 3rd follow-up) (Table 31).

Table 31. Presence of dental examinations at baseline and the three follow-ups

Dental examinations					
Baseline	12-M F-U	24-M F-U	36-M F-U	Number of children	Number of exams
Yes	No	No	No	10	1
Yes	Yes	No	No	3	2
Yes	Yes	Yes	No	11	3
Yes	No	Yes	Yes	1	3
Yes	Yes	Yes	Yes	72	4
Total number of children				97	1 to 4

Table 32 shows person-level crude caries incidence (CCI), when unerupted surfaces at the beginning of the time period were included (assuming that unerupted surfaces were sound, because these teeth were no exposed to the risk factors in the oral environment yet). CCI did not count for true reversals (due to remineralization of the dental lesion) nor false reversals (due to examiner's misclassification).

Table 32 shows that approximately 65.8% of the children who were examined at both baseline and 3rd follow-up (n=73) had at least one site with new caries experience during the mentioned period, with a range from 1 to 72 new sites with caries experience. Also, one year incidence rates were 12.8%, 38.6% and 58.9% starting at baseline, 1st follow-up and 2nd follow-up, respectively.

Table 32. Crude person-level incidence of ECC for all study time periods-unerupted surfaces included

Incidence period	Number of children with dental exams at both the beginning and the end of the time period	Incidence% (dmfs increment range)
Baseline to 1st follow-up	86	12.79%(1-30)
Baseline to 2nd follow-up	84	39.29%(1-64)
Baseline to 3rd follow-up	73	65.75% (1-72)
1st follow-up to 2nd follow-up	83	38.55% (1-53)
1st follow-up to 3rd follow-up	72	66.67% (1-57)
2nd follow-up to 3rd follow-up	73	58.90% (1-47)

Table 33 shows person-level net caries incidence rates (NCI), when unerupted surfaces at the beginning of the time period were included. NCI was identical to the crude caries incidence for all study time periods, except for the incidence period from 2nd follow-up to the 3rd follow-up (NCI=56.1% and CCI=58.9%), due to the presence of reversals.

Table 33. Net person-level incidence of ECC for all study time periods-unerupted surfaces included

Incidence period	Number of children with dental exams at both the beginning and the end of the time period	Incidence% (dmfs increment range)
Baseline to 1st follow-up	86	12.79% (1-30)
Baseline to 2nd follow-up	84	39.29% (1-64)
Baseline to 3rd follow-up	73	65.75% (1-72)
1st follow-up to 2nd follow-up	83	38.55% (1-53)
1st follow-up to 3rd follow-up	72	66.67% (1-57)
2nd follow-up to 3rd follow-up	73	56.16% (1-47)

Table 34 shows person-level CCI, when only erupted surfaces at the beginning of the time periods were included. Approximately 32.8% of the children who were examined at both baseline and 3rd follow-up (n=67) had at least one site with new caries experience during the three-year follow-up, with a range from 1 to 38 new sites with caries experience. Also, the one year incidence rates were 12.5%, 38.6% and 57.5% starting at baseline, 1st follow-up and 2nd follow-up, respectively.

Table 34. Crude person-level incidence of ECC for all study time periods-only erupted surfaces included

Incidence period	Number of children with dental exam at both the beginning and the end of the time period	Incidence% (dmfs increment range)
Baseline to 1st follow-up	80	12.50% (1-20)
Baseline to 2nd follow-up	78	23.08% (1-38)
Baseline to 3rd follow-up	67	32.84% (1-38)
1st follow-up to 2nd follow-up	83	38.55% (1-53)
1st follow-up to 3rd follow-up	72	59.72% (1-52)
2nd follow-up to 3rd follow-up	73	57.53% (1-47)

Table 35 shows NCI rates at the person-level, when only erupted surfaces at the beginning of the time periods were included. NCI was identical to the crude caries incidence for all study time periods, except for the incidence period from 2nd follow-up to the 3rd follow-up (NCI=54.8% and CCI=57.5%).

Table 35. Net person-level incidence of ECC for all study time periods-only erupted surfaces included

Incidence period	Number of children with dental exam at both the beginning and the end of the time period	Incidence% (dmfs increment range)
Baseline to 1st follow-up	80	12.50% (1-20)
Baseline to 2nd follow-up	78	23.08% (1-38)
Baseline to 3rd follow-up	67	32.84% (1-38)
1st follow-up to 2nd follow-up	83	38.55% (1-53)
1st follow-up to 3rd follow-up	72	59.72 %(1-52)
2nd follow-up to 3rd follow-up	73	54.79% (1-47)

The percentages of surfaces that developed new caries for all study time periods, when unerupted surfaces were included are shown in Table 36. Approximately 10.4% of the surfaces that were examined at baseline and 3rd follow-up (n=7,401) developed new caries during the three-year follow-up.

Table 36. Percentage of surfaces that developed new caries for all study time periods-unerupted surfaces included

Incidence period	# surfaces available at both the beginning and the end of the time period	% of surfaces having new caries experience
Baseline to 1st follow-up	8,772	1.14%
Baseline to 2nd follow-up	8,568	6.16%
Baseline to 3rd follow-up	7,401	10.43%
1st follow-up to 2nd follow-up	8,466	5.15%
1st follow-up to 3rd follow-up	7,299	9.43%
2nd follow-up to 3rd follow-up	7,376	5.67%

Table 37 shows the percentages of surfaces that developed new caries for all study time periods, when only erupted surfaces were included. Approximately 11.5% of the surfaces that were examined at baseline and 3rd follow-up (n=2,400) developed new caries during the three-year follow-up.

Table 37. Percentage of surfaces that developed new caries for all study time periods- only erupted surfaces included

Incidence period	# surfaces available at both the beginning and the end of the time period	% of surfaces having new caries experience
Baseline to 1st follow-up	2,695	3.04%
Baseline to 2nd follow-up	2,670	8.61%
Baseline to 3rd follow-up	2,400	11.50%
1st follow-up to 2nd follow-up	7,246	5.79%
1st follow-up to 3rd follow-up	6,310	9.11%
2nd follow-up to 3rd follow-up	7,318	5.63%

Table 38 shows the incidence at the person-level by tooth type for all study time periods, when unerupted surfaces were included. During the three-year follow-up, 35.6%, 9.6%, 27.4% and 49.3% of the children developed new caries on incisors, canines, 1st molars and 2nd molars, respectively. The percentage of children who developed new sites of caries incidence on the 2nd molars was higher than 1st molars, in spite of the fact that 1st molars erupt earlier and are exposed to the risk factors of oral environment for a longer time than 2nd molars.

Table 38. Incidence by tooth type for all study time periods-unerupted surfaces included

Incidence period	Number of children	Person-level incidence by tooth-type			
		Incisors	Canines	1st molars	2nd molars
Baseline to 1st follow-up	80	11.63%	1.16%	5.81%	1.16%
Baseline to 2nd follow-up	78	28.57%	3.57%	19.05%	14.29%
Baseline to 3rd follow-up	67	35.62%	9.59%	27.40%	49.32%
1st follow-up to 2nd follow-up	83	22.90%	3.61%	19.28%	14.46%
1st follow-up to 3rd follow-up	72	33.33%	9.72%	27.78%	50.00%
2nd follow-up to 3rd follow-up	73	23.29%	6.85%	16.44%	43.84%

When only erupted surfaces are included (Table 39), approximately 29.9%, 16.7% and 26.3% of the children developed new carious lesions on incisors, canines, 1st molars, and, respectively. Since only one child had all of his/her 2nd molars erupted and he/she was lost to follow-up, the denominators of the incidence rates from baseline were equal to zero, so the values of the incidence rates were. undetermined (any value divided by zero=undetermined value) at all incidence periods from baseline (Note: this subject had all four erupted). However, dental caries incidence on 2nd molars was substantial from the 1st follow-up to the 3rd follow-up (two years) with more than 52.6% of the children developed new sites of dental caries incidence from 1st follow-up to the 3rd follow-up. The incidence of dental caries on 2nd molars from 1st follow-up to the 2nd follow-up was even higher than dental caries incidence on 1st molars (27.8%) during the same period.

Table 39. Incidence by tooth type for all possible study time periods-only erupted surfaces included

Incidence period	Person-level incidence by tooth-type			
	Incisors (N)	Canines (N)	1st molars (N)	2nd molars (N)
Baseline to 1st follow-up	11.25% (80)	8.33% (12)	10.00% (20)	Undetermined (0)
Baseline to 2nd follow-up	23.92% (78)	8.33% (12)	20.00% (20)	undetermined (0)
Baseline to 3rd follow-up	29.85% (67)	16.67% (12)	26.32% (19)	undetermined (0)
1st follow-up to 2nd follow-up	22.90% (83)	3.80% (79)	19.28% (83)	23.81% (42)
1st follow-up to 3rd follow-up	33.33% (72)	10.00% (70)	27.78% (72)	52.63% (38)
2nd follow-up to 3rd follow-up	21.92% (73)	6.85% (73)	16.44% (73)	42.47% (73)

Descriptive Analyses/Baseline Questionnaire

Height and weight were not assessed at baseline when the children were 3-22 months old (mean age=one year), and few children had their height (mean=33.0 inches) and weight (mean=29.4 pounds) measured at the 1st follow-up (~31.3%) when the children had a mean age of two years. Mean height increased to 37.8 inches and 41.4 inches at the 2nd follow-up and 3rd follow-up when the children had mean age of three and four years, respectively. Mean weight increased to 35.5 pounds and 40.1 pounds at the 2nd follow-up and 3rd follow-up, respectively, for the different sample sizes at each time period (Table 40).

Table 40. Height and weight of children at the 1st, 2nd and 3rd follow-up*

Dental examination	Number of children	Height (inches)			Weight (pounds)		
		Mean	SD	Median	Mean	SD	Median
1st follow-up	27 (Missing=59)	32.96	3.10	33.00	29.37	7.55	27.00
2nd follow-up	83 (Missing=1)	37.79	2.34	37.00	35.51	6.75	35.00
3rd follow-up	71 (Missing=2)	41.44	2.49	41.00	40.11	9.81	38.00

* Height and weight were not assessed at baseline

Birth-related information which was collected at baseline (Table 41) showed that approximately 11.3% of the children were not delivered full-term (37 to 42 weeks) and approximately 21.7% of the children had low birth weight (<5 lbs. 8 oz.). Also, delivery process information showed that approximately 46.4% of the children were delivered by C-section procedure, while the others (53.6%) were delivered by standard procedure.

Table 41. Baseline descriptive analyses of birth-related explanatory variables

Variable	Category	Number	Percentages
Full-term delivery (37 to 42 weeks)	Yes	86	88.66%
	No	11	11.34%
Low birth weight (<5 lbs. 8 oz.)	Yes	76	78.35%
	No	21	21.65%
Delivery process	C-section	45	46.39%
	Standard	55	53.61%

Medical information collected at baseline (Table 42) showed that 20.6% of the children had a history of at least one chronic systemic disease, such as heart disease, rheumatic fever, diabetes, hepatitis, epilepsy, bone or kidney disease, being HIV positive, having a birth defect, or other chronic medical conditions. In addition, approximately

59.3% of the children reportedly used antibiotics in the previous six months and 44.3% of all children reportedly used amoxicillin, while 6.2% of all children used other kinds of antibiotics, such as ciprofloxacin. Also, more than half of the children (56.7%) had history of acute illness in the previous 6 months.

Table 42. Baseline descriptive analyses of medical explanatory variables

Variable	Category	Number	Valid percentage	Missing
Presence of systemic	Yes	20	20.62%	0
	No	77	79.38%	
Use of antibiotic in the previous six months	Yes	48	59.26%	16
	No	33	40.74%	
Period of taking antibiotics in the previous six months	None	34	40.96%	17
	1-2 week	26	31.33%	
	3-6 weeks	9	10.84%	
	7-12 weeks	8	9.64%	
	>12 weeks	6	7.23%	
Amoxicillin use	Yes	43	44.33%	0
	No	54	55.67%	
Other antibiotic use	Yes	6	6.19%	0
	No	91	93.81%	
Use of antibiotic (ever)	Yes	48	59.26%	16
	No	33	40.74%	
Presence of acute illness in the previous six months	Yes	55	56.70%	16
	No	42	43.30%	

Information about feeding practices collected at baseline (Table 43) showed that almost all of the children were never breast-fed (96.9%) and approximately 28.9% of the children had a history of ever night or nap bottle-feeding. Most of the children had a history of drinking liquids other than water throughout the day (92.78%), while the rest had a history of drinking liquids other than water at meal/snack times (7.22%).

Table 43. Baseline descriptive analyses of feeding-practices explanatory variables

Variable	Category	Number	Percentage
Breastfeeding (ever)	Yes	3	3.09%
	No	94	96.91%
Time of consumption of liquids except water (at baseline)	Throughout the day	90	92.78%
	A meal/snack times	7	7.22%
Night or nap bottle-feeding history (ever)	Yes	28	28.87%
	No	69	71.13%

Oral health behaviors information collected at baseline (Table 44) showed that approximately 41.5% of the children had their teeth brushed (13.64% reportedly brushed their own teeth) and 25.5% of the children had their teeth brushed twice a day. However, approximately 66.7% of the children did not use toothpaste. Also, none of the children had a history of use of fluoridated drops or tablets ever (0%) and only 2.1% had a regular dentist. Approximately 2.1% used well water, 75.3% used bottled water, and approximately one-third used city water (37.1%), keeping in mind that the parents had the opportunity to check more than one source of water.

Table 44. Baseline descriptive analyses of oral health behaviors

Variable	Category	Number	Valid percentage	Missing
Toothbrushing	Yes	39	41.49%	3
	No	55	58.51%	
Frequency of toothbrushing	None	55	58.51%	3
	Once/day	15	15.96%	
	Twice/day	24	25.53%	
Use of toothpaste*	Yes	28	33.33%	13
	No	56	66.67%	
Self-brushing	Yes	12	86.36%	9
	No	76	13.64%	
Use of vitamin drops or tablets with fluoride (ever)	Yes	0	0.00%	3
	No	94	100.00%	
Presence of regular dentist	Yes	2	2.08%	1
	No	94	97.92%	
Reason for the last visit to the dentist	Regular	2	100.00%	95
	Other reasons	0	0.00%	
Bottled water use	Yes	73	75.26%	0
	No	24	24.74%	
City water use	Yes	36	37.11%	0
	No	61	62.89%	
Well water use	Yes	2	2.06%	0
	No	95	97.94%	

Family-related information collected at baseline (Table 45) showed that the mean number of the children's family members was approximately 4.8 and standard deviation of 1.9, with range from 2 to 9. Twenty-fifth percentile of the number of household members was 3.0, while fiftieth percentile of the number of household members was 4.0. Seventy-fifth percentile of the number of household members was 6.0, while ninetieth percentile of number of household members was 8.0.

Also, baseline dental exams of the mothers showed that the mean DMFS score was approximately 20.7 and standard deviation of 19.9, with a range from 0 to 95. Twenty-fifth percentile of the number of DMFS was 5.0, while fiftieth percentile of the number of DMFS was 16.0. Seventy-fifth percentile of the number of DMFS was 28.0, while ninetieth percentile of number of DMFS was 50.0.

Dietary information was collected at baseline (Tables 46-48). Descriptive analyses (Table 46) showed that almost all of the children consumed milk, infant formula, water, and 100% natural juice (36.1%, 61.9%, 99.0% and 91.6%, respectively), while few children consumed other beverages (1.0%). Juices included 100% natural juice with no sugar added, such as orange juice, tomato juice, and apple juice, while "other beverages" includes all the soft drinks such as, Coke™ and other carbonated beverages, lemonade, Hi-C™, Hawaiian Punch™, Caprisun™, and Koolaid™, but not 100% natural juice. Drinks like Gatorade™, Vitamin Water™, and Red Bull™ also were defined as soft drinks, although they were sometimes called "sport drinks". Also, "other beverages" included tea, milk, and coffee. Ninety-five of the 97 (~98%) children with baseline questionnaires reported using either milk or infant formula and there were two missing data.

Table 45. Baseline descriptive analyses of family-related explanatory variables

Variable	Sample size	Missing	Mean	SD	Min	5%	10%	25%	50%	75%	90%	95%	Max
Number of household members	97	0	4.8	1.9	2.0	2.0	3.0	3.0	4.0	6.0	8.0	9.0	9.0
Mothers' DMFS	74	23	20.7	19.9	0.0	0.0	1.0	5.0	16	28	50	59	95

Table 46. Baseline dietary descriptive analyses-prevalence*

Beverage	Category	Number	Percentage
Milk	Yes	35	36.08%
	No	62	63.92%
Infant formula	Yes	60	61.85%
	No	37	38.15%
Water	Yes	96	98.96%
	No	1	1.04%
Juice*	Yes	89	91.58%
	No	8	8.42%
Other beverages**	Yes	1	1.03%
	No	96	98.97%

*Juice includes all kinds of 100% natural juice.

**Other beverages include all sugar-added beverages, including juice drinks.

Among only those consuming these categories of beverages, the means of frequency of consumption were 3.3, 4.2, 2.4, 2.4 and 3.0 times per day for milk, infant formula, water, juice, and sugar-added beverages, respectively (Table 47), and the means of quantity of consumption were 3.3, 4.2, 2.4, 2.4 and 3.0 cups per day for milk, infant formula, water, juice and other beverages, respectively (Table 48). Juices included 100% natural juice, such as orange juice, tomato juice, and apple juice. Other beverages include all the soft drinks such as, Coke™ and other carbonated beverages, lemonade, Hi-C™, Hawaiian Punch™, Caprisun™, and Koolaid™, but not 100% natural juice. Also, other beverages included tea, milk, and coffee.

Table 47. Baseline dietary descriptive analyses-frequency/day only among consumers

Variable	Number of consumers	Mean	SD	Min	5%	10%	25%	50%	75%	90%	95%	Max
Milk	35	3.3	1.0	1.0	2.0	2.0	3.0	3.0	4.0	4.0	5.0	5.0
Infant formula	60	4.2	1.4	2.0	2.0	3.0	3.0	4.0	5.0	6.0	6.5	9.0
Water	96	2.4	1.3	1.0	1.0	1.0	1.0	2.0	3.0	4.0	5.0	6.0
Juice*	89	2.4	1.0	1.0	1.0	1.0	2.0	2.0	3.0	4.0	4.0	5.0
Other beverages**	1	3.0	-	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0

*Juice includes all kinds of 100% natural juice.

**Other beverages include all sugar-added beverages, including juice drinks.

Table 48. Baseline dietary descriptive analyses-amount (cups/day) only among consumers

Variable	Number of consumer	Mean	SD	Min	5%	10%	25%	50%	75%	90%	95%	Max
Milk	35	3.3	1.0	1.0	2.0	2.0	3.0	3.0	4.0	4.0	5.0	5.0
Infant formula	60	4.2	1.2	2.0	2.0	3.0	3.0	4.0	5.0	6.0	6.0	8.0
Water	96	2.4	1.4	1.0	1.0	1.0	1.0	2.0	3.0	4.0	5.0	6.0
Juice*	89	2.4	1.1	1.0	1.0	1.0	2.0	2.0	3.0	4.0	4.0	6.0
Other beverages**	1	3.0	-	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0	3.0

*Juice includes all kinds of 100% natural juice.

**Other beverages include all sugar-added beverages, including juice drinks.

Descriptive Analyses/Repeated Questionnaires

As mentioned in Chapter Three, dietary information (frequencies and quantities of different kinds of beverages, sweets and sweetened foods) was also collected at the visits numbered 10, 40, 50, 60, 70 and 90, when the mean ages of the children were 0.9, 1.5, 1.9, 2.5, 3.1 and 4.1 years, respectively (Table 49). As of the time of these analyses, information at visit 90 had been collected for only 23 children, so information at that visit was excluded from the analyses.

Table 49. Number of children with questionnaires at each visit and mean ages (SD)

Questionnaire	Number of children	Mean age (SD)
Visit 10 (recruitment)	97	0.96 (0.30)
Visit 40	89	1.51 (0.31)
Visit 50 (1st follow-up exam)	87	1.99 (0.33)
Visit 60	85	2.49 (0.32)
Visit 70 (2nd follow-up exam)	81	3.06 (0.40)
Visit 90 (3rd follow-up exam)	23	4.07 (0.32)

Area-under-the-curve analyses (AUC) of responses numbered 40 to 70 (40, 50, 60 and 70) were conducted, including only children who had questionnaires both at visits 40 and 70, while not requiring the presence of responses at visits 50 and 60. AUC of responses 40 to 70 showed that mean daily frequencies of consumption of milk, water, 100% juice and sugar-added beverages were 3.4, 0.9, 4.1 and 1.9 times, respectively (Table 28). It is important to mention that the frequencies (times/day) and the quantities (cups/day) of all the beverages at all visits were almost identical (99.4% identical over all time periods and intervals).

Table 50 shows that the AUC daily frequency of consumption of sweets, such as candy and gum, was low (mean=0.15 times per day) compared to the AUC frequency of consumption of sweetened foods, such as sugared cereals and Pop Tarts™ (mean=1.7 times per day). However, none of the parents reportedly added sugar to the children's food or drinks (0%, AUC). Oral hygiene information was also collected at visits 40, 50, 60 and 70. AUC analyses of responses at visits 40 to 70 showed that the mean daily frequency of toothbrushing was 1.7 (Table 50). Table 29 shows that approximately 95% of the children used toothpaste (AUC) (add more notes about toothpaste). Also, more than half of the children reportedly brushed their own teeth and had a history of previous dentist visit (both 57%), while approximately 25% of the children had a regular dentist (AUC, Table 51). However, none of the children reportedly took dietary fluoride supplements (0%, AUC).

Table 50. Descriptive analyses of the daily continuous dietary and oral hygiene variables (AUC visits 40 to 70*)

Continuous variables**	N	Mean	SD	Median	Min	Max
Daily frequency of consumption of milk	81	3.36	1.00	3.51	0.00	5.00
Daily frequency of consumption of 100% juice	81	0.88	0.63	0.82	0.00	2.28
Daily frequency of consumption of water	81	4.09	0.89	4.12	1.57	6.17
Daily frequency of consumption of sugar-added beverages***	81	1.88	0.90	1.92	0.27	4.04
Daily frequency of consumption of sweets****	78	0.15	0.26	0.01	0.00	1.09
Daily frequency of consumption of sweetened foods*****	77	1.65	0.46	1.82	0.21	3.00
Daily frequency of brushing	78	1.77	0.35	2.00	0.72	2.17

*Area-under-the-curve for visits 40, 50, 60 and 70.

** None of the children reportedly added sugar to food or drinks.

***Sugar-added beverages include all beverages, except milk, water, 100% juice.

****Sweets include candy, gum, etc.

*****Sweetened foods include Pop Tarts™, sugared cereals, etc.

Table 51. Descriptive analyses of the daily dichotomous dietary and oral hygiene* variables (AUC visits 40 to 70**)

Variables	Number	Category	Percentage
Consumption of sweets***(yes/no)	78	Yes	19%
		No	81%
Consumption of sweetened foods**** (yes/no)	77	Yes	99%
		No	1%
Consumption of milk*****	87	Yes	99%
		No	1%
Consumption of 100% juice	87	Yes	84%
		No	16%
Consumption of water	87	Yes	99%
		No	1%
Consumption of sugar-added beverages	87	Yes	99%
		No	1%
Use of toothpaste (yes/no)	76	Yes	94%
		No	6%
Self-brushes (yes/no)	76	Yes	56%
		No	44%
Regular dentist (yes/no)	74	Yes	25%
		No	75%
Seen dentist-ever (yes/no)	66	Yes	57%
		No	43%

* None of the children reportedly took dietary fluoride supplements.

** Area-under-the-curve at visits 40, 50, 60 and 70.

*** Sweets include candy, gum, etc.

**** Sweetened foods include Pop Tarts™, sugared cereals, etc.

***** All children reportedly consumed cow's milk at all visits, except one child at only visit 40 who reportedly consumed soy milk.

Table 52 shows that the mean daily frequencies of consumption of milk, water, 100% juice and sugar-added beverages at the 2nd follow-up (visit 70) were 3.3, 2.0, 4.7 and 0.7 times, respectively. In addition, approximately 37% of the children reportedly consumed sweets, such as candy and gum (Table 53), with mean frequency of 0.35 times per day, while all the children reportedly consumed sweetened foods (100%), such as Pop Tarts™ and sugared cereals, with mean frequency of 1.76 times per day (Tables 52 and 53). However, none of the parents reportedly added sugar to the children's food or drinks (Table 53).

Table 52 also shows that the mean daily frequency of tooth brushing among the study children at the 2nd follow-up (visit 70) was 1.9 times. Table 53 shows that all the children used toothpaste (100%). Approximately 78%, 51% and 64% of the children reportedly brushed their own teeth, had a regular dentist and had a history of previous dental visit, respectively (Table 53). However, none of the children reportedly took fluoride supplements (Table 53).

Table 52. Descriptive analyses of the daily continuous dietary and oral hygiene variables at the 2nd follow-up (visit 70)

Continuous variables*	N	Mean	SD	Median	Min	Max
Frequency of consumption of milk per day	81	3.25	1.30	3.00	0	5.00
Frequency of consumption of 100% juice per day	81	1.99	1.39	2.00	0	5.00
Frequency of consumption of water per day	81	4.65	1.09	5.00	0	6.00
Frequency of consumption of sugar-added beverages per day	81	0.69	1.23	0	0	4.00
Frequency of consumption of sweets** per day	79	0.35	0.58	0	0	2.00
Frequency of consumption of sweetened foods per day	79	1.76	0.53	2.00	0.29	3.00
Frequency of brushing per day	80	1.94	0.24	2.00	1.00	2.00

* None of the parents reportedly added sugar to the children's food or drinks.

** Sweets include candy, gum, etc.

Table 53. Descriptive analyses of the daily dichotomous dietary and oral hygiene[*] variables at the 2nd follow-up (visit 70)

Variabes	Number	Category	Percentage
Consumption of sweets[**](yes/no)	79	Yes	37%
		No	63%
Consumption of sweetened foods[***] (yes/no)	79	Yes	100%
		No	0%
Use of toothpaste (yes/no)	81	Yes	100%
		No	0%
Self-brushes (yes/no)	81	Yes	78%
		No	22%
Regular dentist (yes/no)	76	Yes	51%
		No	49%
Seen dentist-ever (yes/no)	73	Yes	64%
		No	36%

*None of the children reportedly took dietary fluoride supplements.

**Sweets include candy, gum, etc.

***Sweetened foods include Pop Tarts™, sugared cereals, etc.

At the baseline and follow-up questionnaires, the parents were asked about the brand names of the toothpaste their children used and the question was an open-ended question. Each of the parents at each of the visits mentioned only one brand name, although there were no restrictions for providing more than one brand name. The purpose of asking the parents to provide the brand names of the toothpaste was to know about the fluoride content of the toothpaste. However, because the question was an open-ended question and the parents completed the questionnaires themselves with the interviewers providing help only upon request, the provided information was not enough to decide whether the toothpastes used by the children were fluoridated or not. For example, many parents reported that their kids used Orajel™ toothpaste, without giving more details. According to the Orajel™ website, there are two main types of Orajel™: "Orajel™ for Kids, which is fluoridated, and "Orajel™ for Toddlers", which is non-fluoridated. Thus, it is difficult to define a fluoride status based on the information provided. Also, some parents reported that their children used "Oral-B™ training gel". However, there is no such brand name in the market. Instead, there is "Oral-B™ Stages", which is fluoridated according to Oral-B™ website. However, Oral-B™ customer service staff explained over the telephone that non-fluoridated Oral-B™ Stages toothpaste is also available in the market[1]. Table 54 summarizes all the brand names of toothpaste the children reportedly used at the baseline and follow-up visits. Table 54 shows that, at recruitment (visit 10), information about brand name of toothpaste was obtained for 28 (33.3%) children. At visit 40 (mean age of the children=1.5 years approximately), information about brand name of toothpaste was obtained for 63 children (75%). At visits 50, 60 and 70 when the children were approximately 2, 2.5 and 3 years, information about brand name of toothpaste was obtained for 80 (97.6%), 82 (98.8%) and 80 (100%) of the children, respectively. Most of the children reportedly used either Colgate™ or Crest™, while few children reportedly used Thomas The train™, Oral-B Training Gel™, Orajel™, Ultra Closeup™ or Aquafresh™. However, data were not available from the University of Alabama at Birmingham (UAB) about the brand name of the toothpaste they provided to the study subjects as part of the benefits provided to all the children and their families at every visit.

Table 54. Brand names of toothpastes the children reportedly used at baseline and follow-up visits and the total number of children at each visit with completed questionnaires

Visit	Number of children who used toothpaste							Total number of children
	Colgate™	Oral-B™	Crest™	Aim™	Others (N)	Missing	Total	
10	16	1	6	0	Thomas the Train (1) Oral-B Training Gel (1) Orajel (2)	1	28	84
40	26	4	17	1	Thomas the Train (1) Oral-B Training Gel (1) Orajel (5) Ultra/Closeup (1) Unknown (3)	4	63	84
50	46	0	19	2	Thomas The train (1) Oral-B Training Gel (2) Orajel (7) Ultra/Closeup (1) Aquafresh (1) Unknown (1)	0	80	82
60	45	0	22	1	Thomas the Train (1) Oral-B Training Gel (2) Orajel (7) Ultra/Closeup (1) Unknown (3)	0	82	83
70	46	0	25	1	Thomas the Train (1) Oral-B Training Gel (2) Orajel (4) Unknown (1)	1	81	81

Bivariate Analyses

Introduction

Bivariate analyses were conducted with SAS 9.3 (SAS Institute Inc., Cary, NC, USA), using logistic regression and negative binomial models for dichotomous and count dependent variables, respectively. The associations between the incidence of ECC from baseline to the 3rd follow-up (dichotomous/count) and the three one-year incidence (dichotomous/count) and different risk factors were assessed. A significance level of $\alpha=0.15$ was used as a cutoff for inclusion in the multivariable analyses.

Incidence as a Dichotomous Dependent Variable with Baseline Information

Table 55 shows that there were no statistically significant relationships between the incidence of ECC (dichotomous) from baseline to 1st follow-up, 1st follow-up to 2nd follow-up, 2nd follow-up to 3rd follow-up or baseline to 3rd follow-up and sex of the child. There was a statistically significant relationship between ECC incidence (dichotomous) and age of the child in months at the beginning of the time period for the baseline to 1st follow-up (OR=1.24) and 2nd follow-up to 3rd follow-up (OR=0.92), with P-values of 0.01 and 0.09, respectively. There was a statistically significant association between ECC incidence (dichotomous) from the 2nd follow-up to 3rd follow-up time (P-value=0.11) and BMI at the beginning of the time period, with those who had higher BMI having lower ECC incidence (OR=0.85). The associations between BMI and ECC incidence from baseline to 1st follow-up and baseline to 3rd follow-up were not assessed, because BMI was not measured at baseline.

Table 55. Bivariate analyses of associations of baseline demographic factors with ECC incidence rates* (dichotomous) for the three one-year periods and the three-year period**

Factor	Incidence period							
	Baseline to 1st follow-up		1st follow-up to 2nd follow-up		2nd follow-up to 3rd follow-up		Baseline to 3rd follow-up	
	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value
Child's sex Male Female	ref 1.12 (0.32-3.99)	0.86	ref 0.98 (0.39-2.41)	0.97	ref 1.64 (0.64-4.26)	0.32	ref 0.99 (0.37-2.64)	0.98
Age (months) at the beginning of the time period	1.24 (1.06-1.46)	**0.01**	1.03 (0.93-1.15)	0.53	0.92 (0.83-1.01)	**0.09**	1.03 (0.91-1.16)	0.66
BMI at the beginning of the time period	***		1.05 (0.89-1.25)	0.55	0.85 (0.69-1.04)	**0.11**	***	

* dmfs (yes/no) at the cavitated level.

** Logistic regression was used in the analyses.

*** BMI was defined as (weight in lb. /2.2)/ (height in inches**2/ (39.37**2)).

Table 56 shows that there was a statistically significant association between premature delivery and ECC incidence from baseline to 3rd follow-up (P-value=0.08), with those who were delivered prematurely having lower ECC incidence (OR=0.29). However, there was no statistically significantly association between premature delivery and ECC incidence from baseline to 1st follow-up, 1st follow-up to 2nd follow-up or 2nd follow-up to 3rd follow-up time periods. There was no statistical significant association between delivery type and ECC incidence from baseline to 1st follow-up, 1st follow-up to 2nd follow-up, 2nd follow-up to 3rd follow-up or baseline to 3rd follow-up. Low birth weight was statistically significantly associated with ECC incidence from 1st follow-up to 2nd follow-up (P-value=0.12), with those who had low birth weight having less ECC incidence (OR=0.38). However, low birth weight was not statistically significantly associated with ECC incidence from baseline to 1st follow-up, 2nd follow-up to 3rd follow-up or baseline to 3rd follow-up.

Table 57 shows that there were no statistically significant associations between different medical risk factors, such as presence of systemic diseases, presence of acute illness, use of antibiotics and amoxicillin use, and ECC incidence (dichotomous). However, there was a statistically significant association between antibiotic use other than amoxicillin and ECC incidence (dichotomous) from baseline to 1st follow-up time period only (P-value=0.14), with those who used antibiotics other than amoxicillin having lower ECC incidence.

Table 58 shows that there were no statistically significant associations between different feeding-practices risk factors, such as history of breastfeeding, time of consumption of liquids other than water and sleeping with a bottle, and ECC incidence from baseline to 1st follow-up, 1st follow-up to 2nd follow-up, 2nd follow-up to 3rd follow-up or baseline to 3rd follow-up. Table 59 shows that there were statistically significant associations between toothbrushing (yes/no) and frequency of toothbrushing and ECC incidence (dichotomous) from the 1st follow-up to 2nd follow-up only (P-values=0.08 and 0.07, respectively). Toothbrushing and increased frequency of toothbrushing were associated with lower incidence of ECC during this time period, with odds ratios of 0.42 and 0.59, respectively.

Table 60 shows that having higher number of household members was associated with lower ECC incidence (dichotomous) from the baseline to 1st follow-up (OR=0.72), with P-value 0f 0.11.

Table 56. Bivariate analyses of associations of baseline birth-related factors with ECC incidence rates* (dichotomous) for the three one-year incidence periods and the three-year period**

Baseline factor	Incidence period							
	Baseline to 1st follow-up		1st follow-up to 2nd follow-up		2nd follow-up to 3rd follow-up		Baseline to 3rd follow-up	
	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value
Premature delivery***	****	0.35	0.34 (0.07-1.81)	0.22	0.50 (0.13-1.95)	0.32	0.29 (0.08-1.14)	**0.08**
Delivery Standard C-section	Ref 0.16 (0.16-2.18)	0.43	Ref 1.22 (0.51-2.96)	0.67	Ref 1.07 (0.43-2.67)	0.89	Ref 0.52 (0.19-1.39)	0.19
Low birth weight*****	0.34 (0.04-2.86)	0.32	0.38 (0.11-1.28)	**0.12**	1.08 (0.36-3.29)	0.89	0.59 (0.19-1.85)	0.37

* dmfs (yes/no) at the cavitated level.

** Logistic regression was used in the analyses.

*** Full term delivery was defined as 37-42 weeks.

**** OR is indeterminate, since no premature children had ECC incidence from baseline to 1st follow-up (P-value from Fisher's exact test).

***** Low birth weight was defined as <5 lbs. 8 oz.

Table 57. Bivariate analyses of associations of baseline medical factors with ECC incidence rates* (dichotomous) for the three one-year incidence periods and the three-year period**

Baseline factor	Incidence period							
	Baseline to 1st follow-up		1st follow-up to 2nd follow-up		2nd follow-up to 3rd follow-up		Baseline to 3rd follow-up	
	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value
Presence of systemic diseases	1.39 (0.33-5.82)	0.66	1.37 (0.48-3.94)	0.57	1.40 (0.41-4.78)	0.59	1.93 (0.48-7.78)	0.36
Presence of acute illness in the previous six months	1.38 (0.38-5.09)	0.64	1.29 (0.53-3.18)	0.57	0.74 (0.29-1.87)	0.52	1.09 (0.42-2.88)	0.86
No use of antibiotics in the previous six months	0.39 (0.08-2.04)	0.27	0.87 (0.33-2.29)	0.77	1.12 (0.39-3.19)	0.83	0.81 (0.27-2.51)	0.43
Amoxicillin use (ever)	1.01 (0.29-3.59)	0.99	1.27 (0.52-3.07)	0.61	1.26 (0.49-3.20)	0.63	1.96 (0.71-5.39)	0.19
Antibiotic use other than amoxicillin (ever)	0.39 (0.63-24.68)	**0.14**	0.79 (0.14-4.55)	0.79	1.69 (0.15-19.44)	0.68	1.05 (0.09-12.09)	0.98

* dmfs (yes/no) at the cavitated level.

**Logistic regression was used in the analyses.

Table 58. Bivariate analyses of associations of baseline feeding-practices factors with ECC incidence rates* (dichotomous) for the three one-year periods and the three-year incidence period**

Baseline factor	Incidence period							
	Baseline to 1st follow-up		1st follow-up to 2nd follow-up		2nd follow-up to 3rd follow-up		Baseline to 3rd follow-up	
	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value
History of breastfeeding (ever)	3.65 (0.31-44.02)	0.31	1.62 (0.09-26.73)	0.74	***	1.00	***	1.00
Time of consumption of liquids besides water Throughout the day At meal/snack	Ref 1.15 (0.13-10.58)	0.91	Ref 1.22 (0.26-5.82)	0.81	Ref 0.39 (0.07-2.23)	0.29	Ref 0.49 (0.09-2.63)	0.41
Night or nap bottle feeding history (ever)	1.47 (0.39-5.55)	0.57	0.86 (0.33-2.27)	0.76	0.75 (0.28-1.99)	0.57	1.07 (0.38-2.98)	0.91

* dmfs (yes/no) at the cavitated level.

** Logistic regression was used in the analyses.

*** OR is indeterminate, since the single child with breastfeeding history had ECC incidence. (P-values from Fisher's exact test).

Table 59. Bivariate analyses of associations of baseline dental factors with ECC incidence rates* (dichotomous) for the three one-year periods and the three-year period**

Baseline factor	Incidence period							
	Baseline to 1st follow-up		1st follow-up to 2nd follow-up		2nd follow-up to 3rd follow-up		Baseline to 3rd follow-up	
	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value
Toothbrushing	1.49 (0.37-6.04)	0.58	0.42 (0.16-1.11)	**0.08**	0.96 (0.37-2.49)	0.93	0.88 (0.32-2.37)	0.79
Daily frequency of toothbrushing	1.19 (0.55-2.59)	0.66	0.59 (0.33-1.04)	**0.07**	1.01 (0.58-1.74)	0.99	0.89 (0.57-1.58)	0.69
Use of tooth paste	1.87 (0.49-7.13)	0.36	0.52 (0.19-1.38)	0.19	0.58 (0.21-1.59)	0.29	0.52 (0.18-1.52)	0.23
Use of bottled water	1.64 (0.33-8.23)	0.55	0.93 (0.33-2.59)	0.88	0.96 (0.33-2.59)	0.94	0.68 (0.21-2.17)	0.51
Use of city water	1.08 (0.29-4.02)	0.92	0.84 (0.33-2.14)	0.71	1.44 (0.53-3.92)	0.48	1.74 (0.59-5.18)	0.33

Table 59-Continued

Use of well water	***	1.00	***	0.52	0.82 0.05-13.64)	0.89	0.51 (0.03-8.52)	0.64
Presence of regular dentist	****	1.00	****	0.53	****	0.21	****	0.12

* dmfs (yes/no) at the cavitated level.

** Logistic regression was used in the analyses.

*** OR is indeterminate, since the two children who reportedly consumed well water had no ECC incidence (P-value from Fisher's exact test).

**** OR is indeterminate, since two children with regular dentist had no ECC incidence (P-value from Fisher's exact test).

Table 60. Bivariate analyses of associations of baseline family-related factors with ECC incidence rates* (dichotomous) for the three one-year periods and the three-year period**

Baseline factor	Incidence period							
	Baseline to 1st follow-up		1st follow-up to 2nd follow-up		2nd follow-up to 3rd follow-up		Baseline to 3rd follow-up	
	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value
Number of household members	0.72 (0.48-1.68)	**0.11**	0.98 (0.77-1.26)	0.88	1.08 (0.83-1.41)	0.59	1.11 (0.83-1.48)	0.49
Mother's DMFS***	1.02 (0.98-1.06)	0.37	1.02 (0.99-1.04)	0.35	0.99 (0.97-1.03)	0.89	0.99 (0.97-1.03)	0.88

* dmfs (yes/no) at the cavitated level.

**Logistic regression was used in the analyses.

*** Dental caries was assessed at the cavitated-level only.

Table 61 shows that there were no statistically significant relationships between the frequencies of consumption of milk, 100% juice and sugar added beverages and ECC incidence (dichotomous) from baseline to 1st follow-up, 1st follow-up to 2nd follow-up, 2nd follow-up to 3rd follow-up or baseline to 3rd follow-up time periods. Increased frequency of water consumption was associated with higher incidence of ECC (dichotomous) for baseline to 1st follow-up time period (OR=1.38), with P-value of 0.15. Increased frequency of consumption of infant formula was associated with lower incidence of ECC (dichotomous) from baseline to 1st follow-up time period (OR=0.65), with P-value of 0.03.

Table 62 shows that there were no statistically significant relationships between the quantities of consumption of milk, 100% juice and sugar-added beverages and ECC incidence (dichotomous) from baseline to 1st follow-up, 1st follow-up to 2nd follow-up, 2nd follow-up to 3rd follow-up or baseline to 3rd follow-up. Increased frequency of water consumption was associated with higher incidence of ECC (dichotomous) from 2nd follow-up to 3rd follow-up (OR=1.31), with P-value of 0.13. Increased frequency of consumption of infant formula was associated with lower incidence of ECC (dichotomous) from baseline to 1st follow-up time period (OR=0.65), with P-value of 0.03.

Natural Juice (100% juice) included all juices without added sugar, such as tomato juice, orange juice, apple juice, etc. Other juices (sugar-added beverages) included all juice drinks with added sugar, such as Coke™ and other carbonated beverages, lemonade, Hi-C™, Hawaiian Punch™, Caprisun™, Koolaid™, Gatorade™, Vitamin Water™, and Red Bull™.

Table 61. Bivariate analyses of associations of baseline frequency of beverages consumption (times/day) with ECC incidence rates* (dichotomous) for the three one-year periods and the three-year period**

Baseline frequency of intake of beverage type	Incidence period							
	Baseline to 1st follow-up		1st follow-up to 2nd follow-up		2nd follow-up to 3rd follow-up		Baseline to 3rd follow-up	
	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value
Milk	1.26 (0.88-1.79)	0.21	0.83 (0.63-1.09)	0.19	0.88 (0.67-1.15)	0.33	0.89 (0.67-1.17)	0.39
Water	1.38 (0.89-2.14)	**0.15**	0.82 (0.58-1.16)	0.26	1.29 (0.89-1.83)	0.17	1.23 (0.85-1.79)	0.29
100% juice	1.41 (0.82-2.42)	0.22	1.62 (0.69-1.51)	0.92	1.09 (0.74-1.67)	0.69	1.14 (0.76-1.73)	0.54
Sugar-added beverages	1.39 (0.81-2.39)	0.24	1.05 (0.71-1.55)	0.82	1.06 (0.72-1.57)	0.77	1.16 (0.77-1.75)	0.49
Infant formula	0.65 (0.44-0.94)	**0.03**	1.02 (0.85-1.23)	0.88	1.04 (0.86-1.25)	0.73	1.02 (0.84-1.24)	0.89

* dmfs (yes/no) at the cavitated level.

**Logistic regression was used in the analyses.

Table 62. Bivariate analyses of associations of baseline amount of beverages consumption (cups/day) with ECC incidence rates* (dichotomous) for the three one-year periods and the three-year period**

Baseline quantity of intake beverage type	Incidence period							
	Baseline to 1st follow-up		1st follow-up to 2nd follow-up		2nd follow-up to 3rd follow-up		Baseline to 3rd follow-up	
	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value	OR (95%CI)	P-value
Milk	1.26 (0.89-1.79)	0.21	0.83 (0.63-1.09)	0.19	0.88 (0.67-1.15)	0.33	0.89 (0.67-1.15)	0.33
Water	1.32 (0.87-2.01)	0.19	0.81 (0.58-1.15)	0.23	1.31 (0.93-1.86)	**0.13**	1.25 (0.87-1.79)	0.24
100% juice	1.35 (0.82-2.23)	0.25	1.02 (0.69-1.48)	0.95	1.09 (0.75-1.58)	0.69	1.14 (0.76-1.69)	0.55
Sugar-added beverages	1.37 (0.84-2.24)	0.22	1.09 (0.77-1.57)	0.63	1.02 (0.71-1.48)	0.91	1.11 (0.75-1.64)	0.61
Infant formula	0.65 (0.45-0.94)	**0.03**	1.04 (0.86-1.26)	0.73	1.01 (0.83-1.23)	0.91	0.99 (0.81-1.22)	0.96

* dmfs (yes/no) at the cavitated level.

**Logistic regression was used in the analyses.

Incidence as a Count Variable with Baseline Information

Table 63 shows that there were no statistically significant relationships between the incidence of ECC (count) and sex or age of the child in months from baseline to 1^{st} follow-up, 1^{st} follow-up to 2^{nd} follow-up, 2^{nd} follow-up to 3^{rd} follow-up or baseline to 3^{rd} follow-up. There was a statistically significant association between ECC incidence (count) from the 2^{nd} follow-up to 3^{rd} follow-up (P-value=0.02) and BMI at the beginning of the time period, with those who had higher BMI having lower ECC incidence (linear coefficient estimate=-0.26). The associations between BMI and ECC incidence (count) from the baseline to 1^{st} follow-up and baseline to 3^{rd} follow-up were not assessed, because BMI was not measured at baseline.

Table 64 shows that there were no statistically significant associations between ECC incidence (count) and premature delivery from baseline to 1st follow-up, 1st follow-up to 2nd follow-up, 2nd follow-up to 3rd follow-up or baseline to 3rd follow-up. There was no statistically significant association between delivery type and ECC incidence (count) from baseline to 1st follow-up, 1^{st} follow-up to 2^{nd} follow-up, 2^{nd} follow-up to 3^{rd} follow-up or baseline to 3^{rd} follow-up. Low birth weight was not statistically significantly associated with ECC incidence (count) from baseline to 1^{st} follow-up, 1^{st} follow-up to 2^{nd} follow-up, 2^{nd} follow-up to 3^{rd} follow-up or baseline to 3^{rd} follow-up.

Table 65 shows that there were no statistically significant associations between different potential medical risk factors, such as presence of systemic diseases, presence of acute illness, use of antibiotics, amoxicillin use or antibiotic use other than amoxicillin, and ECC incidence (count) from the baseline to 1^{st} follow-up, 1^{st} follow-up to 2^{nd} follow-up, 2^{nd} follow-up to 3^{rd} follow-up or baseline to 3^{rd} follow-up.

Table 63. Bivariate analyses of associations of demographic factors with ECC incidence rates[*] (count) at the three one-year periods and the three-year incidence period[**]

Baseline factor	Incidence period							
	Baseline to 1st follow-up		1st follow-up to 2nd follow-up		2nd follow-up to 3rd follow-up		Baseline to 3rd follow-up	
	Linear coefficient estimate (SE)	P-value	Linear coefficient estimate (SE)	P-value	Linear coefficient estimate (SE)	P-value	Linear coefficient estimate (SE)	P-value
Child's sex Male Female	ref 0.89 (1.07)	0.40	ref 0.59 (0.59)	0.33	ref 0.29 (0.47)	0.53	ref 0.46 (0.44)	0.29
Age at the beginning of the incidence period	0.17 (0.18)	0.33	0.00 (0.07)	0.99	0.03 (0.06)	0.65	0.07 (0.05)	0.26
Body mass index[***]	****		-0.03 (0.11)	0.82	-0.26 (0.11)	**0.02**	****	

* dmfs at the cavitated level.

**Negative binomial model was used in the analyses.

*** BMI was defined as (weight in lb. /2.2)/ (height in inches**2/ (39.37**2)).

****BMI was not assessed at baseline.

Table 64. Bivariate analyses of associations of baseline birth-related factors with ECC incidence rates* (count) at the three one-year periods and the three-year period**

Baseline factor	Incidence period							
	Baseline to 1st follow-up		1st follow-up to 2nd follow-up		2nd follow-up to 3rd follow-up		Baseline to 3rd follow-up	
	Linear coefficient estimate (SE)	P-value	Linear coefficient estimate (SE)	P-value	Linear coefficient estimate (SE)	P-value	Linear coefficient estimate (SE)	P-value
Premature delivery***	****		-0.56 (0.91)	0.54	-0.11 (0.69)	0.87	-0.41 (0.63)	0.52
Delivery Process Standard C-section	Ref -0.15 (1.88)	0.89	Ref -0.11 (0.59)	0.86	Ref -0.12 (0.46)	0.79	Ref -0.44 (0.43)	0.31
Low birth weight*****	-1.85 (1.36)	0.18	-0.59 (0.72)	0.42	0.57 (0.54)	0.29	-0.04 (0.52)	0.95

* dmfs at the cavitated level.

** Negative binomial model was used in the analyses.

*** Full term delivery was defined as 37-42 weeks.

**** The iteration process did not converge.

***** Low birth weight was defined as <5 lbs. 8 oz.

Table 65. Bivariate analyses of associations of baseline medical factors with ECC incidence rates* (count) for the three one-year periods and the three-year period**

Baseline factor	Incidence period							
	Baseline to 1st follow-up		1st follow-up to 2nd follow-up		2nd follow-up to 3rd follow-up		Baseline to 3rd follow-up	
	Linear coefficient estimate (SE)	P-value	Linear coefficient estimate (SE)	P-value	Linear coefficient estimate (SE)	P-value	Linear coefficient estimate (SE)	P-value
Presence of systemic diseases	0.73 (1.29)	0.57	0.52 (0.71)	0.47	-0.14 (0.61)	0.82	0.48 (0.56)	0.39
Presence of acute illness in the previous six months	0.88 (1.08)	0.42	0.24 (0.59)	0.69	0.29 (0.46)	0.54	0.23 (0.43)	0.59
No use of antibiotics in the previous six months	-0.87 (1.24)	0.49	-0.19 (0.64)	0.77	-0.19 (0.44)	0.68	0.74 (0.76)	0.33
Amoxicillin use (ever)	0.77 (1.07)	0.48	0.13 (0.59)	0.83	0.59 (0.47)	0.21	0.59 (0.43)	0.17
Antibiotic use other than amoxicillin (ever)	0.59 (2.11)	0.78	0.79 (1.23)	0.52	-0.56 (1.15)	0.63	-0.06 (1.11)	0.34

* dmfs at the cavitated level.

* Negative binomial model was used in the analyses.

Table 66 shows that there were no statistically significant associations between different feeding-practices risk factors, such as history of breastfeeding and time of consumption of liquids other than water, and ECC incidence from baseline to 1st follow-up, 1st follow-up to 2nd follow-up, 2nd follow-up to 3rd follow-up or baseline to 3rd follow-up time periods. However, sleeping with the bottle was associated with less incidence of ECC (count) for the 2nd follow-up to 3rd follow-up time period (linear coefficient estimate=-0.72), with P-value of 0.15.

Table 67 shows there were no statistically significant associations between toothbrushing, daily frequency of toothbrushing, use of toothpaste, bottled water, city water and ECC incidence (count) for baseline to 1st follow-up, 1st follow-up to 2nd follow-up, 2nd follow-up to 3rd follow-up or baseline to 3rd follow-up. Fluoride content of toothpaste was not determined because of the limited available information (parents did not provide us with enough information to rule in or rule out the presence of fluoride in the toothpaste). There were very few children with reported the use of well water, fluoridated vitamin drops, and presence of regular dentist, so an error was reported in logistic regression showing that the iteration process did not converge, when using negative binomial models.

Table 68 shows that higher mother's DMFS was significantly associated with higher ECC incidence (count) from the baseline to 1st follow-up time (linear coefficient estimate=0.08), and baseline to the 3rd follow-up (linear coefficient estimate=0.02), with P-values of 0.10 and 0.13, respectively. Number of household members was not associated with ECC incidence (count) for baseline to 1st follow-up, 1st follow-up to 2nd follow-up, 2nd follow-up to 3rd follow-up or baseline to 3rd follow-up.

Table 66. Bivariate analyses of associations of baseline feeding-practices factor with ECC incidence rates[*] (count) for the three one-year periods and the three-year period[**]

Baseline factor	Incidence period							
	Baseline to 1st follow-up		1st follow-up to 2nd follow-up		2nd follow-up to 3rd follow-up		Baseline to 3rd follow-up	
	Linear coefficient estimate (SE)	P-value	Linear coefficient estimate (SE)	P-value	Linear coefficient estimate (SE)	P-value	Linear coefficient estimate (SE)	P-value
Breastfeeding (ever)	0.57 (2.93)	0.82	-1.25 (1.98)	0.53	2.04 (1.85)	0.27	1.53 (1.81)	0.40
Time of consumption of liquids besides water Throughout the day At meal/snack times	Ref -2.17 (2.16)	0.32	Ref 0.56 (1.13)	0.62	Ref 0.17 (0.89)	0.85	Ref 0.29 (0.78)	0.72
Night or nap bottle feeding history (ever)	-0.49 (1.19)	0.68	-0.26 (0.65)	0.69	-0.72 (0.49)	**0.15**	-0.38 (0.46)	0.41

* dmfs at the cavitated level.

** Negative binomial model was used in the analyses.

Table 67. Bivariate analyses of associations of baseline dental factors with ECC incidence rates* (count) for the three one-year periods and the three-year period**

Baseline factor	Incidence period							
	Baseline to 1st follow-up		1st follow-up to 2nd follow-up		2nd follow-up to 3rd follow-up		Baseline to 3rd follow-up	
	Linear coefficient estimate (SE)	P-value	Linear coefficient estimate (SE)	P-value	Linear coefficient estimate (SE)	P-value	Linear coefficient estimate (SE)	P-value
Toothbrushing	1.06 (1.19)	0.38	-0.49 (0.67)	0.47	-0.51 (0.49)	0.31	-0.09 (0.47)	0.85
Daily frequency of toothbrushing	0.53 (0.76)	0.49	-0.55 (0.41)	0.19	-0.19 (0.27)	0.46	-0.12 (0.27)	0.66
Use of tooth paste	0.23 (1.71)	0.85	-0.44 (0.61)	0.47	-0.55 (0.49)	0.26	-0.21 (0.45)	0.65
Use of bottled water	0.67 (1.24)	0.59	-0.06 (0.69)	0.94	-0.07 (0.54)	0.89	0.44 (0.49)	0.38
Use of city water	0.67 (1.12)	0.56	-0.04 (0.63)	0.96	-0.54 (0.49)	0.27	0.18 (0.46)	0.71
Use of well water	***		***		***		***	

Table 67-Continued

Ever use of vitamin drops or tablets with fluoride	***		***		***		***	
Presence of regular dentist	***		***		***		***	
Reason for last visit to dentist	***		***		***		***	

* dmfs at the cavitated level.

* Negative binomial model was used in the analyses.

** The iteration process did not converge, so could not be determined.

Table 68. Bivariate analyses of associations of baseline family-related factors with ECC incidence rates* (count) at the three one-year periods and the three-year period**

Baseline factor	Incidence period							
	Baseline to 1st follow-up		1st follow-up to 2nd follow-up		2nd follow-up to 3rd follow-up		Baseline to 3rd follow-up	
	Linear coefficient estimate (SE)	P-value	Linear coefficient estimate (SE)	P-value	Linear coefficient estimate (SE)	P-value	Linear coefficient estimate (SE)	P-value
Number of household members	0.03 (0.24)	0.92	-0.06 (0.13)	0.64	0.13 (0.14)	0.36	0.07 (0.11)	0.52
Mother's DMFS***	0.08 (0.05)	**0.10**	0.03 (0.02)	0.19	0.01 (0.02)	0.44	0.02 (0.02)	**0.13**

* dmfs at the cavitated level.

** Negative binomial model was used in the analyses.

*** Dental caries was assessed at the cavitated-level only.

Table 69 shows that there were no statistically significant relationships between the frequencies of consumption of milk, water and infant formula and ECC incidence (count) from baseline to 1st follow-up, 1st follow-up to 2nd follow-up, 2nd follow-up to 3rd follow-up or baseline to 3rd follow-up. Increased frequency of 100% juice consumption was associated with higher incidence of ECC (count) from the 2nd follow-up to 3rd follow-up (linear coefficient estimate=0.29), with P-value of 0.09. Increased frequency of consumption of sugar-added beverages was associated with higher incidence of ECC (count) from the 2nd follow-up to 3rd follow-up (linear coefficient estimate=0.29), with P-value of 0.11.

Table 70 shows that there were no statistically significant relationships between the amounts of consumption of milk, water and infant formula and ECC incidence (count) from baseline to 1st follow-up, 1st follow-up to 2nd follow-up, 2nd follow-up to 3rd follow-up or baseline to 3rd follow-up. Increased frequency of 100% juice consumption was associated with higher incidence of ECC (count) from the 2nd follow-up to 3rd follow-up (linear coefficient estimate=0.29), with P-value of 0.11. Increased frequency of consumption of sugar-added beverages was associated with higher incidence of ECC (count) from the 2nd follow-up to 3rd follow-up (linear coefficient estimate=0.29), with P-value of 0.11.

Natural Juice (100% juice) included all juices without added sugar, such as tomato juice, orange juice, apple juice, etc. Other juices (sugar-added beverages) included all juice drinks with added sugar, such as Coke™ and other carbonated beverages, lemonade, Hi-C™, Hawaiian Punch™, Caprisun™, Koolaid™, Gatorade™, Vitamin Water™, and Red Bull™.

Table 69. Bivariate analyses of associations of baseline frequency of beverages consumption (times/day) with ECC incidence rates* (count) for the three one-year periods and the three-year period**

Baseline frequency of intake of beverage type	Incidence period							
	Baseline to 1st follow-up		1st follow-up to 2nd follow-up		2nd follow-up to 3rd follow-up		Baseline to 3rd follow-up	
	Linear coefficient estimate (SE)	P-value	Linear coefficient estimate (SE)	P-value	Linear coefficient estimate (SE)	P-value	Linear coefficient estimate (SE)	P-value
Milk	0.04 (0.37)	0.91	-0.19 (0.19)	0.32	0.06 (0.15)	0.71	-0.03 (0.14)	0.84
Water	0.29 (0.68)	0.68	-0.08 (0.21)	0.73	-0.21 (0.16)	0.19	-0.07 (0.16)	0.68
100% juice	-0.05 (0.45)	0.93	0.01 (0.28)	0.99	0.29 (0.18)	**0.09**	0.12 (0.19)	0.54
Sugar-added beverages	-0.05 (0.44)	0.91	-0.01 (0.28)	0.99	0.29 (0.18)	**0.11**	0.11 (0.18)	0.57
Infant formula	-0.21 (0.29)	0.48	-0.03 (0.14)	0.86	-0.07 (0.09)	0.49	-0.04 (0.09)	0.69

* dmfs at the cavitated level.

** Negative binomial model was used in the analyses.

Table 70. Bivariate analyses of associations of baseline amount of beverages consumption (cups/day) with ECC incidence rates* (count) for the three one-year periods and the three-year period**

Baseline quantity of intake beverage type	Incidence period							
	Baseline to 1st follow-up		1st follow-up to 2nd follow-up		2nd follow-up to 3rd follow-up		Baseline to 3rd follow-up	
	Linear coefficient estimate (SE)	P-value	Linear coefficient estimate (SE)	P-value	Linear coefficient estimate (SE)	P-value	Linear coefficient estimate (SE)	P-value
Milk	0.04 (0.37)	0.91	-0.87 (0.19)	0.32	0.06 (0.15)	0.71	0.06 (0.15)	0.71
Water	0.23 (0.66)	0.73	-0.08 (0.21)	0.67	-0.22 (0.16)	0.18	-0.08 (0.16)	0.61
100% juice	-0.04 (0.44)	0.92	0.01 (0.28)	0.99	0.29 (0.19)	**0.11**	0.11 (0.19)	0.55
Sugar-added beverages	-0.02 (0.45)	0.97	0.04 (0.29)	0.89	0.29 (0.18)	**0.11**	0.12 (0.18)	0.52
Infant formula	-0.19 (0.29)	0.51	-0.01 (0.14)	0.94	-0.06 (0.09)	0.57	-0.03 (0.09)	0.78

* dmfs at the cavitated level.

** Negative binomial model was used in the analyses.

Bivariate Analyses—Repeated Questionnaires

Some of the risk factors were assessed not only at baseline, but also at later time points. These include the frequencies and the quantities of consumption of different beverages, frequency of toothbrushing, use of toothpaste and presence of a regular dentist. These variables were also assessed at visits 10, 40, 50, 60, 70 and 90, when the mean ages of the children were 0.9, 1.5, 1.9, 2.5, 3.1 and 4.1 years, respectively. Since only 23 children provided information at visit 90 (3rd follow-up examination), information at that visit will not be included in the analyses. Some new questions were added to the follow-up questionnaires, such as those asking about eating sweets (yes/no), the daily frequency of eating sweets (times per day, times per week and times per month), eating sweetened food (yes/no), the daily frequency of eating sweetened food (times per day, times per week and times per month), adding sugar to foods or drinks (yes/no), and the daily amount (teaspoons per day). All the frequencies were converted to times per day (times per week/7 and times per month/30).

Table 71 shows the results of bivariate logistic regression analyses relating the three-year incidence of ECC (dichotomous) to both the concurrent area-under-the-curve (AUC) estimates (using visits 40, 50, 60 and 70) for the variables that were assessed on the follow-up questionnaires and the cross-sectional report of these variables at the 2nd follow-up only (visit 70). There were no statistically significant relationships (at the level of α=0.15) between the three-year incidence and the frequencies of all of the beverages, except for 100% juice (OR=0.54 and P-value=0.13) for the concurrent AUC. Children who consumed sweetened food one more time per day in the concurrent AUC had about seven times the odds, with statistically significant results (P-value=0.003).

At the 2nd follow-up, all subjects were using toothpaste and all were consuming sweetened foods. Since there was no variability in either of these measures, logistic regression equation coefficients were not estimable. For AUC consumption of sweetened foods, all children but three reported consuming sweetened foods in visits 40, 50, 60 and 70. Those three children reported not consuming sweetened foods initially (visit 40, age<two years), but did consume sweetened foods thereafter. This made the variability in AUC sweetened foods quite low (although not zero). The variability was, however, low enough that logistic regression resulted in estimates with extremely high standard errors. Since those estimates were based on all but three subjects with consistent high exposure to sweetened foods, they have not been presented in the table.

Table 72 shows the results of negative binomial regression analyses relating the three-year incidence rate of ECC (count) to both the concurrent area-under-the-curve estimates (AUC) (using visits 40, 50, 60 and 70) for the variables that were assessed on the follow-up questionnaires and the cross-sectional report of these variables at the 2nd follow-up only (visit 70). Children who had a previous visit to a dentist for the concurrent AUC and at 2nd follow-up alone had more new dfs during the three-year period than those who had no history of previous visit to a dentist, with P-values of 0.02 and 0.15, respectively.

At the 2nd follow-up, all subjects were using toothpaste and all were consuming sweetened foods. Since there was no variability in either of these measures, negative binomial regression equations with these variables were not estimable. For AUC consumption of sweetened foods, all children but three reportedly were consuming sweetened foods at visits 40, 50, 60 and 70 consuming sweetened foods. Those three children reported not consuming sweetened foods initially (visit 40, age<two years), but did consume sweetened foods thereafter. This made the variability in AUC sweetened foods quite low (although not zero). The variability was, however, low enough that negative binomial regression equations resulted in estimates with extremely high standard errors, so AUC consumption of sweetened foods was excluded.

Table 71. Bivariate logistic regression of the incidence of ECC (dichotomous) from baseline to the 3rd follow-up examinations and behavioral risk factors defined using concurrent AUC* and 2nd follow-up only

Variables**	AUC*		2nd follow-up only	
	OR (95% CI)	P-value	OR (95% CI)	P-value
Daily frequency of consumption of milk	1.18 (0.74-1.89)	0.49	1.07 (0.74-1.52)	0.77
Daily frequency of consumption of 100% juice	0.54 (0.24-1.19)	**0.13**	0.92 (0.65-1.30)	0.63
Daily frequency of consumption of water	1.23 (0.71-2.10)	0.46	1.07 (0.71-1.64)	0.72
Daily frequency of consumption of sugar-added beverages	1.28 (0.74-2.21)	0.38	1.28 (0.83-1.98)	0.26
Daily frequency of brushing	0.68 (0.16-2.94)	0.57	0.61 (0.06-6.19)	0.68
Use of toothpaste	0.37 (0.01-15.84)	0.60	NA***	NA***
Self-brushes	0.57 (0.15-2.23)	0.60	0.35 (0.07-1.76)	0.20
Regular dentist (current)	1.85 (0.31-10.99)	0.50	1.67 (0.59-4.69)	0.33
Seen dentist-ever	1.86 (0.42-8.21)	0.41	1.56 (0.51-4.78)	0.44
Consumption of sweets (yes/no)	1.09 (0.18-6.77)	0.92	1.29 (0.46-3.62)	0.63
Daily frequency of consumption of sweets	1.96 (0.26-15.15)	0.52	1.71 (0.65-4.52)	0.28
Consumption of sweetened foods (yes/no)	NA***	NA***	NA***	NA***
Daily frequency of consumption of sweetened foods	6.67 (1.89-23.53)	**0.003**	1.76 (0.70-4.38)	0.23

* AUC includes visits 40, 50, 60 and 70, when mean ages of children were 1.5, 2.0, 2.5 and 3.1 years, respectively.

** The variables use of dietary fluoride supplements and adding sugar to foods and beverages were excluded, because none of the children's caregivers reported that they had them.

*** At the 2nd follow-up and AUC, all subjects were using toothpaste and all were consuming sweetened foods. Since there was no variability in either of these measures, logistic regression coefficients were not estimable.

Table 72. Bivariate negative binomial regression analyses of the incidence of ECC (count) from baseline to the 3rd follow-up examination and behavioral risk factors defined using concurrent AUC* and the 2nd follow-up.

Variables	AUC		2nd follow-up only	
	Estimate (SE)	P-value	Estimate (SE)	P-value
Daily frequency of consumption of milk	-0.41 (0.19)	0.48	-0.08 (0.16)	0.66
Daily frequency of consumption of 100% juice	-0.14 (0.19)	0.34	0.09 (0.14)	0.52
Daily frequency of consumption of water	-0.01 (0.23)	0.68	-0.04 (0.16)	0.80
Daily frequency of consumption of sugar-added beverages	0.14 (0.24)	0.56	-0.12 (0.17)	0.48
Daily frequency of brushing	-0.73 (0.68)	0.29	-1.03 (0.92)	0.26
Use of toothpaste	0.37 (1.59)	0.82	NA*	NA*
Self-brushes	-0.36 (0.58)	0.53	-0.28 (0.57)	0.63
Regular dentist	1.02 (0.75)	0.17	0.42 (0.45)	0.94
Seen dentist-ever	1.58 (0.66)	**0.02**	0.69 (0.49)	**0.15**
Daily consumption of sweets (yes/no)	0.78 (0.96)	0.42	0.30 (0.45)	0.50
Frequency of consumption of sweets	0.33 (1.01)	0.74	0.09 (0.46)	0.84
Daily consumption of sweetened foods	6.47 (6.33)**	0.31	NA*	NA*
Dairy frequency of consumption of sweetened foods	0.02 (0.56)	0.26	0.39 (0.47)	0.40

* At the 2nd follow-up, all subjects were using toothpaste and all were consuming sweetened foods. Since there was no variability in either of these measures, negative binomial regression equations with these variables were not estimable.

** All children but three reportedly were consuming sweetened foods at visits 40, 50, 60 and 70. This made the variability in AUC sweetened foods very low, so negative binomial regression equations resulted in estimates with extremely high standard errors, so consumption of sweetened food was excluded.

Tables 73 and 74 show the results of bivariate logistic regression analyses relating the incidence of ECC from 1st follow-up to 2nd follow-up (dichotomous and count, respectively) to both the concurrent area-under-the-curve (AUC) estimates for the variables that were assessed on the follow-up questionnaires and the cross-sectional report of these variables at the 2nd follow-up only (visit 70). Table 73 shows that there were no statistically significant relationships (at the level of α=0.15) between the incidence from 1st follow-up to 2nd follow-up and the frequencies of any of the beverages for either the concurrent AUC or at 2nd follow-up alone.

At the 2nd follow-up, all subjects were using toothpaste and all were consuming sweetened foods. Thus, logistic regression equations were inestimable. For AUC consumption of sweetened foods, all children but three reported consuming sweetened foods in visits 40, 50, 60 and 70. Those three children reported not consuming sweetened foods initially (visit 40, age<two years), but did consume sweetened foods thereafter. This made the variability in AUC sweetened foods quite low (although not zero). The variability was, however, low enough that logistic regression equations resulted in estimates with extremely high standard errors.

Children who reportedly had a regular dentist at AUC had more than four times the odds of incidence of ECC as those who did not have a regular dentist, with P-value of 0.07 (OR=4.29). Also, children who had an earlier history of a previous visit to a dentist at AUC had more than four times the odds of incidence of ECC as those who had no previous history of a dental visit, with P-value of 0.05 (OR=4.48). Children who reportedly had a regular dentist and had a history of a previous visit to a dentist at the 2nd follow-up only had more than two times the odds of incidence of ECC as those who did not have a regular dentist and had no previous history of a dental visit, with P-values of 0.12 and 0.11, respectively

Table 74 shows that at the 2nd follow-up, all subjects were using toothpaste and all were consuming sweetened foods. Since there was no variability in either of these measures, negative binomial regression equations with these variables were not estimable. For AUC consumption of sweetened foods, all children but three reported consuming sweetened foods at visits 40, 50, 60 and 70. Those three children reported not consuming sweetened foods initially (visit 40, age<two years), but did consume sweetened foods thereafter. This made the variability in AUC sweetened foods quite low (although not zero). The variability was, however, low enough that negative binomial regression equations resulted in estimates with extremely high SE (not shown)

Children who reportedly had a regular dentist or had a history of a previous visit to a dentist had more new dfs than those who of did not have a regular dentist or had no previous history of a dental visit, with P-values of 0.02 and 0.03, respectively. Also, children who reportedly had a regular dentist and had a history of a previous visit to a dentist at the 2nd follow-up had more carious surfaces than those who did not have a regular dentist and had no previous history of a dental visit, with P-values of 0.04 and 0.12, respectively. Surprisingly, children who brushed their teeth had fewer new carious lesions than those who did not brush their own teeth, with P-value of 0.15.

Children who reportedly had a regular dentist or had a history of a previous visit to a dentist had more new dfs than those who of did not have a regular dentist or had no previous history of a dental visit, with P-values of 0.02 and 0.03, respectively. Also, children who reportedly had a regular dentist and had a history of a previous visit to a dentist at the 2nd follow-up had more carious surfaces than those who did not have a regular dentist and had no previous history of a dental visit, with P-values of 0.04 and 0.12, respectively. Surprisingly, children who brushed their teeth had fewer new carious lesions than those who did not brush their own teeth, with P-value of 0.15.

Table 73. Bivariate logistic regression of the incidence of ECC (dichotomous) from 1st follow-up to the 2nd follow-up examination and behavioral risk factors defined using concurrent AUC and 2nd follow-up only.

Variables	AUC		2nd follow-up only	
	OR (95% CI)	P-value	OR (95% CI)	P-value
Daily frequency of consumption of milk	1.17 (0.74-1.86)	0.50	1.02 (0.72-1.45)	0.90
Daily frequency of consumption of 100% juice	1.02 (0.49-2.1)	0.96	1.11 (0.79-1.55)	0.53
Daily frequency of consumption of water	0.99 (0.59-1.65)	0.96	0.91 (0.60-1.36)	0.64
Daily frequency of consumption of sugar-added beverages	1.18 (0.72-1.95)	0.51	0.98 (0.67-1.42)	0.90
Daily frequency of brushing	0.53 (0.14-1.94)	0.33	0.99 (0.16-6.29)	0.99
Use of toothpaste	0.26 (0.01-6.74)	0.42	NA*	NA*
Self-brushes	0.53 (0.15-1.86)	0.32	0.45 (0.15-1.36)	0.16
Regular dentist	4.29 (0.91-20.35)	**0.07**	2.16 (0.82-5.63)	**0.12**
Seen dentist-ever	4.48 (1.01-19.94)	**0.049**	2.42 (0.82-7.19)	**0.11**
Consumption of sweets (yes/no)	0.62 (0.10-3.6)	0.59	0.93 (0.36-2.41)	0.89
Daily frequency of consumption of sweets	1.58 (0.27-9.29)	0.61	1.46 (0.67-3.17)	0.34
Consumption of sweetened foods (yes/no)	NA**	NA*	NA*	NA*
Daily frequency of consumption of sweetened foods	1.56 (0.51-4.41)	0.46	1.23 (0.50-2.99	0.65

* At the 2nd follow-up, all subjects were using toothpaste and all were consuming sweetened foods. Since there was no variability in either of these measures, logistic regression equations with these variables were not estimable.

** All children but three reportedly were consuming sweetened foods at visits 40, 50, 60 and 70. This made the variability in AUC sweetened foods very low, so logistic regression equations resulted in estimates with extremely high standard errors, so consumption of sweetened food was excluded.

Table 74. Negative binomial regression analyses of the incidence of ECC (count) from 1st follow-up to the 2nd follow-up examination and behavioral risk factors at the concurrent AUC and the 2nd follow-up.

Variables	AUC		2nd follow-up only	
	Estimate (SE)	P-value	Estimate (SE)	P-value
Daily frequency of consumption of milk	-0.25 (0.28)	0.38	0.00 (0.28)	0.99
Daily frequency of consumption of 100% juice	0.03 (0.49)	0.95	0.25 (0.19)	0.20
Daily frequency of consumption of water	-0.03 (0.29)	0.91	0.16 (0.27)	0.55
Daily frequency of consumption of sugar-added beverages	0.066 (0.31)	0.83	-0.16 (0.23)	0.50
Daily frequency of brushing	-0.35 (0.92)	0.71	-0.63 (1.19)	0.60
Use of toothpaste	1.34 (2.33)	0.57	NA*	NA*
Self-brushes	-0.86 (0.89)	0.34	-1.02 (0.70)	**0.15**
Regular dentist	2.17 (0.96)	**0.02**	1.29 (0.61)	**0.04**
Seen dentist-ever	2.09 (0.96)	**0.03**	1.04 (0.67)	**0.12**
Consumption of sweets (yes/no)	0.63 (1.42)	0.66	-0.09 (0.63)	0.89
Frequency of consumption of sweets	0.33 (1.01)	0.74	-0.23 (0.62)	0.71
Consumption of sweet foods	NA**	NA**	NA*	NA*
Frequency of consumption of sweet foods	0.87 (0.84)	0.30	0.89 (0.69)	0.20

* At the 2nd follow-up, all subjects were using toothpaste and all were consuming sweetened foods. Since there was no variability in either of these measures, negative binomial regression equations were not estimable.

** All children but three reportedly were consuming sweetened foods at visits 40, 50, 60 and 70. This made the variability in AUC sweetened foods very low, so logistic regression equations resulted in estimates with extremely high standard errors, so consumption of sweetened food was excluded.

Multivariable Analyses

Introduction

In this section, the results of multivariable analyses will be summarized. Multivariable models were developed based on assessing results of the bivariate analyses. Variables that met the screening cut-off of 0.15 in the bivariate analyses were included in the multivariable models. Tens of multivariable analyses models could have been built with different combinations of dependent and independent variables. However, since the prevalence of ECC at baseline was very low (1.1%), the incidence from baseline to 1st follow-up, baseline to 2nd follow-up and baseline to 3rd follow-up were almost the same as the prevalence at 1st follow-up, 2nd follow-up and 3rd follow-up, respectively. Thus, prevalence of ECC at baseline and the three follow-up examinations were excluded as dependent variables in order to prevent redundancy.

For the incidence of ECC as a dependent variable, we had twelve major possibilities (the six incidence periods with dichotomous outcomes and with count outcomes, but two incidence periods and four dependent variables were assessed: incidence from baseline to the 3rd follow-up (dichotomous and count) and incidence from 1st follow-up to the 2nd follow-up (dichotomous and count). The rationale behind choosing these two incidence periods was that, for the incidence from baseline to the 3rd follow-up (n=73), the multivariable model would cover the whole three-year period of the study, and for the incidence from 1st follow-up to 2nd follow-up, the sample size is relatively larger (n=83), with high incidence of ECC, so it provided greater power to detect differences in incidence of ECC among children with different dietary and oral hygiene practices.

Multivariable Models

As mentioned before, four multivariable models were developed with incidence from baseline to third follow-up (dichotomous and count) and 1st follow-up to 2nd follow-up (dichotomous and count) as dependent variables. Each of the models was developed both without adjustment for age (A) and after adjustment for age (B). The significance level of 0.10 was considered statistically significant for the model selection, due to the limited sample size. Models with significance level of 0.05 also were explored.

Model 1-A Three-year Incidence from Baseline to 3rd Follow-up (Dichotomous)-not Adjusted for Age

In the bivariate analyses conducted in the previous section using three-year incidence of ECC from baseline to 3rd follow-up (dichotomous) as a dependent variable, premature delivery (baseline), AUC of daily frequency consumption of 100% juice (visits 40, 50, 60 and 70) and AUC of consumption of sweetened foods (yes/no) met the inclusion criterion of having bivariate logistic regression P-values of 0.15 or less (Table 75). A multivariable model was built including all of these factors as independent variables (Table 76). All the variables in this model were statistically significantly related to the three-year incidence of ECC (dichotomous) at $\alpha=0.10$. Thus, no additional model-trimming was conducted. Table 76 shows that children who consumed sweetened food (AUC) one more time per day had approximately 9.2 times the odds of developing caries for the three-year incidence period as those who consumed sweetened food (AUC) less frequently per day (P-value=0.002). Also, children who were delivered prematurely and consumed 100% juice (AUC) one more time per day had approximately 80% and 60% lower odds of developing dental caries for the three-year incidence of ECC, compared to those who were delivered full-term and consumed 100% juice (AUC) less frequently per day, respectively (both p-values=0.049). Two-way interactions were not significant between different combinations of the variables (all P-values>0.10). When assessing the interaction between AUC of daily frequency of consumption of sweetened food and AUC of daily frequency of consumption of 100% juice, the latter was changed into a dichotomous variable in order to assess whether the effect of AUC of daily frequency of consumption of sweetened food on ECC incidence was different in different levels of AUC of daily frequency of consumption of 100% juice. It is important to notice that all the variables in Model 1-A were statistically significantly associated with the three-year incidence rate at α=0.05 as well.

Table 75. Bivariate logistic regression for three-year incidence of ECC from baseline to 3rd follow-up for variables with P-values of 0.15 or less

Variables	OR (95%CI)	P-value
AUC* of daily frequency of consumption of sweetened food	6.67 (1.89-23.53)	0.003
Premature delivery (baseline)	0.29 (0.08-1.14)	0.08
AUC* of daily frequency of consumption of 100% juice	0.54 (0.24-1.19)	0.13

* AUC includes visits 40, 50, 60 and 70, when mean ages of children were 1.5, 2.0, 2.5 and 3.1 years, respectively.

Table 76. Multivariable logistic regression for three-year incidence of ECC from baseline to 3rd follow-up (full and final model, n=67)

Variables*	OR (95%CI)	P-value
AUC** of daily frequency of consumption of sweetened food	9.22 (2.32-36.67)	0.002
Premature delivery (baseline)	0.21 (0.04-0.99)	0.049
AUC** of daily frequency of consumption of 100% juice	0.37 (0.13-0.99)	0.049

* No two-way interactions were significant between pairs of variables (all P-value>0.10).

** AUC includes visits 40, 50, 60 and 70, when mean ages of children were 1.5, 2.0, 2.5 and 3.1 years, respectively.

Model 1-B: Three-year Incidence from Baseline to 3rd Follow-up (Dichotomous) -adjusted for Age

In the bivariate analyses conducted in the previous section using three-year incidence of ECC from baseline to 3rd follow-up (dichotomous) as a dependent variable, premature delivery (baseline), AUC of daily frequency consumption of 100% juice (visits 40, 50, 60 and 70) and AUC of consumption of sweetened foods (yes/no) met the inclusion criterion of having logistic regression P-values of 0.15 or less (Table 75). A multivariable model was built including all of these factors as independent variables with age forced into the model (Table 77).

All the variables in this model except age were statistically significantly related to the three-year incidence of ECC (dichotomous) at $\alpha=0.10$. Thus, no additional model-trimming was conducted. Table 77 shows that children who consumed sweetened food (AUC) one more time per day had approximately 9.7 times the odds of developing caries for the three-year incidence period as those who consumed sweetened food (AUC) less frequently per day (P-value=0.002). Also, children who were delivered prematurely and consumed 100% juice (AUC) one more time per day had approximately 80% and 60% lower odds of developing dental caries for the three-year incidence of ECC, compared to those who were delivered full-tern and consumed 100% juice (AUC) less frequently per day, respectively (p-values=0.049 and 0.059, respectively). Two way interactions were not significant between different combinations of the variables (all P-value>0.1).

Table 77. Multivariable logistic regression for three-year incidence of ECC from baseline to 3rd follow-up (full model, n=67)

Variables*	OR (95%CI)	P-value
Age	0.39 (0.01-2.15)	0.28
AUC** of daily frequency of consumption of sweetened food	9.66 (2.35-39.74)	0.002
Premature delivery (baseline)	0.21 (0.04-0.99)	0.048
AUC** of daily frequency of consumption of 100% juice	0.37 (0.14-1.04)	0.059

* No two-way interactions were significant between pairs of variables (all P-value>0.1).

** AUC includes visits 40, 50, 60 and 70, when mean ages of children were 1.5, 2.0, 2.5 and 3.1 years, respectively.

When developing additional models exploring 0.05 as the significance level, a step-back, manual elimination was used and the consumption of 100% juice (AUC for visits 40 to 70) was eliminated first due to having the highest P-value of 0.059 (Table 78). Then, the variable premature delivery was eliminated for having p-value of 0.10 (Table 79). Table 5 shows that at the significance level of 0.05, children who consumed sweetened foods (AUC) one more time per day had approximately seven times the odds of developing caries for the three-year incidence period as compared to those who consumed sweetened food (AUC) less frequently per day, after adjustment for age (P-value=0.003).

Table 78. Multivariable logistic regression for three-year incidence of ECC from baseline to 3rd follow-up (first reduced model, n=67)

Variables*	OR (95%CI)	P-value
Age	0.36 (0.07-1.82)	0.22
AUC** of daily frequency of consumption of sweetened food	7.32 (1.93-27.69)	0.003
Premature delivery (baseline)	0.28 (0.06-1.25)	0.10

* There was no significant two-way interaction (P-value>0.05).

** AUC includes visits 40, 50, 60 and 70, when mean ages of children were 1.5, 2.0, 2.5 and 3.1 years, respectively.

Table 79. Multivariable logistic regression for three-year incidence of ECC from baseline to 3rd follow-up (final reduced model, n=67)

Variables*	OR (95%CI)	P-value
Age	0.38 (0.08-1.84)	0.23
AUC** of daily frequency of consumption of sweetened food	7.17 (1.93-26.59)	0.003

* There was no significant two-way interaction (P-value>0.05).

** AUC includes visits 40, 50, 60 and 70, when mean ages of children were 1.5, 2.0, 2.5 and 3.1 years, respectively.

Comparison Between Model 1-A and Model 1-B

Model 1-A showed that daily frequency of consumption of sweetened food (AUC), premature delivery and daily frequency of consumption of 100% juice (AUC) were statistically significantly associated with the three-year incidence of ECC, with P-values of 0.002, 0.049 and 0.049, respectively. After adjustment for age (Model 1-B), similar results were obtained with p-values of 0.002, 0.048 and 0.059, respectively, when using a P-value of 0.10.

However, when using P-value of 0.05, Model 1-B showed that only daily frequency of consumption of sweetened food was statistically significantly associated with the three-year incidence of ECC (P-value=0.003). Thus, if we selected the statistical level of alpha=0.10, we would end up by having results in Model 1-B similar to that in Model 1-A by selecting the model in Table 77 as the final model.

Model 2-A: Three-year Incidence from Baseline to 3rd Follow-up (dmfs Count)-not Adjusted for Age

In the bivariate analyses conducted in the previous section using three-year incidence from baseline to 3rd follow-up (count) as a dependent variable, mother's DMFS (baseline) and presence of history of previous dental visit at the 2nd follow-up (yes/no) met the inclusion criterion of having P-values of 0.15 or less (Table 80). History of previous dental visit (AUC visits 40 to 70) was not included in the analyses, because it was statistically significantly correlated with the presence of previous dental visit at the 2nd follow-up (P-value=0.001). Another reason was that, after running detailed descriptive analyses, we noticed that there were 19 children who reportedly had no history of a visit to a dentist (ever) in the 2nd follow-up data, but had AUC more than zero, and eight of them had AUC>=0.5. Also, only five children who reportedly had no previous visit to a dentist at the 2nd follow-up had AUC of a previous visit to a dentist of zero (this issue will be addressed in detail in the discussion section).

A multivariable model was built including mother's DMFS (baseline) and presence of history of previous dental visit at the 2nd follow-up (yes/no) as independent variables (Table 81). A step-back, manual elimination algorithm was used and mother's DMFS was eliminated first due to having the highest P-value (0.41). Table 82 shows the next step in model trimming with a model that had the presence of history of previous dental visit at 2nd follow-up. At both α=0.10 and α=0.05, the presence of history of previous dental visit at 2nd follow-up was not statistically significantly associated with ECC incidence from baseline to the 3rd follow-up (P-value=0.15). In other words, none of the variables met the alpha level of 0.1.

Table 80. Bivariate negative binomial regression for three-year incidence of ECC for variables with P-values of 0.15 or less

Variables	Estimate (SD)	Incidence rate ratio*	P-value
Mother's DMFS (baseline)	0.02 (0.02)	1.02	0.13
Presence of a history of previous visit to a dentist (2nd follow-up)	0.69 (0.49)	1.99	0.15

* Incidence Rate Ratio (IRR) obtained from the exponentiation of the estimates.

Table 81. Multivariable negative binomial regression for three-year incidence of ECC (full model, n=50)

Variables	Estimate (SD)	Incidence rate ratio*	P-value
Mother's DMFS (baseline)	0.01 (0.02)	1.01	0.41
Presence of a history of previous visit to a dentist (2nd follow-up)	0.54 (0.50)	1.71	0.28

* IRR obtained from the exponentiation of the estimates.

Table 82. Multivariable negative binomial regression for three-year incidence of ECC (final reduced model, n=60)

Variables	Estimate (SD)	IRR	P-value
Presence of a history of previous visit to a dentist (2nd follow-up)	0.70 (0.49)	2.01	0.15

Model 2-B: Three-year Incidence from Baseline to 3rd Follow-up (dmfs Count)-adjusted for Age

In the bivariate analyses conducted in the previous section using three-year incidence from baseline to 3rd follow-up (count) as a dependent variable, mother's DMFS (baseline) and presence of history of previous dental visit at the 2nd follow-up (yes/no) met the inclusion criterion of having P-values of 0.15 or less (Table 80). A multivariable model was built including both of these factors as independent variables with age which was forced into the model (Table 83). History of previous dental visit (AUC visits 40 to 70) was not included in the analyses, because it was statistically significantly correlated with the presence of previous dental visit at the 2nd follow-up (P-value=0.001). A step-back, manual elimination algorithm was used and mother's DMFS was eliminated first due to having the highest P-value (0.39). Table 84 shows the next step in model trimming with a model that had the presence of history of previous dental visit at 2nd follow-up and age. Table 84 shows that, after adjustment for age, having a history of a previous visit to a dentist at the 2nd follow-up was not statistically significantly associated with the three-year incidence rate (P-value=0.16).

Table 83. Multivariable negative binomial regression for three-year incidence of ECC (full model, n=50)

Variables	Estimate (SD)	Incidence rate ratio*	P-value
Age	0.51 (0.81)	1.66	0.53
Mother's DMFS (baseline)	0.01 (0.02)	1.01	0.39
Presence of a history of previous visit to a dentist (2nd follow-up)	0.49 (0.49)	1.64	0.32

* IRR obtained from the exponentiation of the estimates.

Table 84. Multivariable negative binomial regression for three-year incidence of ECC (final reduced model, n=60)

Variables	Estimate (SD)	IRR*	P-value
Age	0.27 (0.79)	1.32	0.73
Presence of a history of previous visit to a dentist (2nd follow-up)	0.68 (0.49)	1.98	0.16

* IRR obtained from the exponentiation of the estimates.

Comparison Between Model 2-A and Model 2-B

Both Model 2-A and Model 2-B showed that the presence of a history of a previous visit to a dentist the 2nd follow-up was not statistically significantly associated with the three-year incidence of ECC (before and after adjustment for age, respectively) at either $\alpha=0.10$ or $\alpha=0.05$. Thus, there were no important differences between the two models regarding the mentioned variables.

Model 3-A: Incidence from 1st follow-up to 2nd Follow-up (Dichotomous)-not Adjusted for Age

In the bivariate analyses conducted in the previous section using incidence of ECC from 1st follow-up to 2nd follow-up (dichotomous) as a dependent variable, low birth weight, toothbrushing (yes/no, baseline), daily frequency of toothbrushing (baseline), presence of a regular dentist (2nd Follow-up) and presence of a history of previous visit to a dentist (2nd follow-up) met the inclusion criterion of having logistic regression P-values of 0.15 or less (Table 85). History of previous dental visit (AUC visits 40 to 70) and presence of a regular dentist (AUC visits 40 to 70) were not included in the analyses, because they were statistically significantly correlated with the presence of previous dental visit at the 2nd follow-up (P-value=0.001) and the presence of a regular dentist at the 2nd follow-up (P-value<0.0001), respectively. Also, since toothbrushing (yes/no) and the frequency of toothbrushing at baseline were collinear (P-value<0.001), toothbrushing (yes/no) was manually eliminated from this model because the variable frequency of toothbrushing provided more information (none/once per day/twice per day, etc.) than toothbrushing (yes/no).

A multivariable model was built including all of the mentioned factors as independent variables (Table 86). A step-back, manual elimination algorithm was used and the presence of a regular dentist at the 2nd follow-up was eliminated first because of having the highest p-value of 0.99 (Table 87). This was followed by eliminating the variable low birth weight with P-value of 0.47, resulting in a final reduced model that had daily frequency of toothbrushing (baseline) and the presence of a history of a previous visit to a dentist at the 2nd follow-up (Table 88). Table 88 shows that increased daily frequency of toothbrushing at baseline was statistically significantly associated with lower incidence of ECC from 1st to 2nd follow-up (OR=0.34, P-value=0.01), while having a history of previous visits to a dentist at the 2nd follow-up was associated with higher incidence of ECC from 1st follow-up to 2nd follow-up (OR=4.57, P-value 0.03). No significant two-way interaction was detected between the two variables in the final model (daily frequency of toothbrushing at baseline and presence of a history of a previous visit to a dentist at the 2nd follow-up) at the significance level of 0.10. When exploring 0.05 as the significance level, no differences in the results of the multivariable analyses and interaction between daily frequency of toothbrushing at baseline and presence of a history of a previous visit to a dentist at the 2nd follow-up were detected, since all the P-values in the final reduced model were less than 0.05.

Table 85. Bivariate logistic regression for incidence of ECC from 1st follow-up to 2nd follow-up for the variable that had P-values of 0.15 or less

Variables	OR (95%CI)	P-value
Low birth weight (baseline)	0.38 (0.11-1.28)	0.12
Toothbrushing (yes/no, baseline)	0.42 (0.16-1.11)	0.08
Daily frequency of toothbrushing	0.59 (0.33-1.04)	0.07
Presence of a history of a regular dentist (2nd follow-up)	2.14 (0.82-5.63)	0.12
Presence of a history of previous visit to a dentist (2nd follow-up)	2.42 (0.82-5.63)	0.11

Table 86. Multivariable logistic regression for incidence of ECC from 1st follow-up to 2nd follow-up (full model, n=57)

Variables*	OR (95%CI)	P-value
Low birth weight** (baseline)	0.55 (0.11-2.67)	0.46
Daily frequency of toothbrushing (baseline)	0.35 (0.16-0.77)	0.01
Presence of a history of a regular dentist (2nd follow-up)	1.00 (0.16-6.24)	0.99
Presence of a history of previous visit to a dentist (2nd follow-up)	4.26 (0.58-31.54)	0.16

* Toothbrushing (yes/no) was eliminated due to collinearity with daily frequency of toothbrushing (Pearson correlation=0.93, P-value<0.001).

** Children who were less than 5 pounds and 8 ounces were reported as low birth weight children

Table 87. Multivariable logistic regression for incidence of ECC from 1st follow-up to 2nd follow-up (1st reduced model, n=58)

Variables	OR (95%CI)	P-value
Low birth weight (baseline)	0.56 (0.12-2.70)	0.47
Daily frequency of toothbrushing (baseline)	0.34 (0.15-0.76)	0.01
Presence of a history of previous visit to a dentist (2nd follow-up)	4.50 (1.13-17.97)	0.03

Table 88. Multivariable logistic regression for incidence of ECC from 1st follow-up to 2nd follow-up (2nd reduced model, n=58)

Variables*	OR (95%CI)	P-value
Daily frequency of toothbrushing (baseline)	0.34 (0.15-0.75)	0.01
Presence of a history of previous visit to a dentist (2nd follow-up)	4.57 (1.14-18.25)	0.03

* The two-way interaction was not significant (P-value>0.10).

Model 3-B: Incidence from 1st Follow-up to 2nd Follow-up (Dichotomous)-adjusted for Age

In the bivariate analyses conducted in the previous section using incidence of ECC from 1st follow-up to 2nd follow-up (dichotomous) as a dependent variable, low birth weight, toothbrushing (yes/no, baseline), daily frequency of toothbrushing (baseline), presence of a regular dentist (2nd follow-up) and presence of a history of previous visit to a dentist (2nd follow-up) met the inclusion criterion of having logistic regression P-values of 0.15 or less (Table 85). History of previous dental visit (AUC visits 40 to 70) and presence of a regular dentist (AUC visits 40 to 70) were not included in the analyses, because they were statistically significantly correlated with the presence of previous dental visit at the 2nd follow-up (P-value=0.001) and the presence of a regular dentist at the 2nd follow-up (P-value<0.0001), respectively. Also, since toothbrushing (yes/no) and the frequency of toothbrushing at baseline were collinear (P-value<0.001), toothbrushing (yes/no) was manually eliminated from this model because the variable frequency of toothbrushing provided more information (none/once per day/twice per day, etc.) than the toothbrushing (yes/no).

A multivariable model was built including all of these factors and age as independent variables (Table 89). Then, a step-back, manual elimination algorithm was used and presence of a regular dentist at the 2nd follow-up was eliminated first because of having the highest p-value of 0.94 (Table 90). This was followed by eliminating the variable low birth weight with P-value of 0.48, resulting in a final reduced model that had daily frequency of toothbrushing (baseline) and presence of a history of a previous visit to a dentist at the 2nd follow-up (Table 91). Table 91 shows that increased daily frequency of toothbrushing at baseline was statistically significantly associated with decreased incidence of ECC from 1st to 2nd follow-up after adjustment for age (OR=0.30, P-value=0.01), while having a history of previous visits to a dentist at the 2nd follow-up was associated with higher incidence of ECC from 1st follow-up to 2nd follow-up, after adjustment for age (OR=4.53, P-value 0.03). No significant two-way interaction was detected among the variables in the final model at the significance level of 0.10. When exploring 0.05 as the significance level, no differences were detected, since all the P-values in the final reduced model were less than 0.05. Also, no significant two-way interaction was detected among the variables in the final model at the significance level of 0.05

Table 89. Multivariable logistic regression for incidence of ECC from 1st follow-up to 2nd follow-up (full model*, n=57)

Variables	OR (95%CI)	P-value
Age	2.50 (0.52-12.67)	0.26
Low birth weight (baseline)	0.56 (0.12-2.64)	0.46
Daily frequency of toothbrushing (baseline)	0.31 (0.13-0.73)	0.01
Presence of a history of a regular dentist (2nd follow-up)	0.94 (0.15-6.03)	0.95
Presence of a history of previous visit to a dentist (2nd follow-up)	4.34 (0.57-32.74)	0.16

* Toothbrushing (yes/no) was eliminated due to collinearity with daily frequency of toothbrushing (Pearson correlation=0.93, P-value<0.001)

Table 90. Multivariable logistic regression for incidence of ECC from 1st follow-up to 2nd follow-up (1st reduced model, n=58)

Variables	OR (95%CI)	P-value
Age	2.36 (0.50-11.21)	0.28
Low birth weight (baseline)	0.57 (0.12-2.72)	0.48
Daily frequency of toothbrushing (baseline)	0.31 (0.13-0.72)	0.01
Presence of a history of a regular dentist (2nd follow-up)	4.43 (2.11-17.69)	0.04

Table 91. Multivariable logistic regression for incidence of ECC from 1st follow-up to 2nd follow-up (2nd reduced model, n=58)

Variables	OR (95%CI)	P-value
Age	2.38 (0.51-11.18)	0.27
Daily frequency of toothbrushing (baseline)	0.30 (0.13-0.71)	0.01
Presence of a history of previous visit to a dentist (2nd follow-up)	4.53 (1.13-18.14)	0.03

* The two-way interaction was not significant (P-value>0.10).

<u>Comparison Between Model 3-A and Model 3-B</u>

Both the daily frequency of toothbrushing at baseline and presence of a history of a previous visit to a dentist at the 2nd follow-up were statistically significantly associated with the incidence of ECC from the 1st follow-up to the 3rd follow-up, even after adjustment for age at both α=0.1 and 0.05. Thus, there were not any differences between the two models regarding the mentioned variables.

<u>Model 4-A: Incidence from 1st Follow-up to 2nd Follow-up (Count)-not Adjusted for Age</u>

In the bivariate analyses conducted in the previous section using ECC increment from 1st follow-up to 2nd follow-up (dmfs count) as a dependent variable, presence of a regular dentist (2nd follow-up), previous history of a visit to a dentist (2nd follow-up) and self-brushing (2nd follow-up) met the inclusion criterion of having a logistic regression P-value of 0.15 or less (Table 92). History of previous dental visit (AUC visits 40 to 70) and presence of a regular dentist (AUC visits 40 to 70) were not included in the analyses, because they were statistically significantly correlated with the presence of previous dental visit at the 2nd follow-up (P-value=0.001) and the presence of a regular dentist at the 2nd follow-up (P-value<0.0001), respectively.

A multivariable model was built including all of these factors as independent variables (Table 93). A step-back elimination algorithm was used and previous history of a visit to a dentist (2nd follow-up) was eliminated first, because of having the highest p-value of 0.71 (Table 94). Then self-brushing was deleted for having P-value of 0.102, resulting in the final reduced model that had the history of previous visit to a dentist at the 2nd follow-up only (Table 95). Table 95 shows that children with a history of previous visit to a dentist had statistically significantly higher ECC dmfs count from 1st follow-up to 2nd follow-up than those who had no history of previous visit to a dentist at the 2nd follow-up (P-value of 0.03). When exploring 0.05 as the significance level, no differences were detected, since the P-value in the final reduced model was less than 0.05. Since there was only one single variable in the final reduced model (history of a previous visit to a dentist), we did not assess interaction.

Table 92. Bivariate negative binomial regression for incidence of ECC from 1st follow-up to 2nd follow-up for variables with P-values of 0.15 or less.

Variables	Estimate (SD)	IRR*	P-value
Presence of a history of a regular dentist (2nd follow-up)	1.29 (0.61)	3.63	0.04
Presence of a history of previous visit to a dentist (2nd follow-up)	1.04 (0.67)	2.83	0.12
Self-brushing (2nd follow-up)	-1.02 (0.70)	0.36	0.15

* IRR obtained from the exponentiation of the estimates.

Table 93. Multivariable negative binomial regression for incidence of ECC from 1st follow-up to 2nd follow-up (full model, n=61)

Variables	Estimate (SD)	IRR*	P-value
Presence of a regular dentist (2nd follow-up)	0.82 (1.08)	2.26	0.45
Presence of history of previous visit to a dentist (2nd follow-up)	0.45 (1.19)	1.57	0.71
Self-brushing (2nd follow-up)	-1.06 (0.93)	0.35	0.25

* IRR obtained from the exponentiation of the estimates.

Table 94. Multivariable negative binomial regression for incidence of ECC from 1st follow-up to 2nd follow-up (first reduced model, n=69)

Variables	Estimate (SD)	IRR*	P-value
Presence of a regular dentist (2nd follow-up)	1.25 (0.60)	3.48	0.04
Self-brushing (2nd follow-up)	-1.24 (0.76)	0.29	0.102

*IRR obtained from the exponentiation of the estimates.

Table 95. Multivariable negative binomial regression for incidence of ECC from 1st follow-up to 2nd follow-up (second reduced model, n=71)

Variables	Estimate (SD)	IRR*	P-value
Presence of a regular dentist (2nd follow-up)	1.29 (0.61)	3.63	0.03

* IRR obtained from the exponentiation of the estimates.

Model 4-B: Incidence from 1st Follow-up to 2nd Follow-up (Count)-adjusted for Age

In the bivariate analyses conducted in the previous section using ECC increment from 1st follow-up to 2nd follow-up (dmfs count) as a dependent variable, presence of a regular dentist (2nd follow-up), previous history of a visit to a dentist (2nd follow-up) and self-brushing (2nd follow-up) met the inclusion criterion of having a logistic regression P-value of 0.15 or less (Table 92). History of previous dental visit (AUC visits 40 to 70) and presence of a regular dentist (AUC visits 40 to 70) were not included in the analyses, because they were statistically significantly correlated with the presence of previous dental visit at the 2nd follow-up (P-value=0.001) and the presence of a regular dentist at the 2nd follow-up (P-value<0.0001), respectively

A multivariable model was built including all of mentioned variables as independent variables (Table 96). A step-back elimination algorithm was used and previous history of a visit to a dentist (2nd follow-up) was eliminated first, because of having the highest p-value of 0.85 (Table 97) when using α=0.10. Table 97 shows that that children who had a history of a previous visit to a dentist at the 2nd follow-up had statistically significantly higher ECC dmfs count from 1st follow-up to 2nd follow-up (P-value of 0.03) after adjusting for age. Also, children who brushed their own teeth by themselves (self-brushing=yes) had statistically significantly lower ECC dmfs count from 1st follow-up to 2nd follow-up (P-values of 0.09) after adjusting for age. Two-way interaction was assessed and there was no statistically significant two-way interaction between history of previous visit to a dentist at the 2nd follow-up and self-brushing at alpha level of 0.10.

Table 96. Multivariable negative binomial regression for incidence of ECC from 1st follow-up to 2nd follow-up (full model, n=61)

Variables	Estimate (SD)	IRR*	P-value
Age	0.56 (0.84)	1.74	0.51
Presence of a regular dentist (2nd follow-up)	1.02 (1.13)	2.78	0.36
Presence of history of previous visit to a dentist (2nd follow-up)	0.23 (1.24)	1.26	0.85
Self-brushing (2nd follow-up)	-0.98 (0.96)	0.37	0.31

* IRR obtained from the exponentiation of the estimates.

Table 97. Multivariable negative binomial regression for incidence of ECC from 1st follow-up to 2nd follow-up (first reduced model, n=69)

Variables	Estimate (SD)	IRR*	P-value
Age	0.70 (0.84)	2.02	0.40
Presence of a regular dentist (2nd follow-up)	1.38 (0.63)	3.96	0.03
Self-brushing (2nd follow-up)	-1.30 (0.76)	0.27	0.09

* IRR obtained from the exponentiation of the estimates.

When exploring 0.05 as the significance level, self-brushing (2nd follow-up) was eliminated with P-value of 0.09, resulting in the final reduced model including only presence of a regular dentist at the 2nd follow-up (Table 98). Table 98 shows that children who had a history of a previous visit to a dentist at the 2nd follow-up had statistically significantly higher ECC dmfs count from 1st follow-up to 2nd follow-up (P-value of 0.03).

Table 98. Multivariable negative binomial regression for incidence of ECC from 1st follow-up to 2nd follow-up (second reduced model, n=71)

Variables	Estimate (SD)	IRR*	P-value
Age	0.54 (0.79)	1.72	0.49
Presence of a regular dentist (2nd follow-up)	1.33 (0.62)	3.78	0.03

* IRR obtained from the exponentiation of the estimates.

Comparison Between Model 4-A and Model 4-B

Both models showed that presence of a regular dentist at the 2nd follow-up was statistically significantly associated with the incidence of ECC from the 1st follow-up to the 2nd follow-up, even after adjustment for age at both α=0.10 and 0.05. Before adjusting for age, self-brushing was not statistically significantly associated with ECC incidence (P-value of 0.102). However, after adjusting for age, self-brushing was statistically significantly associated with ECC incidence, when using α=0.10 as the significance level (P-value=0.09). When 0.05 was used as the significance level, no differences were detected between Model 4-A and Model 4-B.

CHAPTER 5

DISCUSSION

Prevalence of ECC

Prevalence Findings in our Study

The prevalence and incidence of Early Childhood Caries (ECC) and the proportions of tooth surfaces with caries experience increased with age during the 3-year follow-up in this study (from approximately age one to approximately age four years). The prevalence of dmfs was 1.1% (all decayed) at baseline (n=91); 12.8% (decayed=10.5% and filled=4.7%) at the 1st follow-up (n=86); 39.3% (decayed=21.4%, missing=2.4% & f=22.6%) at 2nd follow-up (n=84); and 65.8% (decayed=28.8%, missing=5.5% & filling=46.6%) at 3rd follow-up (n=73). The percentages of total surfaces of all teeth that were decayed/missing/filled were 0.1%, 1.4%, 6.2% and 10.4% at the baseline and three follow-up examinations, respectively.

The percentages of dmft on incisors, canines, first molars and second molars were 0.4%, 0.0%, 0.0% and 0.0% at baseline; 3.6%, 0.3%, 3.3%, and 1.5% at 1st follow-up; 9.8%, 1.8%, 10.1%, and 9.2% at 2nd follow; and 11.1%, 4.1%, 15.8% and 31.2% at 3rd follow-up, respectively The percentages of dmfs on incisors, canines, first molars and second molars were 0.1%, 0.0%, 0.0% and 0.0% at baseline; 2.4%, 0.1%, 0.8%, and 0.6% at 1st follow-up; 8.2%, 0.8%, 7.6%, and 6.3% at 2nd follow-up; and 10.2%, 2.2%, 12.6% and 16.7% at 3rd follow-up, respectively. The increases in the prevalence over time are due partly to increased time of exposure to dental caries risk factors, exposure to new caries risk factors, such as sweets and pop, which are not usually consumed by infants, and increased number of teeth at risk.

Overall Prevalence in Other US and African American Studies in Comparison to Our Study

Although all of the children in our study received fluoride varnish treatments at six-month intervals, substantial percentages of them developed additional dental caries experience. Our study showed that the prevalence rates of ECC among these African-American children from Perry County, Alabama were approximately 2.0%, 18.4% and 31.4% more than those reported by NHANES, 1999-2002, for children aged two, three and four years, respectively[3]. NHANES 1999-2002, did not report caries prevalence among very young children less than two years of age, as we reported in our study.

Also, the results of our study showed higher prevalence of ECC than that reported in most other studies. For example, a randomized controlled trial by Leverett et al.[107] (discussed earlier in Chapter Two) reported that the prevalence rates of ECC were 3.0% and 9.0% among children who were living in non-fluoridated counties in southern Maine and did not have fluoride supplements, when they were three (n=474) and five (n=400) years of age, respectively[107]. The prevalence rates of ECC were 4.3% and 7.5% among children who were living in non-fluoridated counties in southern Maine and received dietary fluoride supplements, when they were three (n=464) and five (398) years of age, respectively[107].

A study by Warren et al.[12] was conducted to assess the prevalence of ECC among 698 children (mostly Caucasian) with mean age of 4.6 (discussed earlier in Chapter Two). Warren et al. reported that the prevalence of ECC was 27% when only cavitated lesions were included in the analyses, compared to 65.8% in our study when the children were four years old. However, a study by Kolker et al.[60] (mentioned earlier in Chapter 2) showed a very high prevalence of ECC among African-American children in Detroit, Michigan. The prevalence of ECC was 64.2% and 74.2% among three and four year old children, respectively, which is even higher than what was reported in our study (39.3% and 65.8%, respectively). In the Kolker et al.[60] study, they followed the International

Caries Detection and Assessment System (ICDAS) criteria, which means that they included non-cavitated lesions in their analyses.

Overall Prevalence in Other International Studies in Comparison to Our Study

The prevalence of ECC reported in our study also was higher than the prevalence reported in many of the European studies. For example, Wendet et al.[31] (discussed previously in Chapter Two) assessed the prevalence of ECC among 671 Swedish children aged one year and followed then for one year. The prevalence of ECC was 0.5% and 7.7% at baseline and follow-up, respectively. Although non-cavitated lesions were included in the analyses and these children did not receive fluoride varnish application, the prevalence was less than that reported in our study (1.1% and 12.9% at age one and two, respectively).

In Japan, Tanaka et al.[108] conducted a cross-sectional study to assess the relationship between breastfeeding and dental caries. More than 2,000 children aged three years were recruited and had dental examinations at the tooth level (cavitated only). Tanaka et al.[108] showed that the prevalence of ECC was 20.7% and the mean dmft was 0.7, which was lower than the prevalence and the mean dmft reported in our study at age three years (39.3% and 1.6, respectively).

Njoroge et al.[109] conducted a cross-sectional study to assess the prevalence and patterns of ECC among 336 three to five year old children in Kiambaa, Kenya. The overall mean dmft was 2.46, while the mean numbers of decayed, missing and filled teeth were 2.36, 0.08 and 0.02, respectively. However, in our study, mean dmft was 1.64 and 3.33 at age three and four, respectively, while mean numbers of decayed, missing and filled teeth were 0.46, 0.06 and 1.12 at the 2nd follow-up (mean age=3 years), respectively, and 0.56, 0.15 and 2.16 at the third follow-up (mean age=4 years), respectively. Although the overall mean dmft is comparable between the Njoroge et al.[109]

study and our study, we can clearly see the substantially higher percentage of filled teeth among the children in our cohort compared to the Kenyan children. This finding might be due to the success of referrals to local dentists in the area, which was basically one of the benefits of our study to the families in addition to fluoride application and oral hygiene instructions. Njoroge et al.[109] showed that the overall prevalence of caries was 47.0% and 55.0% at age three and four, respectively, and it was 39.3% and 65.8% at age three and four years in our study, respectively. The overall prevalence rates of the Njoroge et al.[109] study were high and comparable to the prevalence rates in our study, but we need to acknowledge that Njoroge et al.[109] recruited older children who did not receive fluoride varnish, so they were more vulnerable to dental caries.

Cleaton-Jones et al.[110] conducted a cross-sectional study to assess the prevalence of ECC among black children aged two to five years in South Africa. The results showed a higher prevalence of ECC at age two (36.0%, n=64) compared to our study (12.8%, n=86). However, the prevalence of ECC was lower at both age three (35.0%, n=78) and age four (46.0%, n=76), compared to our study (39.3%, n=84) and (65.8%, n=73), respectively.

Prevalence by Tooth Type in Other Studies in Comparison to Our Study

Chankanka et al.[111] conducted analyses to assess longitudinal associations between dental caries and risk factors in 156 children as part of the ongoing Iowa Fluoride Study. The study showed detailed descriptive analyses by tooth type at age 5 years, with mean dmfs of 0.09, 0.06, 0.61 and 0.37 among incisors, canines, first molars and second molars, respectively. Thus, mean dmfs was the highest among 1st molars, followed by 2nd molars, incisors and canines. However, in our study at age 4 (3rd follow-up), mean dmfs was the highest among second molars (1.25), followed by incisors (0.88), first molars (0.36) and canines (0.16).

In the Njoroge et al.[109] study that assessed the prevalence and patterns of ECC cross-sectionally among 336 three to five year old Kenyan children, the published figure showed that caries experience on 2nd molars was higher than on 1st molars, which was the case in our study at the 3rd follow-up (mean age=4 years).

Unfortunately, very few studies have been conducted to assess the prevalence of ECC at the tooth-type and surface levels among young children, so the comparisons were limited.

Summary of the Prevalence Studies in Comparison to Our Study

Generally speaking, the prevalence and severity (dmft and dmfs) of ECC reported in our study were higher than those reported in most of the published studies and national surveys at age one, two, three and four years in the United States. Furthermore, the prevalence of ECC, mean dmft and mean dmfs were higher than those reported in most of the studies in other developed and developing countries at all the mentioned age groups.

Incidence of ECC

Incidence Findings in Our Study

When unerupted surfaces were included, the three one-year person-level net ECC incidence rates were 12.8% (n=86), 38.6% (n=83), and 56.2% (n=73), from baseline, 1st follow-up and 2nd follow-up, respectively. Two-year and three-year net ECC incidence rates from baseline were 39.3% (n=84) and 65.8% (n=73), respectively, while two-year net ECC incidence from the 1st follow-up to 3rd follow-up was 66.7% (n=72).

The three one-year person-level net ECC incidence rates (unerupted surfaces included) for incisors, canines, 1st molars and 2nd molars were 11.6%, 1.2%, 5.8% and 1.2%, respectively, from baseline (n=80), 22.9%, 3.6%, 19.3% and 14.5%, respectively, from the 1st follow-up(n=83), and 23.3%, 6.9%, 16.4% and 43.8%, respectively, from the

2nd follow-up (n=73). Two-year net ECC incidence rates for incisors, canines, 1st molars and 2nd molars from baseline (n=78) were 28.6%, 3.6%, 19.1% and 14.3%, respectively, while the three-year net ECC incidence rates of incisors, canines, 1st molars and 2nd molars from baseline (n=67) were 35.6%, 9.6%, 27.4% and 49.3%, respectively. The two-year net ECC incidence from the 1st follow-up to 3rd follow-up (n=72) for incisors, canines, 1st molars and 2nd molars were 33.3%, 9.7%, 27.8%, 50.0%, respectively.

The most striking findings from the tooth-level analyses of incidence were that the incidence of ECC on second molars was higher (49%, when unerupted surfaces included during the three-year follow-up) than that of first molars (27%, when unerupted surfaces included). This finding was reported in spite of the fact that first molars erupted first and were exposed to the oral environment, bacteria and other risk factors for longer periods of time, compared to 2nd molars that erupt later on. Biologically, this might be due to the higher susceptibility of newly-erupted teeth to dental caries because of the colonization by microorganisms (window of infectivity). Also, this might be due to the difficulty in accessing these teeth during toothbrushing, considering that 13.6% of the children reportedly brushed their own teeth at baseline and rates of the children brushing their own teeth were approximately 56.0% and 78.0% for the AUC composite variables from approximately age 1.5 to three years and at 2nd follow-up, respectively. In addition, this higher incidence of ECC on 2nd molars compared to 1st molars might be due to the difficulty in applying fluoride varnish on the second molars and the difficulty in ensuring a dry environment needed for proper application of fluoride varnish, compared to 1st molars. Also, it is important to mention, that from the 2nd follow-up to the 3rd follow-up, approximately 43.8% of the children developed new carious lesions on their 2nd molars compared to approximately 16.4% of the children who developed new carious lesions on the 1st molars.

Incidence without Intervention

Few studies were conducted to assess the incidence of ECC as a main primary outcome. For instance, Grindefjord et al.[64] examined caries progression among 692 Swedish children who were examined when they were 2.5 years old and reexamined one year later when the children were 3.5 years old (discussed earlier in Chapter Two). The findings of this study showed that there was an increase of 26.4% in the prevalence of ECC during the one-year follow-up (d1 lesions included), while our study showed that there was a 26.5% increase in the prevalence of ECC (cavitated only) between age two and three years, and there was another 26.5% increase between age three and four years. Also, mean dmfs increment was 1.85 in the Grindefjord et al. study (from age 2.5 to 3.5 years), while mean dmfs increments were 5.1 (from age two to three years) and 4.04 (3-4 years) in our study. Mean dmft increments in our study were similar to those reported by Sakuma et al.[66] (discussed earlier in Chapter Two). Sakuma et al.[66] assessed dmft increments in 5,107 children from four different cities from age 1.5 years to three years, without mentioning if it was at the cavitated, non-cavitated or the combined cavitated and non-cavitated levels. Mean dmft increments ranged from 1.17 to 1.39 in these four different cities, while mean dmfs increments were 0.43 and 1.19 from ages one to two and two to three years in our study, respectively.

Litt et al.[68] examined 184 African-American children at baseline (children were three to four years old) and one-year follow-up. Mean dmfs at the cavitated level was 2.8 at baseline, while it was 4.6 at the one-year follow-up (dmfs increment=1.8), compared to our study that showed higher incidence and dmft/dmfs increments of ECC in spite of semi-annual fluoride varnish application, with mean dmfs increased from 6.28 at age three years to 10.32 at age four years (dmfs increment=4.04).

The prevalence and the incidence rates of ECC were higher among our children than that reported in other studies, despite the fact that children in our study received fluoride varnish application semiannually. Thus, better comparisons will be with children who received regular topical varnish applications.

Incidence with Intervention

As mentioned before, since the children in our study received fluoride varnish application semiannually, better comparisons will be with studies that applied fluoride varnish to the study participants. For example, Holm et al.[112] conducted a study to assess dental caries-preventive fraction among 225 Swedish children aged 3 years at baseline. The children were assigned to either control group (n=113) who did not receive fluoride varnish or test group (n=112) who received fluoride varnish application twice annually for two years. The study showed that mean two-year caries increments (dmfs count) were 2.1 and 3.7 surfaces for the test and the control group, respectively. In our study, mean caries increment (dmfs count) was 4.04 for one year from the 2nd follow-up (mean age=three) to the 3rd follow-up (mean age=four), which was higher even than the control group in Holm et al.[112] study for two years. One more important thing was that Holm et al.[112] used bitewing radiographs in order to rule in or rule out any caries in the interproximal surfaces, so the accuracy of their dmfs increments could have been greater than our study which might have underestimated dmfs increments because no radiographs were used.

Autio-Gold et al.[113] conducted a study to assess the efficacy of fluoride varnish on early enamel carious lesion in the primary dentition of 142 children (mostly African-Americans) aged three to five years at baseline who were recruited from Head Start schools in Alachua County, Fla. The children were randomized into either test group (n=59, 71.2% African-Americans) who received fluoride varnish at baseline and four months later or control group (n=83, 72.8% African-Americans) and followed for 9 months. The study showed that mean dmfs increments were 0.54 and 1.47 and mean dmft increments were 0.05 and 0.5 in the test group and control group, respectively, while mean dmfs increment was 4.04 and mean dmft increment was 1.69 in our study from the 2nd follow-up to the 3rd follow-up (from age three to four years), which was even higher than the control group in the Autio-Gold et al. study[113].

Petersson et al.[114] conducted a study to assess the efficacy of different fluoride prophylactic programs in 376 Swedish children aged three years at baseline. The study showed that the mean dmfs increment among children who received fluoride varnish semiannually and placebo (fluoride-free) dentifrice (n=85) was 1.7 for two years. In our study, mean dmfs increment from 2nd follow-up to 3rd follow-up (from age three to age four years) was 4.04, which was even higher than two-year caries increment in Petterson et al. study.

In summary, the findings of our study showed that the incidence of ECC, dmfs increments, dmft increments were higher than those reported in most of the published studies and they were even higher than the incidence of ECC, dmfs increment, dmft increment among children who did not receive fluoride varnish. To our knowledge up to the time this report was written, there were no other studies that assessed the incidence of ECC among very young children by tooth type.

Oral Hygiene and Dietary Behaviors

Oral Hygiene Behaviors

As mentioned previously, in addition to the semiannual fluoride varnish application, oral hygiene instructions were provided by dental personnel, including the examiners, dental hygienist, and dental residents in the Department of Pediatric Dentistry, UAB. Also, monetary compensation ($20) and preventive oral kits that contained a toothbrush were provided to all the participants at each visit (approximately every six months). Family members were provided with additional supplies such as fluoride toothpaste, dental floss, dental flossers, and extra toothbrushes, as available.

Oral hygiene information for the children was collected at the baseline and follow-up questionnaires. This included questions about toothbrushing (yes/no), daily frequency of toothbrushing, use of toothpaste, brand names of toothpastes and whether the children were reportedly self-brushing. Approximately 41.5% of the children reportedly had their teeth brushed (either self-brushing or by their caregivers) at baseline (mean age=1). Among those who had their teeth brushed, 61.5% reportedly brushed twice per day. All the children reportedly had their teeth brushed (either self-brushing or by their caregivers) in our study (100%) at the 2nd follow-up (mean age=three years). Our study also showed that 33.3%, 94.0% and 100% of the children reportedly used toothpaste at the baseline, and for the AUC composite variables from approximately age 1.5 to three years and at the 2nd follow-up, respectively.

National surveys and other studies reported lower percentages of toothbrushing and use of toothpaste among children at the same age group as in our study. For example, Holm et al.[112] assessed the effectiveness of fluoride varnish application among 225 Swedish children (controls=113 and test=112) for 2 years (mentioned earlier), reporting that 97.4% and 98.2% of the children brush their teeth at the end of the study (mean age=5 years) in the test and the control groups, respectively, and 41.0% and 44.3% of the children brushed their teeth twice or more per day, respectively, which is less than that reported in our study at an earlier age (mean age=3 years), with 100% brushing (either self-brushing or by their caregivers) and a mean daily frequency of toothbrushing of 1.99. Also, Holm et al.[112] showed that 70.5% used toothpaste at age 5, compared to 100% in our study at age 3. Wong et al.[115] conducted a study in Hong Kong to assess caries increment over two years in 358 preschool children aged three to four years at baseline (mean age=3.2 years). Baseline questionnaires showed that 63.7% and 78.7% of the children brushed their teeth twice or more per day and used toothpaste, respectively, compared to our study in which almost all of the children reportedly had their teeth brushed twice per day (either self-brushing or by their caregivers) and used toothpaste at the 2nd follow-up (mean age=3).

Warren et al.[67] (discussed earlier in Chapter Two) assessed risk factors associated with caries incidence for 18 months among very young (age=6 to 24 months at baseline), low SES children. Two-hundred and twelve WIC-enrolled children were recruited (volunteers), and questionnaires were completed at baseline, four to five months, nine months and 13 to 14 months after baseline by the mothers. At baseline, 51.9% and 26.2% of the children reportedly had their teeth brushed and used fluoride toothpaste, respectively, while in our study 41.5% and 33.3% of the children had their teeth brushed (either self-brushing or by their caregivers) and used toothpaste.

Tsai et al.[54] conducted a study (discussed earlier) to assess the risk indicators for ECC in Taiwan among 981 children younger than 6 years of age. Among the children who were one (n=114), two (n=182), three (n=182), four (n=178), five (n=147) and 6 (n=178) years old, 67.7%, 30.0%, 33.6%, 37.2%, 31.9% and 43.9% of the children reportedly had their teeth brushed at least once daily, respectively, while in our study, 41.5%, 100%, 100% of the children reportedly had their teeth brushed at baseline (mean age=1 year), in the AUC composite variables from age 1.5 to three years and 2nd follow-up (mean age=3 years), respectively. Also, Kumarihamy et al.[88] conducted a study to assess the prevalence of ECC in children aged 1-2 years in a semi-urban area of Sri Lanka (discussed earlier in this chapter). The study reported that 32.9% of the children had their teeth brushed once or less than once per day, while 67.1% of the children had their teeth brushed twice or more per day, and this was higher than that reported in our study at baseline when the children were three to 23 months old (74.5% brushed their teeth once or less than once per day, while 25.5% reportedly brushed their teeth twice per day). Also, Kumarihamy et al.[88] reported that 23.1% of the children did not use toothpaste, while in our study, 56% of the children reportedly did not use toothpaste at baseline (mean age=three to 23 months). A national survey in Australia showed that

44.4% of the children who were recruited to represent a national sample of the Australian children aged two to three years (n=4,606) had their teeth brushed at least twice per day (Klipatrick et al.[116]). However, in our study, 93.8% of the children had their teeth brushed twice per day at the second follow-up (mean age=3 years). Tanaka et al.[108] recruited 2,056 Japanese children aged three years and collected oral hygiene practices information from their parents cross-sectionally to assess risk factors associated with ECC (discussed earlier in this chapter). Descriptive analyses showed that approximately 85% of the children reportedly used toothpaste and 44% of the children received regular dental check-ups. In our study and when the children were 3 years old, 100% of the children used toothpaste and 51% had a regular dentist.

Very few studies reported the percentage of the children who brushed their own teeth (self-brushing). For example, Guido et al.[117] in the previously mentioned study showed that 31% of the children who were two to six years old (n=63) brushed their own teeth, while 56% of the children in our study reportedly brushed their own teeth in the AUC composite variables from age 1.5 to three years. Chu et al.[118] conducted a study (discussed earlier in this chapter) to report oral health status and behaviors of preschool children aged four to six years in Hong Kong (n=764). The results showed that 20% of the children reportedly brushed their own teeth, which was lower than that reported in our study at the 2nd follow-up (78%).

It is important to mention that there was a question about the brand name of toothpaste the children reportedly used at baseline and each of the follow-up visits. As it was mentioned before, it was an open-ended question, so the parents had the liberty to report whatever they believed to be correct. However, the information they provided was not sufficient to rule in or rule out the presence of fluoride in the toothpaste. For instance, as mentioned earlier, many of the parents reported that their children used Orajel™ toothpaste, without providing more information. Since there are two types of Orajel™: "Orajel™ for Kids", which is fluoridated, and "Orajel™ for Toddlers", which is non-fluoridated, we were not able to determine whether it was fluoridated or not. Also, some parents reported that their children used "Oral-B™ Training toothpaste" and the Oral-B™ customer service staff confirmed on a phone call that they did not have such a product. Originally, we were planning to use fluoride content of toothpaste as a dichotomous independent variable (children using fluoride toothpaste vs. children using non-fluoridated toothpaste), and assess its relationship with different dependent variables. However, because we could not make a definitive decision about the fluoride content of the toothpaste, we decided to exclude this variable from the analyses. This problem probably could have been avoided by asking the participants to bring the toothpaste with them, providing a detailed list of the full brand/product names of toothpastes available in the market or possibly asking specifically about the fluoride content of toothpaste.

In summary, the results of the our study showed different findings regarding oral hygiene behaviors, such as toothbrushing and the use of toothpaste, compared to dental hygiene practices reported in other published articles. For example, the percentage of children who reportedly brushed their own teeth was high compared to the limited number of studies that reported self-brushing. Unfortunately, most of the published articles that assessed the relationships between different oral hygiene practices and ECC did not show detailed descriptive analyses about the percentages of the children who reportedly had their teeth brushed and used toothpaste, so the comparison was limited to the few studies that showed detailed information.

Dietary Practices

Finding in Our Study

In our study, at baseline, 36.1%, 61.9%, 99.0% and 91.6% of the children reportedly consumed milk, infant formula, water and 100% natural juice, respectively, while only 1.0% of the children reportedly consumed sugar-added beverages. The daily frequencies of consumption of milk, infant formula, water, 100% natural juice and sugar-added beverages were 3.3, 4.2, 2.4, 2.4, 3.0 cups, respectively, among consumers. Also, the AUC composite variable from age 1.5 to three years showed that almost all the children (99.0%) reportedly consumed milk, water and sugar-added beverages, with mean daily frequencies of 3.4, 4.1 and 1.9, respectively, while 84.0% of the children reportedly consumed 100% juice with mean daily frequency of consumption of 0.9 cups.

Also, the AUC composite variable from age 1.5 to three years showed that 19.0% and 99.0% of the children reportedly consumed sweets and sweetened food, respectively, with mean daily frequency of 0.2 and 1.7 times, respectively, among all children (consumers and non-consumers). However, none of the parents reported adding plain sugar to the foods or drinks of their children at any of the visits.

The frequency and the quantity of consumption of beverages were identical at the person level at all the visits (for every single sub-category and for every person). In other words, parents who reported that their children consumed three cups of milk a day also reported that their children consumed milk three times per day. If we had chosen a different measure of quantity other than cups, such as ounces, we could have obtained different results. Thus, we included the frequency of consumption of beverages in the bivariate and multivariable analyses in order to avoid redundancy.

Comparison with Dietary Findings from American Studies

NHANES data showed that the prevalence of consumption of sugar-added beverages (regular soda pop, all soft drinks except diet drinks, sugar-added juices, sugar-added fruit drinks and sport drinks) among children (n=8,627) aged two to 11 years has decreased from a mean of 78% in the 1999-2000 survey to 66% in the 2007-2008 survey and the decrease was statistically significant[119]. However, NHANES data (1999-2008) showed that the odds of consumption of sugar-added beverages among African-American children was 30% more than the odds of consumptions of sugar-added beverages among White-American children (OR=1.3, 95% CI 1.10-1.55), while there were no statistically significant differences in the consumption of sugar-added beverages between Hispanic children and White-American children (OR=1.06, 95% CI 0.87-1.28) nor between children of other races and White-American children (OR=0.86, 95% CI 0.61-1.23). In our study, the results showed that 100% of the children aged three years consumed sugar-added beverages and 98.77% of the children consumed sugar-added beverages in the AUC composite variable from age 1.5 to three years which was higher than that reported in NHANES.

Comparison with Dietary Findings from International Studies

Hsu et al.[120] conducted a study to assess the feeding bottle usage and its association with childhood allergy and asthma. They recruited 14,862 children aged two to seven years from southern Taiwan from more than 201 kindergartens and 259 day care centers. Among the children, 3.7%, 12.4%, 28.4%, 38.1% and 17.4% were less than three, three, four, five and six to seven years old, respectively. The study showed that 61.4% and 98.2% of the children had a history of breastfeeding and use of infant formula, respectively, while in our study only 3.1% of the children had a history of breastfeeding (ever) at baseline and all the children (100%) consumed milk and/or formula at baseline.

Tsai et al.[54] showed that 79.1% of the one-year old children reportedly took a bottle to bed, while in our study, 28.9% of the children reportedly had a history of night or nap bottle feeding at baseline (approximate age one). Also, Tsai et al.[54] showed that, at baseline, 35.2% of the children reportedly consumed sugar-added beverages, while 1.0% of the children in our study reportedly consumed sugar-added beverages at baseline (age=three to 23 months). Guido et al.[117] conducted a study to assess the prevalence of dental caries among children aged 2 to 13 years in the state of Hidalgo, Mexico. Descriptive analyses of this study showed that, among children who were 2-6 years old (n=63), the percentages of children who reportedly consumed 100% juice, juice drinks, water, milk and soft drinks at least once per day were 68.3%, 55.6%, 100%, 93.7% and 71.4%, respectively. However, exact definitions of 100% juice, juice drinks, milk and soft drinks were not mentioned in the article. In our study, 83.9% of the children reportedly consumed 100% juice in the AUC composite variable from age 1.5 to three years, while 98.77% of the children reportedly consumed milk, water and the sugar-added beverages AUC composite variable from age 1.5 to three years. Kumarihamy et al.[88] designed a study to assess the prevalence of ECC among Sri Lankan children aged 1 to 2 years (mentioned before) and the results showed that 83.2% of the children had a history of night bottle-feeding, compared to 28.87% in our study. Also, Kumarihamy et al.[88] reported a very high percentage of the parents (74.7%) added sugar to their children's milk, while none of the parents in our study reported adding plain sugar to the food or the beverages at baseline, intermediate or follow-up questionnaires.

In summary, a very high percentage of the children consumed milk, water or sugar-added beverages (each=98.8%) in the AUC composite variables from age 1.5 to three years and about 83.9% of the children reportedly consumed 100% juice in the AUC composite variable from age 1.5 to three years. The prevalence of consumption of sugar-added beverages in our study was higher than that reported in most of the studies and national reports in the U.S. and worldwide.

Risk Factors

Findings of Our Study

The relationships between the three-year incidence of ECC (baseline to the 3rd follow-up, dichotomous and count) and the incidence of ECC from the 1st follow-up to the 2nd follow-up (dichotomous and count) and different individual demographic, medical, dietary and dental practices were assessed in the bivariate analyses. Variables with P-values of 0.15 or less in the bivariate analyses were included in the multivariable analyses. Since age is an important confounder for the incidence of dental caries, as mentioned before in Chapter Two, and because the children were 3 to 22 months at baseline, we adjusted for age at the beginning of the incidence period. Differences in age at the beginning of the incidence period among children were associated with different number of erupted teeth, and hence different number of surfaces at risk for dental caries. Thus, it was important to adjust for age in order to control for the discrepancies in the incidence of dental caries.

Table 99 shows the multivariable models we developed and the P-values for the variables that remained in the final model, using $\alpha=0.10$ as the significance level.

Despite choosing a p-value of 0.10 as the significance level, we did not adjust for multiple comparisons, because using a more conservative significance level with limited sample size would be associated with greater chance of type II error (failure to reject a false null hypothesis).

As was mentioned earlier, α of 0.10 was used as the significance level because of the exploratory nature of our study and limited sample size which decreased the power of our study to detect differences in the incidence of ECC among children with different behavioral risk factors. However, we also explored models with 0.05 as the significance level.

Findings from Model 1

At $\alpha=0.10$ as the significance level, Model 1 showed that premature delivery and greater AUC of daily frequency consumption of 100% juice were associated with lower three-year incidence of ECC (dichotomous, both P-values=0.049), without adjusting for age. Also, AUC of consumption of sweetened foods was associated with higher three-year incidence of ECC (dichotomous, P-value=0.002) before adjusting for age. For Model 1-A, there would have been no differences in Model 1-A, because all the P-values were less than 0.05.

Table 99. Multivariable models and the variables in the full and final reduced models (with P-values)

Model (dependent variable)	Variables included in the full model (direction of the association)	Variables remained in the final model (P-value)[*]
Model 1-A: Three-year incidence from baseline to 3rd follow-up (dichotomous)-not adjusted for age.	* Premature delivery (-) * AUC of daily frequency consumption of 100% juice (-) * AUC of daily frequency of consumption of sweetened foods (+)	* Premature delivery (0.049) * AUC of daily frequency consumption of 100% juice (0.049) * AUC of consumption of sweetened foods (0.002)
Model 1-B Three-year incidence from baseline to 3rd follow-up (dichotomous) -adjusted for age.	* Age (-) * Premature delivery (-) * AUC of daily frequency consumption of 100% juice (-) * AUC of daily frequency of consumption of sweetened foods (+)	* Age (0.28) * Premature delivery (0.048) * AUC of daily frequency consumption of 100% juice (0.059) * AUC of consumption of sweetened foods (0.002)
Model 2-A: Three-year incidence from baseline to 3rd follow-up (dmfs count)-not adjusted for age.	* Mother's DMFS (+) * Presence of history of previous dental visit at the 2nd follow-up (+)	* Presence of history of previous dental visit at the 2nd follow-up (0.15)
Model 2-B: Three-year incidence from baseline to 3rd follow-up (dmfs count)-adjusted for age.	* Age (+) * Mother's DMFS (+) * Presence of history of previous dental visit at the 2nd follow-up (+)	* Age (0.73) * Presence of history of previous dental visit at the 2nd follow-up (0.16)

Table 99-Continued

Model 3-A: Incidence from 1st follow-up to 2nd follow-up (dichotomous)-not adjusted for age	* Low birth weight (-) * Daily frequency of toothbrushing (-) * Presence of a regular dentist at 2nd Follow-up (+) * Presence of a history of previous visit to a dentist at 2nd follow-up (+)	* Daily frequency of toothbrushing (0.01) * Presence of a history of previous visit to a dentist (0.03)
Model 3-B: Incidence from 1st follow-up to 2nd follow-up (dichotomous)-adjusted for age	* Age (+) * Low birth weight (-) * Daily frequency of toothbrushing (-) * Presence of a regular dentist at 2nd Follow-up (+) * Presence of a history of previous visit to a dentist at 2nd follow-up (+)	* Age (0.27) * Daily frequency of toothbrushing (0.01) * Presence of a history of previous visit to a dentist (0.03)
Model 4-A: Incidence from 1st follow-up to 2nd follow-up (count)-not adjusted for age.	* Presence of a regular dentist at 2nd follow-up (+) * Previous history of a visit to a dentist at 2nd follow-up (+) * Self-brushing at 2nd follow-up (-)	* Presence of a regular dentist (0.04)
Model 4-B: Incidence from 1st follow-up to 2nd follow-up (count)-adjusted for age.	* Age (+) * Presence of a regular dentist at 2nd follow-up (+) * Previous history of a visit to a dentist at 2nd follow-up (+) * Self-brushing at 2nd follow-up (-)	* Age (0.40) * Presence of a regular dentist (0.03) * Self-brushing (0.09)

With adjustment for age, there were no meaningful differences in the findings at $\alpha=0.10$. However, if a P-value of 0.05 had been used for Model 1-B, we would have had only AUC of consumption of sweetened foods statistically significantly associated with three-year incidence of ECC. The relationship between the consumption of 100% juice and dental caries is not well-established. Generally speaking, 100% juices contain fructose and glucose[121]. Cariogenic bacteria, such as *Streptococcus mutans*, are more effective in metabolizing sucrose (present in sugar-added beverages), rather than fructose or glucose, to produce extracellular glycan[122-124]. Extracellular glycan is an important component in the formation of dental plaque, as it facilitates dental plaque adherence to the tooth surface. Extracellular glycan enables acid's diffusion, which increases the risk for dental caries[122-124]. In our study, the increased daily frequency of consumption of 100% juice was correlated with decreased daily frequency of consumption of soda pop and other sugar-added beverages that most commonly have sucrose or high-fructose corn syrup (corn syrup is usually composed of fructose and sucrose). Our descriptive analyses showed that the AUC composite of daily frequency of consumption of 100% juice was negatively correlated with the AUC composite of daily frequency of consumption of sugar-added beverages (Spearman Correlation= -0.60 with P-value<0.0001). The decreased daily frequency of consumption of sugar-added beverages being associated with increased AUC composite of daily frequency of consumption of 100% juice might be a reason for the protective effect of the increased daily frequency of consumption of 100% juice.

Generally speaking, 100% juices are more expensive than regular soda pop and other sugar-added beverages. So, children who reportedly consumed more 100% juice might be from higher SES and more educated families which can afford them more easily. Thus, SES and education are important confounders. However, we were unable to control for these variables in our analyses, because income- and education-level information was not collected.

Our finding regarding the negative association between greater consumption of 100% juice and ECC incidence was consistent with that reported by Chankanka et al.[111] who conducted secondary analyses of the Iowa Fluoride Study to assess the effect of frequencies of consumption of beverages on cavitated and non-cavitated caries incidence in 156 children for the primary (exam 1: age five years), mixed (exam 2: age nine years), and permanent dentitions (exam 3: age 13). The results of that study show that increased frequency of 100% juice was significantly associated with fewer non-cavitated and cavitated caries surfaces, with P-values of 0.003 and <0.0001, respectively.

There were some studies that showed a statistically significant relationship between sweetened food and dental caries. For example, a study by Seow et al.[95] assessed the relationships between ECC among 670 Australian children aged 0-4 years living in non-fluoridated areas and different dietary practices (discussed earlier in Chapter Two). The results showed that added sweeteners to bottles (yes/no), sugar added to weaning solids (yes/no), sugar in fluids (more than two times daily) (yes/no), and sugary snacks (yes/no) were statistically significantly associated with ECC, with P-values of <0.001, 0.021, 0.002, and 0.047, respectively.

Although the relationship between sugar-added beverages (yes/no) and ECC incidence was not assessed, we had limited ability to identify sugar-added beverages (yes/no) as a risk factor, because approximately 99% of the children reportedly consumed sugar-added beverages. Also, relatively high percentage (11.3%) of our children was born prematurely (before 37 weeks). This gave us some variability in detected differences in caries incidence among those who were born prematurely and those who were full term.

Two-way interactions were assessed, using different pairs of the variables that remained in the final model, and there were no statistically significant interactions at $\alpha=0.10$. When assessing the interaction between AUC of 100% juice and AUC of sweetened food, AUC of 100% juice was converted to a dichotomous variable, using different cutoffs (=<one cup vs. >one cup, =<1.8 cups vs. >1.8 and =<two cups vs. > two cups), and there were no statistically significant interactions at α=0.10.

Findings from Model 2

At the level of α=0.10, no variable was statistically significantly associated with the three-year incidence of ECC (count) before or after adjustment for age.

Findings from Model 3

At α=0.10, greater daily frequency of toothbrushing at baseline was statistically significantly associated with decreased ECC incidence from the 1st follow-up to the 2nd follow-up (dichotomous), with P-value of 0.01. Having a history of a previous visit to a dentist was statistically significantly associated with increased incidence of ECC from the 1st follow-up to the 2nd follow-up (dichotomous), with P-value of 0.03. The same pattern of results was reported after adjusting for age.

Many studies showed that toothbrushing and the increased frequency of toothbrushing were associated with decreased prevalence and incidence of dental caries. A study by Tsai et al.[54] assessed the relationship between toothbrushing and prevalence of ECC among 981 Taiwanese children younger than 6 years of age (mentioned earlier in Chapter Two). Multiple logistic regression analyses showed that toothbrushing every night before bedtime was statistically significantly associated with lower prevalence of ECC (P-value<0.05, exact P-value not mentioned). Chankanka et al.[111] assessed the effect of toothbrushing on cavitated and non-cavitated caries incidence in the Iowa Fluoride Study children with primary, mixed, and permanent dentitions (discussed earlier in Chapter Two). The results of this study showed that increased daily frequency of toothbrushing was statistically significantly associated with fewer non-cavitated caries surfaces (P-value of 0.03). However, increased daily frequency of toothbrushing was not statistically significantly associated with non-cavitated caries surfaces (P-value of 0.08).

Other studies found that toothbrushing was not statistically significantly associated with caries prevalence and incidence. For example, Kumarihamy et al.[88] assessed the relationship between the prevalence of ECC in 422 1- to 2-year-old Sri Lankan children and the daily frequency of toothbrushing (mentioned earlier in Chapter Two). The study showed that the difference in the mean dmft (non-cavitated lesions included) between children who had their teeth brushed =<1 per day and those who had their teeth brushed >1 per day was not statistically significant with P-value of 0.18. Warren et al.[86] assessed the relationship between dental caries experience (non-cavitated lesions included) and toothbrushing in 212 1-year-old WIC-enrolled children (discussed earlier in Chapter Two). Bivariate analyses showed that there was no statistically significant difference in the prevalence of ECC between children who had their teeth brushed daily and those who had not (P-value=0.26).

It is important to mention that, in our study, toothbrushing (yes/no) at baseline was omitted from the analyses in spite of its statistically significant relationship with the incidence of ECC in the bivariate analyses, because it was collinear with daily frequency of toothbrushing (P-value<0.001).

The significant relationship between having a history of a previous visit to a dentist and the increased incidence of ECC might be due to self-identified need to see a dentist and/or the referrals to local dentists that have been made by the UAB team when diagnosing children with cavitated lesions. This was one of the benefits to the families of participation in current study, in addition to semiannual fluoride varnish application, oral hygiene instructions and dental kits. Also, visiting a dentist might inflate the number of tooth surfaces with dental caries experience, due to the use of radiographs and involving more surfaces in the treatment according to the dentist's clinical judgment.

Two-way interaction was assessed and there were no statistically significant interactions at $\alpha=0.10$.

Findings from Model 4

At α=0.10, the presence of a regular dentist was statistically significantly associated with increased incidence of ECC from the 1st follow-up to the 2nd follow-up (count), with P-value of 0.04, without adjusting for age. However, after adjusting for age, both presence of a regular dentist and self-brushing were statistically significantly associated with ECC incidence (count) from the 1st follow-up to the 2nd follow-up, with P-values of 0.03 and 0.09, respectively. The significant relationship between presence of a regular dentist and the increased incidence of ECC might be due to the referrals to local dentists that have been done by the UAB team when diagnosing children with cavitated lesions. However, the statistically significant relationship between self-brushing and reduced incidence of ECC is counterintuitive. Kumar et al.[125], in his study “Fluoride in dental public health programs”, mentioned that more than 70 randomized controlled trials showed decreased incidence of dental caries in children when toothbrushing with fluoride toothpaste was supervised. However, the findings of these randomized controlled trials might be due to the compliance of the children to brush their teeth because it was a group activity, rather than perfect brushing of their teeth. Kumar et al.[125] stated that a two-year randomized controlled trial in Scotland showed a 32% reduction in caries increment when five-year-old children brushed their teeth under supervision (P-value not mentioned), while on other study in England showed that supervised toothbrushing was associated with an 11% decrease in two-year caries increment among children aged five to six years (P-value not mentioned). Our finding regarding self-brushing might be due to the fact that the question which asked about self-brushing did not provide a range of options (e.g., exclusively self-brushing, exclusively supervised toothbrushing, sometimes self-brushing, most of the time self-brushing); instead the option was dichotomous (yes/no). So, if a child self-brushed his/her teeth occasionally, even if the parents also brushed the teeth after the child, the parents might have chosen yes for answering this question.

If a significance level of α=0.05 had been used, self-brushing would not have been statistically significantly associated with the incidence of ECC from 1st follow-up to the 2nd follow-up (count) after adjusting for age. Thus, in the final model, we had only the presence of a regular dentist at the 2nd follow-up which was statistically significantly associated with the incidence of ECC from the 1st follow-up to the 2nd follow-up (count) both before and after adjustment for age.

The two-way interaction was assessed and there was no statistically significant interaction at α=0.10.

Limitations

Recruitment and Generalizability

This study has several limitations. The most significant limitation was the small sample size. Ninety-seven children were recruited at baseline and 86, 84 and 73 remained in the study at the 1st, 2nd and 3rd follow-up, respectively. Also, since our sample was not a random sample, the children and their families who participated in our study might not represent the actual population of Perry County, Alabama. Instead, the study participants were recruited by the word of mouth only (convenience sample), so this might result in selection bias. Thus, we need to be careful when interpreting the findings of this study, as it might not be generalizable to other African-American children. Also, these findings might not be generalizable to the African-American children who live in Perry County, Alabama, itself, since the children were not selected randomly. Generally speaking, study subjects who volunteered to participate in a study might not represent the actual population, as there might be factors that encouraged them to participate and remain in the study. Also, since our sample was not probability sample, conducting statistical tests were not appropriate. However, since most readers and journals look for such statistical tests, and for the learning value in this thesis, it was decided they should be conducted anyway.

Limited sample size decreased the study's power to detect differences in the incidence and dmfs counts of ECC among children with different dietary and oral hygiene behaviors. Thus, we decided to use α of 0.10 as the significance level. One of the reasons for the limited sample size was that our study was secondary data analyses for an ongoing longitudinal study which was designed originally to understand the natural history of the acquisition and colonization of MS bacteria in plaque, on the tongue, and in saliva, and to assess the relationship between salivary IgA, which is induced by the presence of MS, and the incidence of dental caries. In addition, the purpose of the original study was to "establish the number and stability of MS genotype" longitudinally in the study subjects, and to determine the similarity in the genotypes of MS among the caregivers and their associations with caries incidence.

Furthermore, our study might have underestimated the prevalence and the incidence of ECC among the index children, because caries was reported at the cavitated level only (non-cavitated lesions were not reported) and due to the lack of use of radiographs, which is an important diagnostic method for interproximal caries.

Questionnaires

Information about the children's dietary and oral hygiene practices was collected by asking the caregivers to complete questionnaires at visits numbers 10, 40, 50, 60, 70 and 90, when the children were mean ages of 0.9, 1.5, 1.9, 2.5, 3.1 and 4.1 years, respectively. However, there were some limitations in the questionnaires. The parents were responsible for completing the questionnaires on their own, so they interpreted the questions according to their own understanding and interpretation. However, there were some coordinators who helped the parents in case they had specific questions. Specifically, the coordinator did not go through the questions one by one with the parents. Keeping in mind that some of the questions could be interpreted in more than one way, the validity of the responses might be questionable. For example, the questionnaires asked about the consumption of different beverages, without providing specific definitions of these beverages. For example, there was a question about juice consumption, without specifying whether it was a 100% natural juice, sugar-added juice drinks, etc. Nevertheless, there were questions about the brand names on which we relied to make categories of different types of beverages. Also, there was a question about soft drinks, without defining them. It might be confusing whether to consider lemonade, Hi-C™, or Hawaiian Punch™ as a juice drink or soft drink. Although these mentioned types of beverages should be considered as soft drinks, parents might include them under the category of “juice”[126]. Thus in our analyses, we regrouped beverages into five groups: 1) milk; 2) infant formula; 3) water; 4) 100% juice, such as pure tomato juice, orange juice, etc.; and 5) sugar-added beverages, which included all the sugar-added soft drinks (not diet beverages), such as Coke™ and other carbonated beverages, lemonade, Hi-C™, Hawaiian Punch™, Caprisun™, Koolaid™, Gatorade™, Vitamin Water™, Red Bull™, or any other drinks with added sugar. Also, it is important to note that there was no question about infant formula in the original questionnaire. Instead, there was a question about the brand name of the milk, and depending on the brand name, the data were categorized as either infant formula or milk. This explains why milk consumption and infant formula consumption were mutually exclusive. However, the percentage of the children who reportedly consumed milk (36.08%) and infant formula (61.05%) did not sum to 100%, due to missing data (parents of two children out of 97 did not answer the question).

In addition, the questions about the frequencies and amounts of beverage referred to consumption referred to “during a typical day”. Different parents might have interpreted this statement differently, especially since it was not clear whether the questions were meant to ask about consumption in the previous day, week or month, and whether it was meant to be about weekdays or weekends. In addition, since the public water in Perry County is not fluoridated, the question that asked about the source of drinking water (bottled water vs. city water vs. well water) might not be meaningful. Since the city water was nonfluoridated, the fluoride content of well waters were unknown, most of the bottled water was nonfluoridated, and brand names of the bottled waters were not provided by the parents, we could not categorize the children according to the fluoride status of the water they reportedly consumed. Water fluoride status could have been an important independent variable, especially if city water was fluoridated.

Some of the options for answering specific questions were limited. For example, there was a question about whether the children brushed their own teeth and the option for answering this question was dichotomous (yes/no), while in several other studies that assessed self-brushing, there were more detailed options, such as yes, no, most of the time, sometimes (occasionally). Similarly, some offered options of child only, parent only, and both. Also, there was no specific question about fluoride status of the toothpaste the children used. Instead, there was an open-ended question about the brand name of toothpaste. Since the answers provided by the parents often provided incomplete product information with inadequate information for assessing the fluoride status, we determined that we would not be able to make valid assumptions regarding the fluoride status of the toothpastes. For example, some of the parents reported that their children used Orajel™ toothpaste. However, there are two types of Orajel™, with one fluoridated and the other nonfluoridated, so it was not possible to make valid assumptions about the fluoride status of the toothpaste. Thus, we did not include type of toothpaste in the bivariate nor the multivariable analyses. For future studies, it would be desirable to ask the parents to bring the toothpastes with them, provide all details (words) of the product, or possibly to ask directly about fluoride status of the toothpaste.

There was a question whether the child had a history of a previous visit to a dentist (ever) in the baseline and follow-up questionnaires. If the parents reported that a child had a history of a previous visit to a dentist at baseline, then they should have answered yes to this question at all the follow-up visits as well, because the question asked about a history (ever). Also, if the parents reported that a child had a history of a previous visit to a dentist at approximately age 1.5 years (visit 40), then they should have answered yes to this question at the later ages of 2, 2.5 and 3 years (visits 50, 60 and 70), but not necessarily at baseline (visit 10). However, as mentioned earlier, 19 children who reportedly had no history of a visit to a dentist (ever) at visit 70 (2nd follow-up data), had an AUC composite for a history of a visit to a dentist from visit 40 to visit 70 more than zero (it should be zero), and eight of them had AUC>=0.50. Also, only five of 73 children who reportedly had no previous visit to a dentist at visit 70 (2nd follow-up) had AUC of a previous visit to a dentist of zero. This might be due to recall bias, or misunderstanding of the question.

Analyses

All the data were analyzed using SAS 9.3 (SAS Institute Inc., Cary, NC, USA). When analyzing the data, an important observation was that the frequencies (times per day) and the quantities (cups per day) were 99.4% identical over all the time points combined (recruitment and the five follow-up questionnaires) for all the beverages and for all the study participants. Part of the reason why it was not 100% identical was that some of the parents could not estimate the exact amount (cups per day) of beverages that their kids consumed, so they left this question unanswered. Since the literature suggests that frequencies of consumption of beverages have greater impact on the demineralization and remineralization of the tooth surface, a decision was made to analyze the relationships between only the frequencies of consumption of beverages and incidence of ECC in order to prevent redundancy. One other limitation of our study was that inter- and intra-examiner reliability were not assessed, because reliability data were not available.

In our study, we were planning to use previous caries experience as a predictor for future caries incidence. However, since the prevalence of ECC among our children at baseline and 1st follow-up was very low (1.1% and 12.8%, respectively), we did not include previous caries experience in the multivariable models with the incidence of ECC from baseline to the 3rd follow-up and from the 1st follow-up to the 2nd follow-up as dependent variables.

It is important to mention that height and weight were not recorded at baseline, and very few children had their heights and weights measured at the first follow-up (31.4%). However, height and weight were systematically measured starting from the 2nd follow-up. Thus, the relationships between BMI measures and the incidence of ECC were not assessed.

Strengths

In spite of the limitations mentioned above, our study is a unique study. There are very few studies that have reported the prevalence and incidence of ECC and dmfs increments among very young, high caries risk, African-American children at the tooth-type level and surface level. Assessment of incidence of ECC is difficult because it should be done longitudinally and it is usually costly, time-consuming and subject to attrition. In our study, we lost about 11% from baseline to the 1st follow-up, 2% from the 1st follow-up to 2nd follow-up and 13% from 2nd follow-up to the 3rd follow-up, with an average of 8.6% annually. Thus, our study provided valuable information regarding the incidence, dmfs increments and dmft increments at the person level. Also, our study provided unique findings regarding the incidence per tooth type (incisors, canines, first molars and second molars). Another strength of our study was that detailed demographic, dietary and oral hygiene information was collected at the recruitment (visit 10, when the children were about 0.9 years), at approximately six-monthly intervals until age three (visits 40, 50, 60 and 70, when the children were approximately 1.5, 2.0, 2.5 and 3 years old, respectively) and at the 3rd follow-up when the children were about four years old. This detailed information at different stages of child development helped us assess the relationships between the incidence and dmfs counts of ECC and different dietary and oral hygiene behaviors reported at baseline, and using AUC composite variables from age 1.5 to three years.

Future Research

Many factors might be associated with the high prevalence and incidence of ECC among our cohort of African-American children, such as living in a non-fluoridated county, low socioeconomic status and poor dietary behaviors. Thus, additional studies evaluating risk factors for caries development are necessary to better understand the disease process and determine foci for prevention of caries. Also, we can do further detailed analyses of caries increments per surface type and jaw (maxillary vs. mandibular). This will help us know which tooth surfaces are more vulnerable to dental decay in this subgroup. Since the preventive approach for occlusal caries is different from that for proximal caries, knowing the incidence per tooth surface type will help us plan for the best preventive intervention modality or combined modalities to decrease the prevalence and incidence of ECC among those children. In our study, we have conducted detailed analyses of the prevalence of ECC by tooth surface for 1st molars and 2nd molars. The results showed that caries experience on the occlusal surfaces of 1st molars increased from 2.99% to 10.12% to 15.42% at 1st follow-up, 2nd follow-up and 3rd follow-up, respectively. Also, caries experience on the occlusal surfaces of 2nd molars increased from 1.46% to 8.59% to 29.11% at 1st follow-up, 2nd follow-up and 3rd follow-up, respectively. The prevalence of occlusal caries experience in our study was higher than that on other individual tooth surfaces (buccal, distal, mesial and lingual) for both 1st molars and 2nd molars and at all the time points.

Survival analyses can be conducted to assess the age at which the children develop their first decayed tooth. Also, incidence density can be used in order to consider the time the teeth were at risk (the denominator=person-time instead of number of persons). For the logistic and negative binomial analyses, multivariate analyses of the one-year incidences (baseline to 1st follow-up, 1st follow-up to 2nd follow-up and 2nd follow-up to 3rd follow-up) can be conducted to provide a composite dependent variable that assesses the periods individually in a combined analysis.

Conclusion

Both the prevalence and incidence of Early Childhood Caries (ECC) and the proportions of tooth surfaces with caries experience increased with age during the three-year follow-up. The increases in the prevalence and incidence over time are due partly to both increased exposure to risk factors and increased number of teeth at risk. Also, greater prevalence and incidence of dental caries among the children in Perry County, Alabama, might be due to living in a non-fluoridated community, eating an extremely sugary and cariogenic diet, and following unhealthy dietary practices and improper oral hygiene practices. Although all of these children received fluoride varnish treatments at six-month intervals, substantial percentages of them developed additional dental caries experience during the three-year follow-up. Broadly speaking, our study had higher prevalence than that reported in most other studies and the prevalence of ECC in our study was approximately 2.0%, 18.4%, and 31.4% more (absolute difference) than the national mean prevalence among a representative sample of all American children (all races and ethnic groups) aged two, three and four years of age, respectively, reported by NHANES 1999-2002.

Also, the incidence of ECC, dmft and dmfs increments reported in this study during different time periods were higher than reported in most other studies. Many factors could be associated with the high prevalence and incidence of ECC among our cohort of African-American children, such as living in a non-fluoridated county, low socioeconomic status and poor dietary behaviors. Thus, additional studies evaluating risk factors for caries development are necessary to better understand the disease process and determine foci for prevention.

The study showed that both increased frequency of toothbrushing and increase AUC composite of daily frequency of consumption of 100% juice were associated with decreased incidence of dental caries both before and after adjustment for age. However, increased AUC composite of daily frequency of consumption of sweetened food was associated with increased incidence of dental caries.

CHAPTER 6

CONCLUSIONS

Our study is considered unique, as it assessed the prevalence and the incidence of Early Childhood Caries (ECC) among very young African-American children, who were one year old at baseline (n=97) and followed for three years. Although the children were living in a nonfluoridated community in Perry County, Alabama, the children received fluoride varnish applications semiannually from the beginning of this ongoing study, in addition to oral hygiene instructions and dental kits (toothbrushes, fluoride toothpastes, dental floss, etc.) at each visit. The analyses of our study also were relatively unique, since we determined the prevalence and the incidence of ECC at the tooth and surface levels and by tooth type (incisors, canines, 1st molars and 2nd molars) and surface type (occlusal, buccal, mesial, lingual and distal). The results of our study showed that both the prevalence and the percentages of tooth surfaces with caries experience increases with time. The prevalence of ECC increased from 1.1% at baseline (mean age was approximately one year) to 12.8%, 39.3% and 65.8% at the 1st follow-up, 2nd follow-up and 3rd follow-up, when the children were approximately two, three and four years old, respectively. Also, the percentages of surfaces with caries experience increased from 0.1% at baseline to 1.4%, 6.2% and 10.4% at the 1st follow-up, 2nd follow-up and 3rd follow-up, respectively. The prevalence and the percentages of tooth surfaces with dental caries experience were higher than those reported in most national surveys and individual studies.

Our study reported the incidence of dental caries during six different time periods, both when unerupted surfaces at the beginning of the incidence period were included, and also when unerupted surfaces at the beginning of the incidence period were excluded: 1) age one to two years; 2) age one to three years; 3) age one to four years; 4) age two to three years; 5) age two to four years; and 6) age three to four years. The results showed that 72 children had all four dental examinations, and when the unerupted surfaces at baseline were included, about 66% of the children developed at least one carious or filled surface (maximum 72 carious or filled surfaces) during the three-year follow-up. However, when only surfaces erupted at baseline were included, about 33% of the children developed at least one carious or filled surface (maximum 38 carious or filled surfaces) during the three-year follow-up. Also, crude caries incidence rates were almost identical to net caries incidence rates, due to presence of a very few reversals.

In addition, our study assessed the incidence of dental caries by tooth type. The results showed that, when unerupted surfaces at baseline were included, there were higher percentages of children who developed new caries during the three year follow-up on the 2nd molars (49%), followed by incisors (36%), 1st molars (27%) and canines (10%). However, when only erupted surfaces at baseline were included, there were higher percentages of children with new caries during the three-year follow-up on their incisors (30%), followed by 1st molars (26%) and canines (17%). Since there was only one child with all his 2nd molars erupted at baseline and this child was lost at the 3rd follow-up, the denominator of the incidence rate at the person level from baseline to the third follow-up for the 2nd molars was zero, so the incidence rate at the person level was undetermined for the 2nd molars from baseline to the 3rd follow-up.

The incidence reported in our study was higher than that reported in most published studies. Also, the incidence of dental caries in our cohort who received semiannual fluoride application was higher than the incidence of dental caries among children who did not receive fluoride treatment at all in other studies.

The high incidence and prevalence of dental caries were reported in spite of the high percentage of children who reportedly brushed their teeth with toothpaste. The study showed that 41.5% and 33.3% of the children in our cohort had their teeth brushed (either self-brushing or by caregivers) and used toothpaste, respectively, at baseline when the children were approximately one year old. However, we could not tell the fluoride status of toothpastes used by the children in our cohort, due to lack of accurate information, as mentioned earlier in the discussion. Also, all of the children (100%) reportedly brushed their teeth and used toothpaste at the 2nd follow-up when the children were three years old. The study showed that increased frequency of toothbrushing was associated with decreased incidence of dental caries (OR=0.37 and P-value=0.049).

The AUC composite of daily frequency of consumption of 100% juices was associated with decreased incidence of ECC. The protective effect of consumption of 100% juice against dental caries might be due to the statistically significantly negative correlation between the consumption of 100% juices and sugar-added beverages (Pearson correlation=-0.60 with P-value <0.0001). Also, children who reportedly consumed more 100% juice might be from higher SES families compared to those who did not consume 100% juices or consumed less. However, we could not adjust for SES or the educational status of the families, due to lack of information.

In summary, the increases in the prevalence and incidence over time with increasing age were due partly to both increased exposure to risk factors and increased number of teeth at risk. Furthermore, after assessing the relationships between dental caries incidence and different demographic risk indicators and dietary and dental risk factors, increased AUC composite of daily frequency of consumption of sweetened foods was statistically significantly associated with increased three-year incidence of dental caries (dichotomous) among the children in our cohort (P-value=0.002 and OR=9.2). It is important to mention that consumption of sugar-added beverages and sweets was not statistically significantly associated with dental caries.

We recommend additional studies be conducted to evaluate risk factors associated with dental caries development in order to define the best approaches for its prevention.

REFERENCES

1. Dye BA, Nowjack-Raymer R, Barker LK, Nunn JH, Steele JG, Tan S, et al. Overview and quality assurance for the oral health component of the National Health and Nutrition Examination Survey (NHANES), 2003–2004. J Public Health Dent. 2008;68:218–26.

2. Beltran-Aguilar ED, Barker LK, Canto MT, Dye BA, Gooch BF, Griffin SO et al. Surveillance for dental caries, dental sealants, tooth retention, edentulism, and enamel fluorosis – United States, 1988–1994 and 1999–2002. MMWR Surveill Summ. 2005;54:1–43.

3. Dye BA, Tan S, Smith V, Lewis BG, Barker LK, Thornton-Evans G, et al. Trends in oral health status—United States, 1988–1994 and 1999–2004. [cited 2010 Jun 1];Vital Health Stat 11. 2007(248) Also available from: URL: http://www.cdc.gov/nchs/data/series/sr_11/sr11_248.pdf.

4. Drury TF, Horowitz AM, Ismail AI, Maertens MP, Rozier RG, Selwitz RH. Diagnosing and reporting early childhood caries for research purposes: A report of a workshop sponsored by the National Institute of Dental and Craniofacial Research, the Health Resources and Services Administration, and the Health Care Financing Administration. J Public Health Dent. 1999;59:192-7.

5. Milnes AR. Description and epidemiology of nursing caries. J Public Health Dent. 1996;56:38–50.

6. Burt BA, Eklund SA. Dentistry, Dental Practice, and the Community, 6th edition. St. Louis, Missouri: Westline Industrial Drive, 2005. Print.

7. Raper AF. A tale of two black belt counties. Chapel Hill: University of North Carolina Press, 1936.

8. Institute of Rural Health Research. http://irhr.ua.edu/blackbelt. Web. February 11, 2012.

9. Drury TF, Horowitz AM, Ismail AI, Maertens MP, Rozier RG, Selwitz RH. Diagnosing and reporting early childhood caries for research purposes: A report of a workshop sponsored by the National Institute of Dental and Craniofacial Research, the Health Resources and Services Administration, and the Health Care Financing Administration. J Public Health Dent. 1999;59:192-7.

10. American Academy of Pediatric Dentistry. Policy Early childhood Caries (ECC): Classifications, consequences, and Preventive Strategies. Pediatr Dent 2008;30 (suppl):83.

11. Ismail AI. Clinical diagnosis of precavitated carious lesions. Community Dent Oral Epidemiol. 1997;2513-23.

12. Warren JJ, Levy S, Kanellis MJ. Dental caries in the primary dentition: Assessing prevalence of cavitated and non-cavitated lesions. J Public Health Dent. 2002;62:109–114.

13. Zafar S., Harnekar SY, Siddiqi A.Early childhood caries: etiology, clinical considerations, consequences and management. http://www.moderndentistrymedia.com/july_aug2009/zafar.pdf. Accessed February 20, 2012.

14. Dye BA, Nowjack-Raymer R, Barker LK, Nunn JH, Steele JG, Tan S, et al. Overview and quality assurance for the oral health component of the National Health and Nutrition Examination Survey (NHANES), 2003–2004. J Public Health Dent. 2008;68:218–26.

15. Kaste LM, Drury TF, Horowitz AM, Beltran E. An evaluation of NHANES III estimate of Early Childhood Caries. J Public Health Dent. 1999;59(3):198-200.

16. Currier GF, Glinka MP. The prevalence of nursing bottle caries or baby bottle syndrome in an inner-city fluoridated community. Virginia Dent J. 1977;54:9-19.

17. Lee GH, McGrath C, Yiu CK, King NM. A comparison of a generic and oral health–specific measure in assessing the impact of early childhood caries on quality of life. Community Dent Oral Epidemol. 2010; 38: 333–339.

18. Chronic disease prevention and health promotion. http://www.cdc.gov/chronicdisease/resources/publications/AAG/doh.htm. Web. Accessed July 10, 2012.

19. Fletcher RH, Fletcher SW. Clinical epidemiology: The essentials. Baltimore, MD: Lippincott Williams & Wilkins, 2005. Print.

20. Milnes AR. Description and epidemiology of nursing caries. J Public Health Dent. 1996,56:38–50.

21. Goose DH. Infant feeding and caries of the incisors: an epidemiological approach. Caries Res. 1967;1:167-73.

22. Goose DH, Gittus E. Infant feeding methods and dental caries. Public Health. 1968;83:72-6.

23. Winter GB, Hamilton MC, James PMC. The role of the comforter as an etiological factor in rampant caries of the deciduous teeth. Arch Dis Child. 1966;41: 207-12.

24. Winter GB, Rule DC, Mailer GP, James PMC, Gordon PH. The prevalence of dental caries in preschool children aged 1 to 4 years. Br Dent J. 197l;130:271-7.

25. Silver DH. The prevalence of dental caries in 3-year-old children. Some social differences and a method of assessing preschool needs at a local level. Br Dent J. 1974;137:123-8.

26. Silver DH. Improvements in the dental health of 3-year-old Herffordshire children after 8 years. Br Dent J. 1982;153:179-83.

27. Holt RD, Joels D, Winter GB. Caries in preschool children: the Camden study. Br Dent J. 1982;153:107-9.

28. Holt RD, Joels D, Bulman J, Maddick IH. A third study of caries in preschool-aged children in Camden. Br Dent J. 1988;165:87-91.

29. Silver DH. A comparison of 3-year-olds caries experience in 1973, 1981 and 1989 in a Herffordshire town,related to family behaviour and social class. Br Dent J. 1992;172:191-7.

30. Wendt LK, Hallonsten AL, Koch G. Dental caries in one- and two-year-old children living in Sweden. Part I-a longitudinal Study. Swed Dent J. 1991;15 1-6.

31. Wendt LK, Hallonsten AL, Koch G. Oral health in preschool children living in Sweden. Part II-a longitudinal study. Findings at three years of age. Swed Dent J. 1992;16:41-9.

32. Paunio P, Rautava P, Helenius H, Alanen P, Sillanpaa M. The Finnish family competence study: the relationship between caries, dental health habits and general health in 3-year-old Finnish children. Caries Res. 1993;27:154-60**.**

33. Aldy D, Siregar Z, Siregar H, Liwijaya *SG*, Tanyati S. A comparative study of caries formation in breast-fed and bottlefed children. Paediatr Indones. 1979;19:308-12

34. Brown JP, Junner C, Liew V. A study of *Streptococcus mutans* levels in both infants with bottle caries and their mothers. Aust Dent J. 1985; 30:96-98.

35. Deyu H, Dawei L. Trends in caries prevalence\ and experience in children in Qlengdu City, West China, 1982-1990. Community Dent Oral Epidemiol. 1992;20:308-9.

36. Wei *SHY*, Holm AK, Tong LSM, Yuen SWH. Dental caries prevalence and related factors in 5-year-old children in Hong Kong. Pediatr Dent. 1993;15:116-19.

37. Cleaton-Jones P, Richardson BD, McInnes PM, Fatti LP. Dental caries in South African white children aged 1-5 years. Community Dent Oral Epidemiol. 1978;6:78-81.

38. Salako NO. Infant feeding profile and dental caries status in urban Nigerian children. Acta Odontol Paediatr. 1985;6:13-7.

39. Kerosuo H, Honkala E. Caries experience in the primary dentition of Tanzanian and Finnish 3-7-year-old children. Community Dent Oral Epidemiol. 1991; 19:272-6.

40. Raadal M, Elkhider EF, Rasmussen P. The prevalence of canes in groups of children aged 4.5 and 7-8 years in Khartoum, Sudan. Int J Pediatr Dent. 1993;3:9-15

41. Soparkar P, Tavares M, Husain J. Nursing bottle syndrome in Kuwait. J Dent Res. 1986;65:74-5.

42. Yagot K, Nazhat NY, Kuder SA. Prolonged nursing-habit *caries* index. J Int Assoc Dent Child. 1990;20:8-10.

43. Powell D. Milk: is it related to rampant caries of the early primary dentition? J Calif Dent Assoc. 1976;4:58-63.

44. Currier GF, Glinka MP. The prevalence of nursing bottle caries or baby bottle syndrome in an inner-city fluoridated community. Virginia Dent J. 1977;54:9-19.

45. Johnsen DC, Schultz DW, Schubot DB, Easley MW. Caries patterns in Head start children in a fluoridated community. J Public Health Dent. 1984;44:61-6.

46. Kelly M, Bruerd B. The prevalence of baby bottle tooth decay among two Native American populations. J Public Health Dent. 1987;47: 94-7.

47. Broderick E, Mabry J, Robertson D, Thompson J. Baby bottle tooth decay in Native American children in Head Start centers. Public Health Rep. 1989;104:50-4.

48. Barnes GP, Parker WA, Lyon TC, Drum MA, Coleman GC. Ethnicity, location, age and fluoridation factors in baby bottle tooth decay and caries prevalence of Head Start children. Public Health Rep. 1992;107:167-73.

49. Derkson GD, Ponti P. Nursing bottle syndrome; prevalence and etiology in a nonfluoridated city. J Can Dent Assoc. 1982;48:389-93.

50. Budowski L. Prevalence of nursing caries in a multi-ethnic, racial and cultural population. Dissertation, University of Toronto, 1989.

51. Albert RJ, Cantin RY, Cross HG, Castaldi *CR.* Nursing caries in the Inuit children. J Can Dent Assoc. 1988;54:751-8.

52. Katz L, Rip LW, Petersen M. Nursing caries in Head Start children, St. Thomas, US Virgin Islands: assessed by examiners with different dental backgrounds. J Clin Pediatr Dent. 1992;16:124-30.

53. Vignarajah S, Williams GA. Prevalence of dental caries and enamel defects in the primary dentition of Antiguan preschool children aged 34 years including an assessment of their habits. Community Dent Health. 1992;9:349-60.

54. Tsai AI, Chen C-Y, Li L-A, Hsiang C-L, Hsu K-H. Risk indicators for ECC in Taiwan. Community Dental Oral Epidemol. 2006;34: 437-445.

55. World Health Organization. Oral health surveys: basic methods, 3rd ed. Geneva: WHO; 1987.

56. Levy SM, Kiritsy MC, Slager SL, Warren JJ. Pattern of dietary fluoride supplement use during infancy. J Public Health Dent. 1998;58:228-33.

57. Levy SM, Kiritsy MC, Slager SL, Warren JJ, Kohout FJ. Pattems of fluoride dentifrice use among infants. Pediatr Dent. 1997;19:50-5.

58. Heilman JR, Kiritsy MC, Levy SM, Wefel JS. Fluoride content of infant foods and cereals. J Am Dent Assoc. 1997;128:857-63.

59. Warren JJ, Kanellis MJ, Levy SM. Fluorosis of the primary dentition: what does it mean for permanent teeth? J Am Dent Assoc. 1999;130:347-56.

60. Kolker JL, Yuan Y, Burt BA, Sandretto AM, Sohn W, Lang SW, Ismail AI. Dental caries and dietary patterns in low-income African-American children. Paediatr Dent. 2007;29:257-64.

61. Pitts N. ICDAS: An international system for caries detection and assessment being developed to facilitate caries epidemiology, research and appropriate clinical management. Community Dent Health. 2004;21:193-8.

62. Mitsugi O, Yoshiko S, Fumiko H, Takako D, Junji S, Kazuo M, Katsuyuki K. Longitudinal study of dental caries incidence associated with Streptococcus mutans and Streptococcus sobrinus in pre-school children. J Med Microbiol. 2005;54:661–665.

63. Ismail AI, Sohn W, Tellez M, Amaya A, Sen A, Hasson H, Pitts NB. (2007), The International Caries Detection and Assessment System (ICDAS): An integrated system for measuring dental caries. Community Dent Oral Epidemol. 2007;35: 170–178.

64. Grindefjord M, Dahllöf M, Modéer T. Caries development in children from 2.5 to 3.5 years of age: a longitudinal study. Caries Res. 1995;29:449–454.

65. Karjalainen S, Soderling E, Sewon L, Lapinleimu H, Simell O: A prospective study on sucrose consumption, visible plaque and caries in children from 3 to 6 years of age. Community Dent Oral Epidemol. 2001;29:136-42.

66. Sakuma S, Nakamura M, Miyazaki H. Predictors of dental caries development in 1.5-year-old high-risk children in the Japanese public health service. J Public Health Dent. 2007;67: 14–19.

67. Warren JJ, Weber-Gasparoni K, Marshall TA, Drake DR, Dehkordi-Vakil F, Dawson DV, Tharp KM. A longitudinal study of dental caries risk among very young low SES children. Community Dent Oral Epidemiol. 2009;37: 116–122.

68. Litt MD, Reisine S, Tinanoff N. Multidimensional causal model of dental caries development. Public Health Rep. 1995;110:607-607.

69. Radike A W: Criteria for diagnosis of dental caries. In Proceedings of the conference on clinical testing of cariostatic agents, 1968. American Dental Association, Chicago, IL, 1972, pp.92-95.

70. O'Sullivan DM, Tinanoff N. The association of early dental caries patterns with caries incidence in preschool children. J Public Health Dent. 1996;56: 81–83.

71. O'Sullivan DM, Tinanoff N. Maxillary anterior caries associated with increased caries risk in other primary teeth. J Dent Res. 1993;72:1577-80.

72. Scalvos S, Porter S, Seow WK. Future caries development in children with nursing bottle caries. J Pedodontics. 1988;13:1-10.

73. Weintraub JA, Ramos-Gomez F, Jue B. Fluoride varnish efficacy in preventing early childhood caries. J Dent Res. 2006;85(2):172–176.

74. Grodzka K, Augustyniak L, Budny J. Caries increment in primary teeth after application of Duraphat fluoride varnish. Community Dent Oral Epidemiol. 1982;10:55-9.

75. Plonka KA, Pukallus ML, Barnett A, Holocombe TF, Walsh LJ, Seow WK. A controlled, longitudinal study of home visits compared to telephone contacts to prevent early childhood caries. Int J Paediatr Dent. 2011;10:1365-85

76. Isokangas P, Soderling E, Pienihakkinen K, Alanen P. Occurrence of dental decay in children after maternal consumption of xylitol chewing gum, a follow-up from 0 to 5 years of age. J Dent Res. 2000;79: 1885-1889.

77. Ismail AI, Hasson H, Sohn W. Dental caries in the second millennium. http://www.nidcr.nih.gov/NR/rdonlyres/5A204F1A-AAF6-4DF8-BAB8-2E18033944D6/0/Amid_Ismail.pdf. Web. Accessed August 3, 2012.

78. Dental Care Matters. http://dentalcarematters.com/tooth-decay. Web. Accessed April 23, 2012.

79. Nair R, Weber-Gasparoni K, Marshall TA, Warren JJ, Levy SM. Factors affecting ECC among WIC-enrolled children in Linn County, Iowa. J Dent Child. 2010;77:158-65.

80. Ramos-Gomez FJ, Weintraub JA, Gansky SA, Hoover CI, Featherstone John D.B., Bacterial, behavioral and environmental factors associated with early childhood caries. J Clin Pediatric Dent. 2002;26:165–73.

81. Karjalainen S, Sewo´n L, Söderling E, Lapinleimu H, Seppänen R, Simell O. Oral health of 3-year-old children and their parents after 29 months of child-focused antiatherosclerotic dietary intervention in a prospective randomized trial. Caries Res. 1997;31:180–5.

82. Edelstein BL, Chinn CH. Update on disparities in oral health and access to dental care for America’s children. Academic Pediatr. 2009;9:415–9.

83. Hallett KB and O’Rourke PK. Pattern and severity of Early Childhood Caries. Community Dent Oral Epidemiol. 2006;34:25–35.

84. Vanchirarojpisan T, Shinada K, Kawaguchi Y, Laungwecchakan P, Somkote T, Detsomboonrat P. Early Childhood Caries in children aged 6-19 months. Community Dent Oral Epidemol. 2004;32:133-42.

85. Dye BA, Arevalo O, Vargas CM. Trends in paediatric dental caries by poverty status in the United States, 1988–1994 and 1999–2004. Int J. Paediatr Dent. 2010;20:132–143.

86. Warren JJ, Weber-Gasparoni K, Marshall TA, Drake DR, Dehkordi-Vakil F, Kolker JL, Dawson DV, Factors associated with dental caries experience in 1-year-old children. J Public Health Dent. 2008,68:70–75.

87. Psoter WJ, Pendrys DG, Morse DE, Zhang H, Mayne ST. Associations of ethnicity/race and socioeconomic status with Early Childhood Caries patterns. J Public Health Dent. 2006;66:23–29.

88. Kumarihamy SL, Subasinghe LD, Jayasekara P, Kularatna SM, Palipana PD: The prevalence of Early Childhood Caries in 1–2 year olds children in a semi-urban area of Sri Lanka. BMC Res Notes. 2011;4:336.

89. Rugg-Gunn AJ, Hackett AF, Appleton DR, Jenkins GN, Eastoe JE. Relationship between dietary habits and caries increment assessed over two years in 405 English adolescent school children. Arch Oral Biology J. 1984;29:983–92.

90. Burt BA, Szpunar SM. The Michigan study: the relationship between sugars intake and dental caries over three years. International Dent Journal. 1994;44:230–40.

91. Gibson S, Williams S. Dental caries in pre-school children: Associations with social class, toothbrushing habit and consumption of sugars and sugar-containing foods. Caries Res. 1999;33:101–13.

92. Persson LÅ, Holm AK, Arvidsson S, Samuelson G. Infant feeding and dental caries – a longitudinal study of Swedish children. Swed Dent J. 1985;9:201–6.

93. Freeman R, Breistein B, McQueen A, Stewart M. The dental health status of five-year-old children in North and West Belfast. Community Dent Health 1997; 14:253–7.

94. Weber KA. Comparison of the effectiveness of three educational interventions in the prevention of "Early Childhood Caries." Iowa City (Doctoral Dissertation). 2003.

95. Seow WK, Clifford H, Battistutta D, Morawska A, Holcombe T. Case-control study of Early Childhood Caries in Australia. Caries Res. 2009;43:25-35.

96. Kaste LM, Gift HC. Inappropriate infant bottle feeding—status of the Healthy People 2000 objective. Arch Pediatr Adolesc Med. 1995;149:786-791.

97. Folayan MO, Sowole CA, Owotade FJ, Sote E. Impact of infant feeding practices on caries experience of preschool children. J clin Pediatr Dent. 2010;34(4):297-302.

98. Forsman B, Ericsson Y. Breastfeeding, formula feeding and dental health in low-fluoride district in Sweden. Community Dent Oral Epidemol. 1974;2:1-6.

99. Grönroos L. Quantitative and qualitative characterization of Mutans Streptococci in Saliva and in the Dentition. http://doria17kk.lib.helsinki.fi/bitstream/handle/10024/1484/quantita.pdf?sequence=1. Web. Accessed April 18, 2012.

100. O'Sullivan DM, Tibodeau EA. Caries experience and mutans streptococci as indicators of caries incidence. Pediatr Dent. 1996;18:371-4.

101. Alaluusua S, Renkonen OV. *Streptococcus mutans* establishment and dental caries experience in children from 2 to 4 years old. Scand Journal Dental Research. 1983;91:453–7.

102. Laitala M, Alanen P, Isokangas P, Söderling E, Pienihäkkinen K. A cohort study on association of early Mutans Streptococci colonization and dental decay. J Caries Res. 2012;46:228-233.

103. UAB Grant. http://grants.nih.gov/grants/oer.htm. Web. Accessed February 20, 2012.

104. Harris NO, Garcia-Godoy F. Primary preventive dentistry, 6th ed. Pearson Education, Upper Saddle River, N.J. 2004.

105. Pinkham JR. Pediatric dentistry: Infancy through adolescence, 4th ed. Elsevier Saunders Co., Philadelphia. 2005.

106. Petrie A, Bulman JS, Osborn JF. Further statistics in dentistry Part 6: Multiple linear regression. Br Dent J. 2002;21;193:675-82.

107. Leverett DH, Adair SM, Vaughan BW, Proskin HM, Moss ME. Randomized clinical trial of the effect of prenatal fluoride supplements in preventing dental caries. Caries Res. 1997;31:174-179.

108. Tanaka K, Miyake Y. Association between breastfeeding and dental caries in Japanese children. J Epidemol. 2012;22:72-77.

109. Njoroge NW, Kemoli AM, Gatheche LW. Prevalence and pattern of early childhood caries among 3-5 year olds in Kiambaa, Kenya. East African Med J. 2010;87:134-137.

110. Cleaton Jone P, Williams S, Green C, Fatti P. Dental caries rates in primary teeth in 2002, and caries surveillance trends 1981-2002, in South African city. Comm Dent Health. 2008;25,79-83.

111. Chankanka O, Cavanaugh JE, Levy SM, Marshal T, Warren JJ, Broffitt B, Kolker JL. Longitudinal associations between children's dental caries and risk factors. J Public Health Dent. 2011;71:289-300.

112. Holm AK. Effect of fluoride varnish (Duraphat) in preschool children. Community Dent. Oral Epidemol. 1979:7:241-245.

113. Autio-Gold JA, Courts F. Assessing the effect of fluoride varnish on early enamel carious lesions in the primary dentistion. JADA 2001;132:1247-53.

114. Petersson LG, Koch G, Rasmusson CG, Stanke H. Effect on caries of different fluoride prophylactic programs in preschool children. A two year clinical study. Swd Dent J 1985; 9:97-104.

115. Wong MC, Lu HX a, LO EC. Caries increment over 2 years in preschool children: a life course approach. Int J Ped Dent. 2011;22:77-84.

116. Kilpatrick NM, Neumann A, Lucas N, Chapman J, Nicholson JM. Oral health inequalities in a national sample of Australian children aged 2-3 and 6-7 years. Australian Dent J. 2012;57:38-44.

117. Guido JA, Martinez Mier EA, Soto A, Eggertsson H, Sanders BJ, Jones JE, Weddell JA, Cruz IV, Anton de la Concha JLA. Caries prevalence and its

association with brushing habits, water availability, and the intake of sugared beveragges. Int J Pead Dent. 2011; 21:432-440.

118. Chu CH, Ho PL, Lo ECM. Oral health status and behaviors of preschool children in Hong Kong. BMC Public Health. 2012; 12:767.

119. Han E, Powell L. Consumption patterns of sugar-seetened beverages in the United States. J Academy of Nutrition and Dietetics. 2013;113 (1): 43-53.

120. Hsu NY, WU PC, Bornehag CG, Sundell J, Su HJ. Feeding bottles usage and the prevalence of childhood allergy and asthma. Clinical and Development immunology. 2012;113(1):43-53.

121. Marshall TA, Levy SM, Broffitt B, Warren JJ, Eichenberger-Gilmore JM, Burns TL, Stumbo PJ. Dental caries and beverage consumption in young children. Pediatrics. 2003;112(3 Pt 1):e184-91.

122. Rolla G, Scheie AA, Ciardi JE. Role of sucrose in plaque formation. Scand J Dent Res. 1985;93:105–111

123. Rolla G. Why is sucrose so cariogenic? The role of glucosyltransferase and polysaccharides. Scand J Dent Res. 1989;97:115–119

124. Petti S, Pezzi R. Effect of sucrose consumption on level of *Streptococcus mutans* in saliva. New Microbiol. 1996;19:133–140

125. Kumar JV, Moss ME. Fluorides in dental public health programs. Dent Clin N Am. 2008;52:387-401.

126. Wikipedia. Soft drinks. http://en.wikipedia.org/wiki/Soft_drink. Accessed May 2, 2013.

APPENDIX

ORAL HEALTH RISK ASSESSMENT FORM (AGE 1 BASELINE)

CHILD'S NAME: ____________________DATE OF BIRTH:_______________
Sex: Male ☐ Female ☐

Child's Medical History

Birth:

1. Did the birthmother experience any of the following problems during pregnancy?
   ☐ Gestational Diabetes ☐ Pregnancy Induced Hypertension ☐ Group B Strep
   ☐ Amnionic Fluid Complications ☐ Placental Complications ☐ other
   If other, please explain:

____

2. Was the child delivered full term (37 to 42 weeks)? ☐ Yes ☐ No
   If no, week of delivery: ☐☐ Weeks

3. Was the child's birth: ☐standard ☐ C-Section ☐ Forcep ☐ other?
   If other, please specify what it was:

____

4. What was your child's birth weight? ☐☐ pounds ☐☐ ounces

Health:

5. Does your child have any allergies or experienced an adverse reaction to anything (drugs, food, latex, etc.)? ☐ Yes ☐ No
   If yes, please explain. ____

6. Has your child ever had any of the following medical conditions?

	YES	NO		YES	NO		YES	NO
Heart	( )	( )	Heart Murmur	( )	( )	Hepatits	( )	( )
Rheumatic Fever	( )	( )	Kidney	( )	( )	Epilepsy	( )	( )
Blood dyscrasia	( )	( )	Endocrine	( )	( )	Immune Deficiencies	( )	( )
Diabetes	( )	( )	Cancer	( )	( )	HIV positive	( )	( )
Bones	( )	( )	Birth defect	( )	( )	Other	( )	( )

If yes to any of these please explain:

____

7. Has your child had any of these illnesses in the last six months? Did he/she receive an antibiotic for the illness?

Illness:	Yes/No	Antibiotic Name
☐ Ear Infection	☐ / ☐	____________________
☐ Sinus Infection	☐ / ☐	____________________
☐ Sore Throat	☐ / ☐	____________________
☐ Skin Infection	☐ / ☐	____________________
☐ Urinary Tract Infection	☐ / ☐	____________________
☐ Chest Cold	☐ / ☐	____________________
☐ Other____________________	☐ / ☐	____________________

8. How many weeks has your child taken antibiotics in the past 6 months:
☐ None ☐ 1-2 wks ☐ 3-6 wks ☐ 7-12 wks
☐ More than 12wk

9. How long ago did your child complete their last antibiotic prescription? ________

☐ days ☐ weeks ☐ months

a. How many days did your child take antibiotics the last time? [|] days

b. What was the name of antibiotic for the last prescription?

____

c. How was the antibiotic taken? liquid ☐ pill ☐ other ☐

10. Please list all medications that your child is currently taking (or has taken in the past month).

Dose, frequency and reason concerning prescribed and over-the-counter medications should be provided:

Medication Name	Dose	Frequency Given	Reason for Use
____________________	__________	____________	____________
____________________	__________	____________	____________
____________________	__________	____________	____________

Childcare History

11. In what type of childcare environment did/does your child spend the most time during the day from birth to the present? (please check only one)

☐ with mother ☐ with father ☐ with grandparent (specifiy) ____________

☐ daycare ☐ other (specify) ____________

a. How many hours per week did/does your child spend in the above environment?

☐☐☐ hours per week

Diet and Nutrition

12. Is/was your child breastfed? ☐ Yes ☐ No

a. If yes, how old was the child when you stopped? ☐☐ Months

b. If still nursing, how many times a day? 1-2x ☐ 3-4x ☐ 5-7x ☐ 8+ ☐

13. What does your child drink during a typical day? (please check all that apply)

Beverage	Brand Name	Times Per Day	Cups Per Day
☐ breast milk	NA	☐☐	☐☐
☐ formula	____________	☐☐	☐☐
☐ milk	____________	☐☐	☐☐
☐ juice	____________	☐☐	☐☐
☐ water	____________	☐☐	☐☐
☐ other	____________	☐☐	☐☐

14. a. Which of the following best describes how your child drinks liquids other than water?

Drinks quickly ☐ Sips over time ☐

b. Which of the following best describes when your child drinks liquids other than water?

Drinks frequently throughout the day ☐ Drinks primarily at meal/snack times ☐

15. Which of the following is used by your child?

Regular cup	☐	Bottle	☐
Sippy cup	☐	Other	________________

16. Does/did your child go to sleep with something to drink? ☐ Yes ☐ No
    a. If yes, what did they drink? (please be specific)

    ____

17. Is your child on a special diet and/or have to avoid certain foods? ☐ Yes ☐ No
    a. If yes, please explain:

    ____

Oral Hygiene

18. Do you (or someone else) brush your child's teeth/gums? ☐ Yes ☐ No
    a. If someone else, who and where (for example: at daycare)?

    ____

19. Do you use a toothbrush used to clean your child's teeth? ☐ Yes ☐ No

20. How many times a day do you clean your child's teeth? ☐☐ times/day

21. Do you use toothpaste to clean your child's teeth? ☐ Yes ☐ No

    a. If yes, what brand of toothpaste is used?

    ____

Dental History

22. What is your child's source of drinking water?

    ☐ bottled water ☐ well water ☐ city water ☐ other

23. Have you given your child vitamin drops or tablets with fluoride? ☐ Yes ☐ No

24. Please check if your child has (had) any of the following dental problems:

Toothache	( )	Teeth bumped	( )	Bleeding gums	( )
Sensitive teeth	( )	Discolored teeth	( )	Other:	

____

ORAL HEALTH RISK ASSESSMENT FORM (AGE 2-4 FOLLOW-UP)

CHILD'S NAME: ______________________ DATE OF BIRTH: ______________

Sex: Male ☐ Female ☐

Child's Medical History

1. Does your child have any allergies or experienced an adverse reaction to anything (drugs, foods, latex, etc.)? ☐ Yes ☐ No

   If yes, please explain. ____

2. Has your child ever had any of the following medical conditions?

	YES	NO		YES	NO		YES	NO
Heart	( )	( )	Heart Murmur	( )	( )	Hepatitis	( )	( )
Rheumatic Fever	( )	( )	Kidney	( )	( )	Epilepsy	( )	( )
Blood dyscrasia	( )	( )	Endocrine	( )	( )	Immune Deficiencies	( )	( )
Diabetes	( )	( )	Cancer	( )	( )	HIV positive	( )	( )
Bones	( )	( )	Birth defect	( )	( )	Other	( )	( )

If yes to any of these please explain:

____

____

____

____

3. Has your child had any of these illnesses in the last six months? Did he/she receive an antibiotic for the illness?

Illness:	Yes/No	Antibiotic Name
☐ Ear Infection	☐ / ☐	____________________
☐ Sinus Infection	☐ / ☐	____________________
☐ Sore Throat	☐ / ☐	____________________
☐ Skin Infection	☐ / ☐	____________________
☐ Urinary Tract Infection	☐ / ☐	____________________
☐ Chest Cold	☐ / ☐	____________________
☐ Other________________	☐ / ☐	____________________

4. How many weeks has your child taken antibiotics in the past 6 months:

   ☐ None ☐ 1-2 wks ☐ 3-6 wks ☐ 7-12 wks ☐ More than 12wk

5. How long ago did your child complete their last antibiotic prescription? ________

☐ days ☐ weeks ☐ months

a. How many days did your child take antibiotics the last time? ☐☐ days

b. What was the name of antibiotic for the last prescription?

____

c. How was the antibiotic taken? liquid ☐ pill ☐ other ☐

6. Please list all medications that your child is currently taking (or has taken in the past month).

Medication Name	Dose	Frequency Given	Reason for Use

Childcare History

7. In what type of childcare environment did/does your child spend the **most time** during the day from birth to the present? (please check only one)

☐ with mother ☐ with father ☐ with grandparent (specifiy)

__________________ ☐ daycare ☐ other (specify) __________________________

b. How many hours per week did/does your child spend in the above environment?

☐☐☐ hours per week

Diet and Nutrition

8. Are you currently breastfeeding your child? ☐ Yes ☐ No

a. If yes, how many times a day? 1-2x ☐ 3-4x ☐ 5-7x ☐ 8+ ☐

9. What does your child drink during a typical day? (please check all that apply)

Beverage	Brand Name	Times Per Day	Cups Per Day
☐ breast milk	NA		
☐ formula			
☐ milk			
☐ juice			
☐ water			
☐ other			

10. a. Which of the following **best** describes how your child drinks liquids **other than water**?

Drinks quickly ☐ Sips over time ☐

b. Which of the following **best** describes when your child drinks liquids **other than water**?

Drinks frequently throughout the day ☐ Drinks primarily at meal/snack times ☐

11. Which of the following is used by your child?

Regular cup ☐ Bottle ☐
Sippy cup ☐ Other ________________

12. Does/did your child go to sleep with something to drink? ☐ Yes ☐ No

a. If yes, what did they drink? (please be specific)

____

13. Is your child on a special diet and/or have to avoid certain foods? ☐ Yes ☐ No

a. If yes, please explain:

____

Oral Hygiene

14. Do you (or someone else) brush your child's teeth/gums? ☐ Yes ☐ No

a. If someone else, who and where (for example: at daycare)?

____

15. Do you use a toothbrush used to clean your child's teeth? ☐ Yes ☐ No

16. How many times a day do you clean your child's teeth? ☐☐ times/day

17. Do you use toothpaste to clean your child's teeth? ☐ Yes ☐ No

    a. If yes, what brand of toothpaste is used?

____

Dental History

18. What is your child's source of drinking water?

    ☐ bottled water ☐ well water ☐ city water ☐ other

19. Have you given your child vitamin drops or tablets with fluoride? ☐ Yes ☐ No

20. Please check if your child has (had) any of the following dental problems:

    Toothache ( ) Teeth bumped ( ) Bleeding gums ( )
    Sensitive teeth ( ) Discolored teeth ( ) Other: ____________

21. Does your child have a dentist? ☐ Yes ☐ No

    c. If yes, when was his/her last visit?

    ____

    d. If yes, what was his/her last visit for? Regular Check-Up ☐ Other ☐
       If other, please explain:

____

Caretaker Signature: ________________________________

Date: _______________

Relationship to Child: ________________________________

Interviewer Signature: ________________________________

Dentist Signature: ________________________________

IRB CERTIFICATION

OMB No. 0990-0263
Approved for use through 3/31/2015

Protection of Human Subjects
Assurance Identification/IRB Certification/Declaration of Exemption
(Common Rule)

Policy: Research activities involving human subjects may not be conducted or supported by the Departments and Agencies adopting the Common Rule (56FR28003, June 18, 1991) unless the activities are exempt from or approved in accordance with the Common Rule. See section 101(b) of the Common Rule for exemptions. Institutions submitting applications or proposals for support must submit certification of appropriate Institutional Review Board (IRB) review and approval to the Department or Agency in accordance with the Common Rule.

Institutions must have an assurance of compliance that applies to the research to be conducted and should submit certification of IRB review and approval with each application or proposal unless otherwise advised by the Department or Agency.

1. Request Type	2. Type of Mechanism	3. Name of Federal Department or Agency and, if known, Application or Proposal Identification No.
[ ] ORIGINAL [X] CONTINUATION [ ] EXEMPTION	[ ] GRANT [X] CONTRACT [ ] FELLOWSHIP [ ] COOPERATIVE AGREEMENT [ ] OTHER:________	
4. Title of Application or Activity Epidemiology of Dental Caries and Immunity in Children (Alabama)		5. Name of Principal Investigator, Program Director, Fellow, or Other CHILDERS, NOEL

6. Assurance Status of this Project *(Respond to one of the following)*

[X] This Assurance, on file with Department of Health and Human Services, covers this activity:
Assurance Identification No. FWA00005960, the expiration date 01/24/2017 IRB Registration No. IRB00000726

[ ] This Assurance, on file with *(agency/dept)*________, covers this activity.
Assurance No.________, the expiration date________ IRB Registration/Identification No.________ *(if applicable)*

[ ] No assurance has been filed for this institution. This institution declares that it will provide an Assurance and Certification of IRB review and approval upon request.

[ ] Exemption Status: Human subjects are involved, but this activity qualifies for exemption under Section 101(b), paragraph________.

7. Certification of IRB Review (Respond to one of the following IF you have an Assurance on file)

[X] This activity has been reviewed and approved by the IRB in accordance with the Common Rule and any other governing regulations.
by: [X] Full IRB Review on (date of IRB meeting) 8/29/2012 or [ ] Expedited Review on (date)________
[ ] If less than one year approval, provide expiration date ________

[ ] This activity contains multiple projects, some of which have not been reviewed. The IRB has granted approval on condition that all projects covered by the Common Rule will be reviewed and approved before they are initiated and that appropriate further certification will be submitted.

8. Comments
Protocol subject to Annual continuing review.

Title F060328001
Epidemiology of Dental Caries and Immunity in Children (Alabama)

IRB Approval Issued: 09-12-12

9. The official signing below certifies that the information provided above is correct and that, as required, future reviews will be performed until study closure and certification will be provided.	10. Name and Address of Institution University of Alabama at Birmingham 701 20th Street South Birmingham, AL 35294
11. Phone No. *(with area code)* (205) 934-3789 12. Fax No. *(with area code)* (205) 934-1301 13. Email: irb@uab.edu	
14. Name of Official Ferdinand Urthaler, M.D.	15. Title Chairman, IRB
16. Signature Ferdinand Urthaler, MD/db	17. Date 09-12-12

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INFORMED CONSENT

Addendum Informed Consent Document

TITLE OF RESEARCH: Epidemiology of Dental Caries and Immunity in Children (Alabama). School and Daycare Study

IRB PROTOCOL: F060328001

INVESTIGATOR: Dr. Noel K. Childers
Dr. Rosalyn Bassett

SPONSOR: National Institute of Dental and Craniofacial Research

For Children/Minors (persons under 19 years of age) participating in this study, the term *You* addresses both the participant ("you") and the parent or legally authorized representative ("your child").

Explanation of Procedures

You are being asked to continue participating in a study to determine the factors related to colonization of teeth with a bacteria that is associated with dental caries (tooth decay). This information will be beneficial in understanding dental caries. If you decide to continue participating, you will have a dental cleaning, fluoride treatment and examination, fill out a questionnaire about caries risk, and have saliva and dental plaque samples collected for analysis of immunity and bacteria colonization levels. "Pure" saliva will be collected by having small plastic suction cups placed in your mouth (cheek) for 5-10 minutes. Dental plaque will be collected from teeth with a sterile toothpick. These samples will be sent to the University of Alabama at Birmingham for analysis. The length of time you will be involved in this study is approximately 4 years with the procedures described above repeated every 12 months.

Risks and Discomforts

The risks involved with this study are no more than minimal risk. The only mild discomfort anticipated is the use of small plastic suction cups used in your mouth to collect saliva.

Benefits

You will receive a dental cleaning and check up for each 12 month visit. If any dental disease is identified, you will be informed and we will attempt to help you find dental care. Additionally, the benefits of the study are to better understand the immunity to bacteria that cause dental caries and how immunity and other factors affect colonization of children with these bacteria when new teeth erupt.

UAB IRB
Date of Approval 09-12-12
Not Valid On 08-29-13

Participant Initials____________

Alternatives

There are no alternatives to the procedures of this study, however, you may choose not to participate.

Confidentiality

The information gathered during this study will be kept confidential to the extent permitted by law. However, your doctor, the National Institutes of Dental and Craniofacial Research, the Office for Human Research Protections (OHRP) and UAB's Institutional Review Board (IRB) will be able to inspect your medical records and have access to confidential information that identifies you by name. The results of the findings from your participation in this study, including laboratory tests may be published for scientific purposes; however, your identity will not be revealed. [UAB only] UAB and UAB Health System affiliated entities

Refusal or Withdrawal without Penalty

You are free to withdraw your consent and to discontinue participation in this project at any time without prejudice against further care that you may receive at this institution.

Cost of Participation

There will be no cost to you from participation in the research. All study related materials, dental cleaning, fluoride treatment, sample collections, and dental examinations will be provided at no cost.

Payment for Participation in Research

You will be paid $20 (or equivalent gift certificate, U.S. Savings Bond) when saliva and dental plaque samples are collected. Payments will be made at the visit when samples are collected.

Payment for Research-Related Injuries

UAB and the National Institute of Dental and Craniofacial Research have made no provision for monetary compensation in the event of injury resulting from the research, and in the event of such injury, treatment is provided, but is not provided free of charge.

Participant Initials______________

Significant New Findings

Any significant new findings that develop during the course of the study that may affect your willingness to continue in the research will be provided to you by Dr. Childers, or his staff.

Questions

If you have any questions about the research or a research related injury, Dr. Noel Childers and/or Dr. Rosalyn Bassett will be glad to answer them. Dr. Childers' phone number is 205-934-3230. Dr. Childers may also be reached after hours by paging at 205-939-9100. Dr. Bassett's phone number is 334-289-9978. If you have questions about your rights as a research participant, or concerns or complaints about the research, you may contact the Office of the Institutional Review Board for Human Use (OIRB) at (205) 934-3789 or 1-800-822-8816. If calling the toll-free number, press the option for "all other calls" or for an operator/attendant and ask for extension 4-3789. Regular hours for the Office of the IRB are 8:00 a.m. to 5:00 p.m. CT, Monday through Friday. You may also call this number in the event the research staff cannot be reached or you wish to talk to someone else.

Legal Rights

You are not waiving any of your legal rights by signing this consent form.

"INTENTIONALLY LEFT BLANK"

Participant Initials______________

Signatures

You are making a decision whether or not to have your child participate in this study. Your signature indicates that you have read (or been read) the information provided above and decided to allow your child to participate.

You will receive a copy of this signed consent form.

____

Signature of Parent Or Legally Authorized Representative Date

____

Reviewed By Principal Investigator Date

____

Signature of Individual Obtaining Consent Date

Signature of Child/Minor

_________________________(name of child/minor) has agreed to participate in research Epidemiology of Dental Caries and Immunity in Children (title of project).

____

Signature of Participant 14-18 Years of Age Date

OR

Waiver of Assent

The assent of _________________________ (name of child/minor) was waived because of:

Age ________ Maturity ________ Psychological state of the child ________

____

Signature of Parent Or Legally Authorized Representative Date

University of Alabama at Birmingham
AUTHORIZATION FOR USE/DISCLOSURE OF HEALTH INFORMATION FOR RESEARCH

What is the purpose of this form? You are being asked to sign this form so that UAB may use and release your health information for research. Participation in research is voluntary. If you choose to participate in the research, you must sign this form so that your health information may be used for the research.

Participant Name:____________________
Research Protocol: Epidemiology of Dental Caries and Immunity in Children (Alabama)

UAB IRB Protocol Number: F060328001
Principal Investigator: Noel K. Childers

Sponsor: National Institute for dental and Craniofacial Research

What health information do the researchers want to use? All medical information and personal identifiers including past, present, and future history, examinations, laboratory results, imaging studies and reports and treatments of whatever kind related to or collected for use in the research protocol.

Why do the researchers want my health information? The researchers want to use your health information as part of the research protocol listed above and described to you in the Informed Consent document.

Who will disclose, use and/or receive my health information? The physicians, nurses and staff working on the research protocol (whether at UAB or elsewhere); other operating units of UAB, HSF, UAB Highlands, The Children's Hospital of Alabama, Callahan Eye Foundation Hospital and the Jefferson County Department of Public Health, as necessary for their operations; the IRB and its staff; the sponsor of the research and its employees; and outside regulatory agencies, such as the Food and Drug Administration.

How will my health information be protected once it is given to others? Your health information that is given to the study sponsor will remain private to the extent possible, even though the study sponsor is not required to follow the federal privacy laws. However, once your information is given to other organizations that are not required to follow federal privacy laws, we cannot assure that the information will remain protected.

How long will this Authorization last? Your authorization for the uses and disclosures described in this Authorization does not have an expiration date.

Can I cancel the Authorization? You may cancel this Authorization at any time by notifying the Director of the IRB, in writing, referencing the Research Protocol and IRB Protocol Number. If you cancel this Authorization, the study doctor and staff will not use any new health information for research. However, researchers may continue to use the health information that was provided before you cancelled your authorization.

Can I see my health information? You have a right to request to see your health information. However, to ensure the scientific integrity of the research, you will not be able to review the research information until after the research protocol has been completed.

Signature of participant:____ Date:__________
or participant's legally authorized representative:____________________ Date:__________
Printed Name of participant's representative:____________________
Relationship to the participant:____________________