

**Smoking, Snuffing and Oral Health with
Special Reference to Dental Caries**

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Abstract

Smoking, Snuffing and Oral Health with Special Reference to Dental Caries

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Background and aims: This thesis describes oral health and the use of tobacco with the emphasis on dental caries and Swedish snuff (snus). There appears to be a general opinion in Sweden that snus protects against caries, but there are no scientific studies that support these speculations. More research is therefore needed and the aims of the present investigations were accordingly: 1) to describe the use of tobacco and changes over time (1983-2003) in randomly selected individuals between 15 and 70 years of age, in relation to socioeconomic conditions and dental care habits, 2) to study the relationship between various intraoral caries-associated variables and the effect of smoking and of using snus on dental caries, 3) to study caries-related factors and the pH fall in dental plaque in a group of Swedish snus users and 4) to investigate pH changes in plaque *in vivo* when using different snus products, both with and without nicotine, and to analyse their carbohydrate content.

Methods: Three previous epidemiological cross-sectional studies carried out in 1983, 1993 and 2003 were the platform for Studies I & II (n=2015 and 1591 respectively). The participants were between 15 and 70 years of age. Study III was a clinical study with middle-aged adults (26-62 years old), who had been using snus for ≥ 10 years (n=102) and a control group (n=101) consisting of non-tobacco users. Study IV, in which plaque pH was measured *in situ*, comprised 10 snus users.

Results: Study I showed that there was a statistically significant reduction from 34% tobacco users in 1983 to 28% in 2003. The decrease was most obvious among smokers, while the number of snus users increased somewhat. More tobacco users than non-users did not visit a dentist regularly. In 1983 and 1993 (Study II), there were no significant differences in mean DFS between smokers and non-users, but there was a statistically significantly higher mean DFS in comparison with snus users. Study III, which was carried out in 2009-2011, showed that the salivary secretion rate was higher ($p < 0.001$) in snus users than in non-users (2.50 vs. 2.16 ml/min). Regarding gingival inflammation, non-users showed a mean of 14.4 ± 13.9 and snus users 20.4 ± 18.2 ($p < 0.009$). No statistically significant differences between these two groups were found regarding plaque index, primary and secondary enamel and dentine caries, DFS and salivary counts of mutans streptococci and lactobacilli. The pH fall after a sucrose rinse was more pronounced among the controls than in the snus users (NS). Snus users had fewer snacks between meals compared with non-users ($p < 0.001$). The intraoral pH measurements in Study IV showed that all four nicotine-containing products increased the plaque pH, in contrast to three of the six nicotine-free products, which lowered the pH. The carbohydrate analyses showed only traces of glucose, fructose and sucrose (0.5-1%) and starch ($\approx 1.5\%$) in the various nicotine-containing snus products. Some of the nicotine-free products, on the other hand, contained up to 6.5% low-molecular-weight carbohydrates and 26.0% starch.

Conclusions: 1) During the 20-year period (1983-2003), there was a reduction in the number of smokers and an increase in the number of snus users. Tobacco users had less frequent dental visits and poorer oral hygiene habits than non-tobacco users. 2) The results of the cross-sectional epidemiological studies, performed in 1993 and 2003, indicate that daily smoking and the use of Swedish snus do not appear to increase the risk of dental caries. 3) The clinical study carried out in 2009-2011 showed only minor or no differences in caries and related factors between daily snus users and non-users. 4) All the tested nicotine-containing snus products increased the plaque pH, in contrast to three of the six nicotine-free products, which lowered the pH.

Key Words: carbohydrates, dental care habits, dental caries, nicotine-containing snus, nicotine-free snus, plaque pH, salivary factors, smokeless tobacco, smoking, snuff, socioeconomic factors, tobacco.

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Original Papers

This thesis is based on the following four original papers, which are referred to by their Roman numerals in the text

- I. Hellqvist L, Rolandsson M, Birkhed D, Hugoson A. Tobacco use in relation to socioeconomic factors and dental care habits among Swedish individuals 15-70 years of age, 1983-2003. *Int J Dent Hygiene* 7: 62-70, 2009.
- II. Hugoson A, Hellqvist L, Rolandsson M, Birkhed D. Dental caries in relation to smoking and the use of Swedish snus: epidemiological studies covering 20 years (1983-2003). *Acta Odontol Scand* 2012; 70: 289-96.
- III. Hellqvist L, Hugoson A, Rolandsson M, Lingström P, Birkhed D.
Dental caries and associated factors in a group of Swedish snus users.
Submitted for publication.
- IV. Hellqvist L, Boström A, Lingström P, Hugoson A, Rolandsson M, Birkhed D.
Effect of nicotine-free and nicotine-containing snus on plaque pH in vivo.
Swedish Dental Journal, in press.

Abbreviations and Definitions

| | |
|-------------------|--|
| ANOVA | Analysis of variance |
| Chewing tobacco | Twist, plug or scrap leaf of tobacco. Used orally and chewed intermittently to mix with saliva |
| DFS | Number of decayed (dentine caries) and filled surfaces |
| GI | Gingival index |
| LB | Lactobacilli |
| MS | Mutans streptococci |
| NS | Non-statistical significance |
| PLI | Plaque index |
| SD | Standard deviation |
| Smokeless tobacco | The powdered tobacco leaves are inhaled through the nose, chewed, or stored in cheek pouches. |
| Snuff | Dry or moist form of tobacco, which can be used orally or nasally. |
| Snus | Swedish oral moist snuff, which is placed under the lip and used orally. |
| SPSS | Statistical Package for the Social Sciences |
| SOC | Sense of Coherence |
| WHO | World Health Organisation |

Introduction

Tobacco products

The leaves from the tobacco plant are used as the main product when producing snus and cigarettes as well as other tobacco products. Swedish snus has been manufactured since the early 1800s. In the late 1960s, it became more popular and has been regulated under the Swedish Food Act since 1971. The tobacco leaves are ground or cut into a powder and then undergo a heat treatment process. Other ingredients are blended to achieve a specific nicotine content, pH, taste, flavour and aroma and a moist non-fermented product (Foulds et al., 2003; Rutqvist et al., 2011). Snus contains tobacco-specific nitrosamines, even if Swedish snus has lower levels than American snuff, according to comparative studies (Rodu et al., 2004). The content of tobacco-specific nitrosamines has been reported as the carcinogenic agent in snus (Gupta et al., 1996; Rodu et al., 2004; Hall et al., 2009). Typical Swedish snus has a pH of about 8.5 (Andersson et al., 1994) and the product is usually placed under the upper lip as portion-packed snus or in loose form. The portion-packed snus contains portion bags of 0.4-2.0 grams of snus. Both kinds are packed in boxes, usually 50 grams for loose snus and portion-packed snus containing 24 and 10 grams (Digard et al., 2009). The salivary pH is significantly higher with a pouch of snus in the mouth during the day than in the morning 6-8 hours after snus use (Andersson et al., 2003).

Snus contains nicotine, which produces dependence comparable to the dependence on other tobacco products such as cigarettes (Foulds et al., 2003). Experience and symptoms of lifetime nicotine dependence were studied and snus users displayed symptoms at least as frequently as cigarette smokers. Dual users had a higher consumption of snus compared with exclusive snus users, despite concurrent cigarette use. Exclusive snus users had a lower incidence of prior attempts at quitting compared with smokers and dual users (Post et al., 2010). The increase in snus and “the underlying mechanisms that contribute to this trend” were studied among young Norwegian snus users. Among the participants, more than 50% reported that they agreed or totally agreed with the beliefs such as “using snus makes me relax”; “using snus satisfies my need for nicotine” and so on. Finally, snus was mainly associated with smoking control and mood regulation (Wiiium et al., 2011).

The marketing of snus is banned in all member states in the European Union EU, except for Sweden. However, the EU scientific committee has concluded that there is not enough evidence to draw any conclusions about the health effects of snus (Ahlbom et al., 2007). In Sweden, since 1993, there have been restrictions on tobacco usage, Swedish Tobacco Act, with amendments added in 1994, 1997, 2002, 2004 and 2005 (SFS 1993:581,). The act replaces earlier legislation on health warnings and a partial ban on advertising. In January 1997, an age limit of 18 years for the purchase of tobacco products was introduced and in 2002 additional restrictions were adopted. They included a ban on indirect tobacco advertising. In 2005, smoking in bars and restaurants was prohibited.

Both nationally and internationally, there are a number of different tobacco products on the market and new products are constantly being introduced. For definitions of smokeless tobacco products, see Table 1.

Table 1. Definition of various smokeless tobacco products

| Products | Definition |
|--------------------|---|
| Chewing tobacco | Twist, plug or scrap leaf of tobacco. Used orally and chewed intermittently to mix it with saliva |
| Nicotine-free snus | Snus substitute, contains no nicotine |
| Smokeless tobacco | The powdered tobacco leaves are inhaled through the nose, chewed, or stored in cheek pouches. |
| Snuff | Dry or moist form of tobacco. Can be used orally or nasally. |
| Snus | Swedish oral moist snuff, which is placed under the lip and used orally. |

Tobacco and general health

The global tobacco epidemic is one of the largest threats public health has ever faced and its use is one of the greatest challenges for prevention in public health. Most tobacco users are unaware of the harm tobacco use causes (World Health Organisation, 2008). Smoking harms almost every organ of the body and diminishes a person's overall health and the medical side-effects of smoking have been reported in a large number of studies (Persson et al., 2000; Conroy et al., 2003; Li et al., 2005). Longitudinal studies in Europe have shown that cigarette smoking is one of the most common risk factors causing cardiovascular disease (Conroy et al., 2003).

In Sweden the prevalence of smoking declined to 10% in men and 12% in women in 2011 (Statens folkhälsoinstitut, 2012). The prevalence in Sweden has decreased in recent decades; in 1980 35% of men were smokers and 28 % of women (Shafey O. et al., 2012). The decline in smoking in Sweden is thought to be mainly due to increasing snus use, since the prevalence of snus use in 2011 in Sweden was 18% for men and 3% for women (Statens folkhälsoinstitut, 2012) the highest prevalence of smokeless tobacco in Europe. When smokers stop smoking, some use snus as an aid to success and according to the author, 50% of the snus users are ex-smokers (Foulds et al., 2003). However, contradictory results showed that the majority of the smokers who stop smoking do so without starting to use snus (Lundqvist et al., 2009). The use of both snus and cigarettes is often called "dual use" and it has also been studied in a recent prospective study of male Norwegian adolescents. The results showed that snus users at baseline, who were dual users at follow-up, had increased odds of being daily snus users and occasional smokers (Grötvedt et al., 2012). There is also a scientific question of whether snus use may increase the risk of taking up smoking. Some studies among adolescents from the USA and Sweden conclude that smokeless tobacco use alone is not a significant risk factor for the subsequent use of cigarettes (Kozlowski et al., 2003; Galanti et al., 2008; Timberlake et al., 2009). While there are other studies with contradictory results, the authors describe this as a "gateway hypothesis" (Haddock et al., 2001; Kozlowski et al., 2003). According to Haddock and co-workers (2001), smokeless tobacco use appears to be a predictor of smoking initiation among young adult males in the US forces and they suggest that smoking prevention and cessation programmes should also include strategies related to use. To conclude, two recent reviews showed that more research is needed to clarify this question (Colilla, 2010; Lee, 2011).

The medical effects of snus use are not so well described, but a Swedish study showed that snus use is a risk factor for pancreatic cancer (Luo et al., 2007). Other studies have shown that snus users run a higher risk of cardiovascular disease (Bolinder et al., 1994; Bolinder et al., 1998). On the other hand, there are studies that have presented contradictory results (Hansson et al., 2009; Lee, 2011). In a recent review, summarising evidence relating snus to health, the conclusion is that snus use is much safer for health than smoking (Lee, 2011). Snus is a product that has traditionally been used in Sweden since the early nineteenth century and it differs from many other smokeless tobacco products due to its lower content of nitrosamines (Österdahl et al., 2004). However, the levels of blood nicotine after one dose of smokeless tobacco at the same levels are the same as after smoking one cigarette and the nicotine doses after one pouch of smokeless tobacco have a longer duration (Benowitz, 1997). The addictive capacity of snus is also comparable to that of smoking (Henningfield et al., 1999; Post et al., 2010). In a recent study carried out in Sweden, symptoms of lifetime nicotine dependence on snus and cigarettes was studied and snus users reported statistically significantly higher symptoms of dependence compared with smokers (Post et al., 2010).

Gender and tobacco use

In most parts of the world, being born male is the greatest predictor of tobacco use. The prevalence globally is about four times higher among men than among women (48 vs. 12%) (Corrao et al., 2000). However, according to the Global Youth Tobacco Survey (Warren et al., 2009), young girls smoke almost as much as young boys. Girls and boys also use non-cigarette tobacco products such as spit tobacco and water pipes at similar rates. Life style is often regarded as an optionally choice for each individual and each person makes choices about his or her behaviour, such as exercise, consumption of food, alcohol and tobacco (Hammarström et al., 1999).

National numbers show that, among men with a low socioeconomic status, smoking is more common (Statens folkhälsoinstitut, 2012). Men have a shorter life expectancy, approximately

four years shorter in Sweden, and also run a higher risk of cardiovascular disease according to statistics in Sweden (SCB Statistics of Sweden, 2010). According to a study performed in Stockholm, men with low and intermediate education level used snus more than men with high educational levels (Engström et al., 2010). Female smokers are more common among women with a low income and low education, compared with individuals with a high education and income. Male smokers are also more represented among individuals with a low education and low income. It is interesting to note that the highest rates among both women and men are found in smokers who are unemployed (Statens folkhälsoinstitut, 2012). These data are also confirmed in a review study of international research published between 1990 and 2010 (Henkel, 2011).

In Sweden, the decreasing prevalence of snus use from north to south is well known. In the northern part of the country, the use of snus is more common than in the south of the country among both women and men (Statens folkhälsoinstitut, 2012). Cultural background can also be a reason for differences in tobacco use. Smoking among citizens born in other countries in Europe but now living in Sweden, among both women and men, shows higher rates than among citizens born in Sweden. The use of snus is a typical Swedish or northern habit and, among immigrants, this use is somewhat uncommon according to national rates (Statens folkhälsoinstitut, 2012). From the beginning, tobacco use, especially the use of snus, was a traditional male habit. In a qualitative study conducted on ice-hockey-playing boys, the result showed that the informants describe the environment as cool and rough and the players were macho and masculine (Rolandsson et al., 2006). The results of this study can be applied to theories of hegemonic masculinity (Connell, 2005) where the power relationships are seen as natural and to legislating the hierarchy among different social groups in the community is accepted.

Tobacco and oral health

A qualitative study aiming to explore and describe attitudes to oral health among adolescents with a high caries risk the authors found that their views of oral health are important for the dental team when encouraging dental caries risk patients to make healthy choices (Hattne et al., 2007). Oral health was also primarily regarded as the health of the teeth and caries was considered to be

the result of a frequent intake of sweetened drinks and candy. This was also confirmed in an epidemiological study conducted in 1973, 1983, 1993 and 2003, where more than half the subjects were aware of the fact that bacteria and sugar are the main sources of acid formation for the further development of dental caries (Hugoson et al., 2005a). Fresh-smelling breath was also seen as being of great importance and a sign of good oral health when Hattne and co-workers studied adolescents (Hattne et al., 2007). Health behaviour and attitudes to health are influenced by different factors such as education, socio-economics, culture and place of residence (Hjern et al., 2001; Källestål et al., 2002; Poutanen et al., 2007). Oral health professionals have traditionally focused on preventive and educational action and altering the behaviour which was seen to be the cause of dental diseases; this is based on a biomedical model of disease. The theory behind this approach is that, when individuals acquire the relevant knowledge and skills, they will then act and approve good oral health. This lifestyle approach has dominated the preventive practice for many decades all over the world (Watt, 2005). In a review (Yevlahova et al., 2009) studying the effectiveness of models for oral health promotion, the authors found that “there is a need for more supportive rather than judgmental approaches to oral health behavior change”. Motivational interviewing (MI), based on theories such as the Transtheoretical Model, has been found to be one of the most effective approaches to altering clients behaviors (Hollister et al., 2004; Yevlahova et al., 2009).

Oral health is connected to habits associated with life style and influenced by a person’s way of acting. The different choices an individual makes regarding oral hygiene, choice of food habits, tobacco use and attitudes to health are examples of lifestyle issues that predict oral health. A person’s “sense of coherence” (SOC) which is based on “Antonovsky’s salutogenic theory” and searches for “the origin of health” focuses on the causes of disease and what creates health (Antonovsky, 1987). An understanding of SOC and the salutogenic perspective can help dental professionals to understand an individual’s choice or behaviour in relation to health. A strong SOC appears to be positively related to oral health-related behaviour, attitudes and knowledge of oral health, which was recently studied in a cross-sectional study among individuals in aged 20-80 years in Sweden (Lindmark et al., 2011). A Finnish cross-sectional study showed that a strong SOC has a positive association with several health and oral health behaviour factors (Savolainen et al., 2009).

A number of scientific studies within oral health research in recent years have demonstrated the effects of tobacco use on both the mucosa and periodontal tissues (Axelsson et al., 1998; Norderyd et al., 1998; Pihlström et al., 2005). A recent study based on epidemiological data showed that cigarette smokers run a significantly higher risk of periodontitis compared with non-tobacco users and snus users (Hugoson et al., 2011).

In periodontal care improving oral hygiene and tobacco use cessation have been necessary to achieve and maintain periodontal health (Axelsson et al., 2004; Needleman et al., 2006; Binnie et al., 2007). Tobacco prevention in dental care is an important task for dental hygienists and dentists. During the last two decades, 90-95 % of all individuals, irrespective of age, have visited the dental team regularly, once every two years and this makes it possible to support and follow up a healthy choice, free from tobacco (Hugoson et al., 2005a; Rolandsson et al., 2005). Regular visits to the dental team were shown to have an impact on affecting and preserving an individual's knowledge and oral health behaviour (Sheiham, 1977; Nuttall, 1997; Rosen et al., 1999). A recent intervention study of adolescents, who were considered to run a potential risk of dental disease, showed that the participants had a negative attitude towards tobacco use both before and after the intervention (Hedman et al., 2010). The authors also found that an important factor that kept the participants away from tobacco use was the approach of parents and friends and the dangerous effects of tobacco.

Smoking and dental caries

Several studies showing an association between dental caries and smoking have been published (Modéer et al., 1980; Hirsch et al., 1991; Axelsson et al., 1998; Fure, 2004; Vellappally et al., 2007; Skudutyte-Rysstad et al., 2009). Axelsson et al. (1998) found a connection between caries and smoking, but oral hygiene was similar compared with non-users. Among professional truck drivers in Mexico, higher age, poorer oral hygiene, higher education and greater tobacco exposure were associated with higher caries experience (DMFT) (Aguilar-Zinser et al., 2008). It has also been shown that smoking is associated with poor oral hygiene and food habits, which could increase the caries risk (Hirsch et al., 1991; Bruno-Ambrosius et al., 2005; Aguilar-Zinser et al., 2008). According to a health survey from 2001, long-term smokers did not visit a dentist as often as non-smokers and the likelihood of an annual visit for an inspection decreased with an

increase in the amount of smoking/day. There was also a connection with smokers who only consumed a small amount of fruit and vegetables (Mucci et al., 2001). A recent study among Italian servicemen showed that heavy smokers had a higher prevalence of caries compared with non-smokers and light smokers (Campus et al., 2011).

Table 2. Summary of studies of the association between tobacco use and dental caries. A positive association is marked in yes.

| First author, year | Study design | Caries |
|----------------------------|-------------------------------------|--------|
| Mod er; 1980 | Clinical | Yes |
| Weintraub; 1987 | Review | |
| Hirsh; 1991 | Clinical | Yes |
| Axelsson; 1998 | Cross- sectional Epidemiological | Yes |
| Tomar; 1999 | Epidemiological | Yes |
| Unell; 1999 | Clinical | Yes |
| Reibel ; 2003 | Review | |
| Fure ; 2004 | Cross-sectional follow- up | Yes |
| Vellappally; 2007 | Review | |
| Bruno-Ambrosius; 2005 | Longitudinal | Yes |
| Rolandsson; 2005 | Clinical | No |
| Aguilar-Zinser; 2008 | Clinical | Yes |
| Skudutyte-Rysstad; 2009 | Clinical | Yes |
| Campus: 2011 | Clinical | Yes |

Snus and dental caries

There is a lack of studies of the way the use of snus influences dental caries. American smokeless tobacco might increase the risk of dental caries, which was shown by (Weintraub et al., 1987). In two reviews, an association was found between smokeless tobacco and dental caries (Vellappally et al., 2007; Lee, 2011). The American snuff and other smokeless products differ from Swedish snus in terms of content and pH (Idris et al., 1998; Tomar et al., 1999; Foulds et al., 2003). This makes it difficult to compare them from a cariological point of view. A clinical study conducted

in 2005 among adolescent snus users and non-users as a control showed that there was no statistically significant difference in terms of filled teeth (Rolandsson, 2005).

Caries-related factors

Dental caries is one of the most common diseases and affects the majority of the population in both the industrialised and developing countries (Petersen, 2003). Caries is a multifactorial disease and it is an interaction between diet, microorganisms and host (Keyes, 1960; Selwitz et al., 2007). These three factors are involved in cycles of demineralisation and remineralisation. The early stages of caries can be reversed depending on the balance in the different defence factors.

A frequent high intake of fermentable carbohydrates, especially sucrose, has been known to be associated with caries initiation and development for a review, see (Paes Leme et al., 2006). This was already established in the Vipeholm Study more than 60 years ago (Gustafsson et al., 1954). A review by (Zero, 2004) made it clear that poor oral hygiene in combination with frequent sugar consumption appears to increase the caries prevalence. However, with frequent exposure to fluoride, this relationship between sugar consumption and caries experience is not so clear (Sundin et al., 1983; Burt et al., 2001). The consumption of sweetened drinks has raised and added to the sugar consumption in the diet of children and adolescents and studies have shown that the frequent intake of caries risk products during the first years of life is associated with caries development during pre-school years (Wendt et al., 1995). However, no evidence showing that simply reducing sugar in the diet is an effective activity in caries-prevention measure was found in a review (Lingström et al., 2003).

Saliva plays an important role in oral health maintenance. It has several caries-prevention functions, even if flushing and neutralising effects, also referred to as “oral clearance” appear to be the most effective caries-preventive functions, according to a review based on saliva and dental caries (Lenander-Lumikari et al., 2000). There is a relationship between salivary secretion capacity, caries activity and DMFS/DMFT values (Lenander-Lumikari et al., 2000). These authors also state that buffer capacity in saliva is in generally dependent on the secretion rate and

more saliva therefore produces a higher buffer capacity. The buffering capacity of saliva also influences the pH of plaque on the tooth surface and thereby the caries process (Humphrey et al., 2001).

Fluoride primarily works in three different ways: 1) It slows down the demineralisation process, 2) it stimulates remineralisation and 3) it affects the bacterial metabolism in plaque. The daily use of fluoride toothpaste is one of the best preventive methods for reducing dental caries (Bratthall et al., 1996). In a review in which the caries-prevention effect of fluoride-containing toothpaste was evaluated, the authors found strong evidence that the daily use of fluoride toothpaste reduces dental caries (Twetman et al., 2003). They also concluded that toothpaste with a higher concentration of fluoride appears to provide better protection from caries than toothpaste with a lower concentration. This was also confirmed in a study in which dentifrice with a high fluoridated content (with 5000 ppm) was tested on young adults running a high risk of dental caries (Nordström et al., 2010).

Oral micro-organisms colonise the tooth surface by forming a biofilm, called dental plaque. If it is not removed, it can increase and build up a thick layer (Takahashi et al., 2011). Epidemiological studies in teenagers have recently revealed high scores for plaque and gingivitis (Abrahamsson et al., 2006; Ericsson et al., 2009). Lactobacilli and mutans streptococci are bacteria with a great ability to produce acid and are regarded as important in the caries process, even if there are other bacteria which influence the process (Russell, 2008; Takahashi et al., 2011).

According to an epidemiological study carried out in Sweden, most individuals (>70%) brushed their teeth at least twice a day in 2003 (Hugoson et al., 2005a). The percentage of individuals who used dental floss every day was approximately 15%. Perception, attitudes and behaviour toward oral health were studied among individuals aged 19 and the conclusion was that adolescents with high scores for plaque and gingivitis had a less positive attitude, perception and behaviour (Ericsson et al., 2012). There are some gender differences in oral hygiene habits; boys generally have poorer oral hygiene than girls (Östberg et al., 2001).

According to Swedish law, dental care should be offered on equal terms in the entire community (SFS 1985:125,). Socioeconomic status has been found to be one of the main factors for inequality in dental health (Locker, 2000). A Swedish cross-sectional study, based on interview data, showed that there is considerable inequality in the use of dental care and oral health in Sweden (Hjern et al., 2001). There was a general tendency for the time between treatments to increase in the 1996–97 survey compared with the 1988–89 survey. This is confirmed in an epidemiological study, where individuals aged 30-40 years did not visit a dentist as regularly in 2003 as in 1993 (Hugoson et al., 2005a). In a recent study in which the oral health and the self-perceived oral treatment need of adults were investigated, the author found “that important factors for the prediction of a high oral treatment need were a low educational level, previous unmet perceived oral treatment need, frequent visiting pattern, perception of worse oral health, external locus of control, and to have received information from one's dental caregiver about a need for oral treatment” (Lundegren, 2012). In a longitudinal study of dental health the most obvious difference in dental health due to gender (Crossner et al., 2007). The experience of proximal caries and bleeding after probing was twice as frequent in males as in females.

Caries risk assessment and plaque pH

Caries risk assessment have become increasingly common in dental health practice for the prevention and care of dental caries (Twetman et al., 2009; Brocklehurst et al., 2011). One example is the Cariogram which is used to score the caries risk of each patient. This computer program is based on ten different factors and the risk is expressed as “the percent age chance of avoiding new lesions in the near future” (Bratthall et al., 2005). The program has been used in school children and among the elderly and adults (Hänsel Petersson et al., 2002; Hänsel Petersson et al., 2003).

Plaque pH is an important variable for measuring pH in situ in order to evaluate the cariogenicity of food products and the microtouch method is commonly used method (Lingström et al., 1993). The registration of plaque pH has become a tool for assessing the individual risk of caries (Scheie et al., 1992; Aranibar Quiroz et al., 2003). A hand-held microelectrode is inserted into the area of

measurement and the resulting pH curve is generally assumed to assess the cariogenic potential of the plaque flora (Lingström et al., 1993; Aranibar Quiroz et al., 2003).

Aims

Many studies have been published on the way the use of tobacco affects oral health in a periodontal perspective, but fewer studies have been published from a cariological perspective.

The aims of this thesis were therefore:

- to describe the use of tobacco and changes in its use over time among individuals living in Jönköping, Sweden, and to analyse tobacco habits in relation to socioeconomic conditions, personality aspects and dental care habits,
- to evaluate, among individuals from the same population living in Jönköping, some intraoral caries-associated variables and tobacco use from a cariological point of view,
- to investigate some caries-related factors and their effect on dental caries in a group of middle-aged habitual snus users and to measure the pH fall in dental plaque in situ in a subgroup after a sucrose rinse,
- to study changes in plaque pH in vivo when using different snus products, both nicotine-free and nicotine-containing, and to analyse their carbohydrate content.

Material and Methods

The material in this thesis is based on four studies (I-IV). Studies I & II were performed in the County of Jönköping and Studies III & IV at Karlstad University. The design and sample size of the four studies are given in Table 3.

Table 3. The four studies and their corresponding design, sample size and sample.

| Study | Design | Sample size (n) |
|------------|--|-----------------|
| I | Epidemiological, cross-sectional study | 2015 |
| II | Epidemiological, cross-sectional study | 1591 |
| III | Clinical study | 203 |
| IV | Experimental study | 10 |

The ethical rules for research laid down in the Declaration of Helsinki (Declaration of Helsinki, World Medical Association 1964.) were followed in all four studies. Studies I & II were approved by the Ethics Committee at the University of Linköping and Studies III & IV by the Ethics Committee at Karlstad University.

Study I

The aim was to describe the use of tobacco and changes in its use over time among individuals living in Jönköping, Sweden, and to analyse tobacco habits in relation to socioeconomic conditions, personality aspects and dental care habits.

Study population and methodology

The study population was taken from three epidemiological cross-sectional studies conducted in Jönköping, Sweden, in 1983, 1993 and 2003 (Hugoson et al., 1986; Hugoson et al., 1995; Hugoson et al., 2005a). One hundred and thirty randomly selected individuals in each of the age cohorts between 15-70 years of age were asked to take part in an oral health examination. A questionnaire was completed at the same time as the examination. The questionnaire for the 15-year olds comprised 23 questions, while the questionnaire for the 20- to 70-year olds contained 101 questions; edentulous individuals answered 68 of these questions. The same questionnaire was used in all three studies. They were focused on demographic, socioeconomic, medical and oral health history, dental habits, tobacco habits and oral hygiene habits. Marital status and financial situation were also noted for the age group of 20 years and above. Smokers and snuff users were defined as daily users and participants who both smoked and used snuff as “dual users”. The individuals were asked to answer Antonovsky’s questionnaire, the SOC scale, relating to SOC in a Swedish version containing 13 questions. A high score indicated a strong SOC.

Study II

The aim was to evaluate, among individuals from the same population living in Jönköping, some intraoral caries-associated variables and tobacco use from a cariological point of view.

Study population and methodology

Study II consisted of the same samples as in Study I, except that, in Study II, no edentulous individuals participated. They were informed of the purpose of the investigation and that clinical and radiographic examinations would be performed. A questionnaire was also completed at the time of the examination. Thirty-four dual tobacco users were excluded from this population.

The clinical examination took approximately 60-90 min. A saliva sample was collected and the secretion rate was expressed as ml/min. Buffer capacity was also estimated. In 1983, the numbers of MS and LB were analysed. The number of teeth was recorded; third molars were excluded. Filled tooth surfaces were registered. All surfaces available for clinical examination were examined for dental caries. All surfaces that could not be evaluated clinically were evaluated on

the radiographs. Oral hygiene was registered as the presence of visible plaque on four tooth surfaces per tooth using Plaque Indices (PLI) 2 and 3. If a participant had had a recent radiographic examination, the radiographs were obtained from the records. According to improved oral health and ethical considerations the radiographic examination changed during the course of the studies.

Study III

The aim was to investigate some caries-related factors and their effect on dental caries in a group of middle-aged habitual snus users and to measure the pH fall in dental plaque in situ in a subgroup after a sucrose rinse.

Study population and methodology

The study was carried out between 2009 and 2011 and the subjects comprised of male and female adults, between 26 and 62 years of age (n=102). They were recruited from individuals working at different companies and with different educational levels living in the city of Karlstad or the surrounding areas. They were considered themselves as healthy, non-smokers and daily snus users ≥ 10 years. Most of the snus users (n=66) consumed less than one box/day, 26 one box/day and eight more than one box/day. Sixty individuals had been using snus for more than 20 years and 37 of them for more than 25 years. The control group was considered themselves as healthy, with no use of tobacco during the ≥ 10 last years. They were selected in order to be as similar to the test group as possible in terms of age, gender and educational level. The final control group (n=101) consisted of 29-to 61-year-old individuals. A total of 203 (102+101) subjects, of which 188 (93%) were males, participated in the study.

All the participants were asked to complete a questionnaire at the same time as the clinical examination. It contained 32 questions about medical health, educational level and tobacco, dietary and oral care habits. The clinical examination was carried out by one of the authors (LH) and the clinical and radiographic examination took around 60-90 min. The number of teeth, as well as the number of DFS, was recorded; third molars were excluded. All tooth surfaces available for clinical evaluation were examined for caries and tooth surfaces that could not be

evaluated clinically were evaluated on radiographs. The participants were instructed not to eat, drink or use snus one hour before the sampling. A paraffin-stimulated saliva sample was collected and expressed as ml/min and buffer capacity was estimated in a chair-side test (CRT Buffer; Vivacare, Schaan, Liechtenstein). One millilitre of the saliva was sent to the Dental School in Gothenburg to estimate the number of MS and LB. The presence of visible plaque was recorded on six surfaces as plaque index PLI grades 2 and 3 (Silness et al., 1964). GI grades 2 and 3 were also assessed at six sites (Løe, 1967). A caries risk assessment was made using the Cariogram computer program (Bratthall et al., 2005).

Twenty subjects, 10 snus users and 10 non-users, were randomly selected from the 203 subjects. They came to the laboratory for a one-hour visit (non-users) or for a two-hour visit (snus users). They were asked not to brush their teeth for three days prior to the test and were told not to eat, drink or use snus for two hours prior to the test. Plaque pH was measured in situ with a microtouch electrode. Measurements were made at three approximal sites in the upper jaw; one in the premolar and two in the front region (mesial 15, distal and mesial 13). Plaque pH was measured before and on eight occasions up to 45 min after a one-minute mouth rinse with 10 ml of a 10% sucrose solution. This was done twice in the 10 snus users. Directly after the first test, one portion of snus was placed under the upper lip, region 13. The subjects then rinsed again with sucrose, after which pH was followed for another 45 min.

Study IV

The aim was to study changes in plaque pH in vivo when using different snus products, both nicotine-free and nicotine-containing, and to analyse their carbohydrate content.

Study population and methodology

Ten subjects, seven males and three females, with full dentition and regarded as being healthy, participated in the study. They were all snus users and non-smokers and they were recruited from students and staff at the university. Each subject came to the laboratory eleven times for a one-hour visit and there was a minimum of one week between each visit. The volunteers were asked not to brush their teeth for three days prior to testing and were not allowed to eat, drink or use snus for two hours before each test. On the first visit, a whole saliva sample was collected by

chewing a piece of paraffin wax to determine the secretion rate, buffer capacity and number of cariogenic micro-organisms.

Plaque pH was measured using a microtouch electrode. Measurements were made at three approximal sites in the upper jaw, mesial 15, distal and mesial 13. pH was measured before and at eight points of time within 45 minutes after a one-minute mouth rinse with a 10% sucrose solution (positive control). On the remaining ten occasions, pH was measured before and after a portion of the snus product was placed under the upper lip, region 13, and pH was measured at the three sites in the same way as the first visit. Ten products were tested in randomised order and all subjects tested all products. Six nicotine-free and four nicotine-containing products were purchased on the open market. They were analysed for the percentage of low-molecular-weight carbohydrates, using high-pressure liquid chromatography (HPLC), and of starch.

Statistical methods (Studies I-IV)

All the analyses were performed using the Statistical Package of Social Sciences (SPSS). Descriptive methods were used to describe the populations and various statistical methods were used in the different studies. In *Study I*, calculations of statistical significance between groups and variables were made using chi-square analyses. Multivariate stepwise logistic regression was used in order to analyse the relationship between groups. The difference in SOC score between tobacco users and non-users was tested using Student's *t*-test. Continuity correction was also used to define differences in the proportion in the groups between different study years. In *Study II*, ANOVA (Analysis of Variance), including the post-hoc test according to Sheffé, was used to study differences between the three groups. ANOVA was also used to study differences over time. Multivariate logistic regressions were used and the odds ratio (OR) was calculated. The analyses in *Study III* were chi-square analyses to estimate mean values between variables and groups. Differences between groups were calculated using Student's *t*-test. The inter-examiner agreement was calculated using Cohen's kappa. In *Study IV*, the mean values of the three sites were calculated. The area under the curve (AUC) for the six nicotine-free products and the area over the curve (AOC) for the four nicotine-containing products were analysed using a computer program. The AUC/AOC values were compared using analyses of variance ANOVA. $p < 0.05$ was considered statistically significant in all four studies.

Drop-outs

The percentage of drop-outs in Studies I and II was (12-28%) in the study years 1983 and 1993 and (26-36%) in 2003. For details, see (Hugoson et al., 2005a). In Study III, a total of 295 individuals were contacted; this meant that there were 92 non-respondents. There were two missing samples regarding measurements of salivary secretion rate among the snus users and two of buffer capacity among the non-users.

Results

Study I

Tobacco habits. Of the sample in 1983, 34% were tobacco users (smokers, users of snuff and mixed users). In 1993 and 2003, the corresponding figures were 27% and 28% respectively. The percentage of tobacco users was lowest in all three cohorts among 15- and 70- year-olds. In all three cohorts, smoking was more frequent than the use of snuff in all age groups, except in 15-year olds in 1983. The total percentage of smokers decreased gradually from 1983 to 2003, from 27%, 18% and 16% in 1983, 1993, and 2003; the reduction was statistically significant ($p < 0.001$) between 1983 and the two last years. The decline was the same for males and females between 1983 and 2003. The use of snuff increased from 6% in 1983 to 10% in 2003 ($p < 0.001$).

The percentage of smokers declined between 1983 and 2003 in all age groups; during the same period, the percentage of snuff users increased in the 20-60 age groups. In 1983, there were no smokers among males in aged 15, while 9% of females were smokers. At the same time, however, 19% of the males used snuff but none of the females. Among women in the 20-year group, there were 49% smokers and the rate among 60-year-old men was 41%, which was the highest percentage value. Snuff use was most common among 20-year-old men (29%). In 2003, 2% of 15-year-old males and 6% of females stated that they were smokers. Among 20-year-old men, 28% said that they were smokers and 26% that they used snuff. The use of snuff increased in the 40-60 age groups in all the study years.

There was no statistically significant difference between low and high earners and low and high educational levels in the different cohorts according to tobacco use. In 2003 a statistically significant difference was found between tobacco users and non-users in terms of marital status. Among single men compared with men living in a partnership, the use of tobacco was higher. No statistically significant difference was found between tobacco users and non-users, either between dual users or non-users, in terms of SOC scores.

In the 2003 cohort, there was a statistically significant difference between tobacco users and non-users regarding the frequency of dental visits; more tobacco users stated that they did not make regular visits to a dental clinic (Table 3). In the other cohorts, no statistically significant

difference was found. There was no significant difference between tobacco users and non-users with respect to reasons for dental visits.

Oral hygiene habits. Most tobacco users and non-users brushed their teeth twice a day. In 1993, there was a statistically significant difference regarding toothbrushing frequency; it was higher among non-users than among tobacco users. In 1983 and 2003, there was a statistically significant difference in the regular use (every day) of toothpicks ($p < 0.000$ and $p < 0.039$) between tobacco users and non-users: use was higher among non-users Table 4.

Table 4. Percentage distribution of tobacco users and non-users according to dental visits and oral hygiene habits in 1983, 1993 and 2003.

| | 1983 | | 1993 | | 2003 | |
|--------------------------------|---------------|-----------|---------------|-----------|---------------|-----------|
| | Tobacco users | Non-users | Tobacco users | Non-users | Tobacco users | Non-users |
| <i>Dental visits</i> | | | | | | |
| Every year | 82 | 83 | 82 | 86 | 62 | 71 |
| Every other year | 10 | 8 | 12 | 11 | 20 | 19 |
| None of the years | 8 | 9 | 6 | 3 | 18 | 8 |
| <i>Toothbrushing frequency</i> | | | | | | |
| Twice or more a day | 89 | 91 | 80 | 90 | 87 | 90 |
| Once a day | 11 | 8 | 18 | 8 | 9 | 7 |
| Now and then | 0 | 2 | 2 | 1 | 4 | 1 |
| Regular users of toothpicks | 32 | 39 | 30 | 35 | 27 | 33 |

Study II

Tobacco habits. The mean \pm SD age of non-tobacco users was 45.9 ± 16.9 years, whereas it was 40.9 ± 15.8 years for smokers and 34.9 ± 14.3 years for snus users. The differences in mean age were statistically significant. The percentage of smokers was almost halved (from 29% to 15%), whilst the use of snus rose from 4% to 9%. There were more female than male smokers and most of the snus users were male. Among the daily smokers, $\geq 60\%$ smoked ≥ 10 cigarettes a day for each year of examination.

Table 5. Number of individuals examined, mean values, 95% CI and *p*-values for non-users, smokers and snus users in Study II. Only significant variables are given.

| | 1983 | | | 1993 | | | 2003 | | |
|-------------------------|----------|----------|-----------------|----------|----------|-----------------|----------|----------|-----------------|
| | <i>n</i> | Mean | <i>p</i> -value | <i>n</i> | Mean | <i>p</i> -value | <i>n</i> | Mean | <i>p</i> -value |
| Number of teeth | | | | | | | | | |
| Non-users | 358 | 23.0±0.6 | 0.010 | 402 | 24.5±0.5 | | 382 | 25.4±0.4 | 0.001 |
| Smokers | 159 | 23.1±1.0 | 0.016 | 103 | 24.0±1.2 | | 83 | 23.3±1.4 | 0.001 |
| Snus users | 22 | 27.1±0.7 | | 38 | 26.0±1.3 | | 44 | 26.8±0.6 | |
| Buffer capacity | | | | | | | | | |
| Non-users | 352 | 4.9±0.1 | 0.001 | 392 | 4.4±0.1 | | 343 | 5.7±0.1 | |
| Smokers | 157 | 4.4±0.2 | 0.004 | 103 | 4.2±0.2 | 0.047 | 75 | 5.6±0.2 | |
| Snus users | 22 | 5.2±0.4 | | 37 | 4.6±0.2 | | 40 | 5.8±0.1 | |
| DFS (% surfaces) | | | | | | | | | |
| Non-users | 358 | 41.3±2.8 | 0.001 | 402 | 35.8±2.4 | 0.003 | 382 | 27.8±2.3 | |
| Smokers | 159 | 41.3±3.5 | 0.001 | 103 | 37.1±5.1 | 0.004 | 83 | 28.8±5.8 | |
| Snus users | 22 | 20.2±4.4 | | 38 | 21.8±5.9 | | 44 | 19.3±5.0 | |
| MS | | | | | | | | | |
| Non-users | 340 | 5.9±0.1 | 0.001 | | | | | | |
| Smokers | 152 | 6.2±0.2 | | | | | | | |
| Snus users | 22 | 5.9±2.0 | | | | | | | |
| LB | | | | | | | | | |
| Non-users | 300 | 5.0±0.1 | 0.001 | | | | | | |
| Smokers | 142 | 5.7±0.2 | 0.044 | | | | | | |
| Snus users | 18 | 4.9±0.5 | | | | | | | |

Clinical variables. Table 5 shows the mean ± SD for each year of examination (1983, 1993 and 2003) and the *p*-values are presented for non-tobacco users, smokers and snus users. Only significant variables are given. Regarding the number of teeth, non-users and smokers had statistically significantly fewer teeth in 1983 than snus users. In 1983, there were statistically significant differences between non-users and smokers, as well as between smokers and snus users, when it came to buffer capacity. In 1983, there was a higher DFS in non-users and smokers compared with snus users (*p*<0.001). In 1993, non-users and smokers had a statistically significantly higher number of DFS than snus users. In 1983, smokers had a statistically significantly higher mean number of MS compared with non-users. Smokers also had a higher number of LB than both non-users and snus users.

The secretion rate for non-users was lower in 2003 compared with 1993. When comparing PLI in 2003 with PLI in 1983 and 1993, there were lower mean plaque values for non-users in 2003 and for snus users compared with 1993, but there was no significant difference for smokers.

Table 6. Results of multiple logistic regression analysis of the association between the outcome variable (DFS %, low/high) and various explanatory variables, adjusted for age, gender and socio-demographic variables in the different years of examination 1983, 1993 and 2003. Only significant variables are given.

| | | | |
|-----------------|------|------------|-------|
| 1983, n = 518 | | | |
| Gender | 2.81 | 1.76–4.49 | 0.001 |
| Age | 1.10 | 1.07–1.12 | 0.001 |
| Employed | 1.56 | 1.25–1.95 | 0.001 |
| Non-users | 1 | | |
| Smokers | 2.32 | 1.41–3.81 | 0.001 |
| 1993, n = 507 | | | |
| Gender | 1.89 | 1.10–3.22 | 0.020 |
| Age | 1.13 | 1.10–1.17 | 0.001 |
| Education (low) | 2.57 | 1.35–4.87 | 0.004 |
| Employed | 3.14 | 1.59–6.19 | 0.001 |
| PLI | 1.02 | 1.01–1.03 | 0.002 |
| 2003, n = 451 | | | |
| Age | 1.12 | 1.078–1.17 | 0.001 |
| Buffer capacity | 0.45 | 0.26–0.77 | 0.004 |

The results of the multiple logistic regression (Table 6) showed that, in 1983, after adjusting for age, gender and socio-demographic variables, there was a statistically significantly higher risk that men vs women and employed vs unemployed individuals would have more DFS and there was also a significant association between DFS and smoking. In 1993, there was a higher risk that men vs women, individuals with a low education vs a high education and employed vs unemployed individuals would have more DFS, as well as an association between DFS and PLI. In 2003, there was no statistically significant association between DFS and gender or DFS and socio-demographic variables, but an association between DFS and buffer capacity was seen (lower risk).

Study III

Demographic data. The age of the snus users was 42.3 ± 8.7 (mean \pm SD) while that of non-tobacco users was 42.3 ± 8.3 years. They had visited a dentist for an oral examination 1.9 ± 2.4 and 1.5 ± 2.2 years ago respectively. All the participants but one stated that they used fluoride toothpaste daily or twice daily. Among the snus users, 78 individuals had a “high” and 22 a “low” educational level. The corresponding figures among non-users were 83 and 18 respectively (no statistical analyses were carried out).

Table 7. Mean and \pm SD of number of teeth, secretion rate, gingival index (GI) in Study III. Only significant variables are given.

| <i>Number of individuals</i> | | | |
|--------------------------------|-------|-----------------|----------------|
| | Males | Females | |
| Snus users | 93 | 7 | |
| Non-users | 93 | 8 | |
| | | <i>Mean</i> | <i>p-value</i> |
| <i>Number of teeth</i> | | | |
| Snus users | 100 | 27.1 ± 1.4 | 0.010 |
| Non-users | 101 | 27.5 ± 0.9 | |
| <i>Secretion rate (ml/min)</i> | | | |
| Snus users | 98 | 2.5 ± 0.8 | 0.005 |
| Non-users | 101 | 2.2 ± 0.8 | |
| <i>GI (%)</i> | | | |
| Snus users | 100 | 20.4 ± 18.2 | 0.009 |
| Non-users | 101 | 14.4 ± 13.9 | |
| <i>GI upper front (%)</i> | | | |
| Snus users | 100 | 14.9 ± 20.6 | 0.003 |
| Non-users | 101 | 7.7 ± 11.9 | |

Clinical data. Table 7 shows the number of females and males and the mean \pm SD number of teeth, salivary secretion rate and GI. Only statistically significant variables are given. Snus users had significantly fewer teeth than non-users, even if the numerical mean difference was small (27.1 vs 27.5). For DFS, there was no significant difference, neither for the whole dentition nor for the upper front area (teeth 13-23), between the groups. There were no statistically significant differences between the groups in terms of initial and manifest caries. The numbers of tooth surfaces with secondary caries were 14 for snus users and 17 for non-users.

Regarding GI, snus users had higher values than non-users. In the upper front area (teeth 13-23), the trend was the same. In terms of PI, there was no statistical significant difference between the two groups.

The mean salivary secretion rate for snus users was higher than for non-users. However, no significant difference was found for buffer capacity or for the number of MS and LB in saliva. There was no statistically significant difference between the test and control group regarding toothbrushing habits and approximal cleaning with tooth picks and an interdental brush (data not shown). However, the use of dental floss was more frequent among non-users ($p=0.001$). A larger number of snus users ($n=23$) did not have any snacks between meals compared with non-users ($n=10$; $p=0.049$). Fewer snus users ($n=18$) stated that they had cookies and buns two to three times a week compared with the controls ($n=44$) and a smaller daily intake between main meals ($n=6$ vs. 12 ; $p<0.001$). There was no significant difference in terms of the intake of candy, sweets and soft drinks between the two groups (data not shown).

Cariogram and plaque pH data. The mean Cariogram value, expressed as the “chance of avoiding new lesions in the near future” was $65.4 \pm 16.2\%$ for snus users and $64.5 \pm 16.4\%$ for non-users (NS). The measurement of plaque pH showed that non-users had a somewhat more pronounced drop than snus users (Fig. 1), but this difference (based on AUC; area under the curve) was not statistically significant. When the users placed a portion of snus under their lip

and then rinsed with sucrose, the pH fall was less than if no snus was present in the mouth ($p < 0.001$).

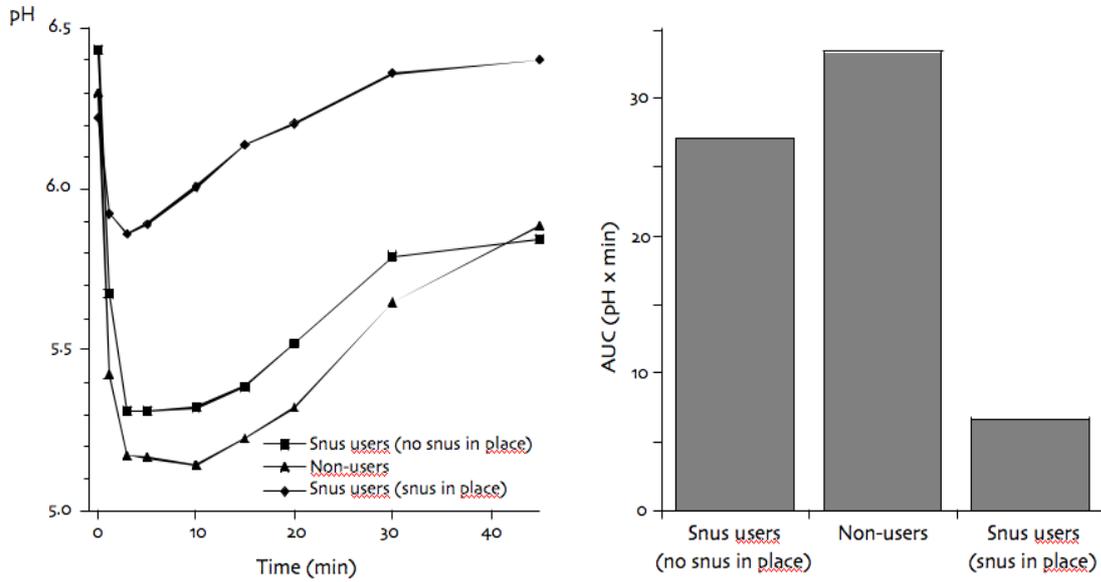


Figure 1. Mean plaque pH and area under the curve (AUC) after rinsing with 10% sucrose in snus users and non-users (10 subjects/group). The snus users repeated the sucrose rinse with a portion of snus present in the mouth.

Study IV

The subjects had a mean salivary secretion rate of 2.9 ± 2.2 ml/min, high levels of buffer capacity and medium numbers of mutans streptococci and lactobacilli (4.2 ± 1.4 and 3.0 ± 1.2 respectively). The mean plaque pH values are shown in Fig. 2.

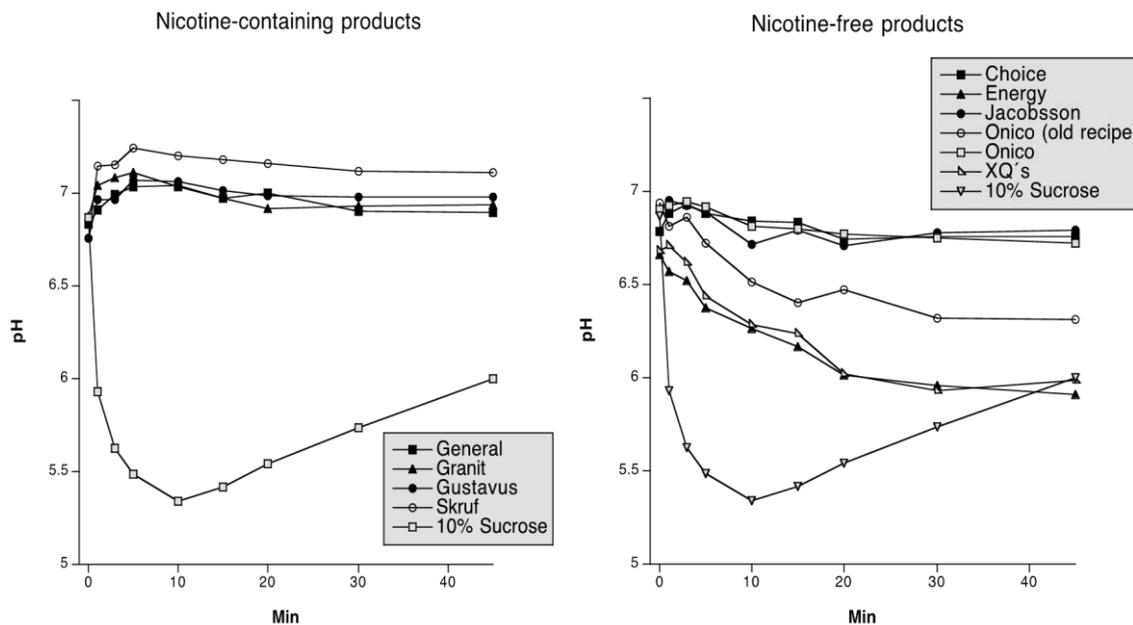


Figure 2. Mean values for pH in plaque of the different nicotine-containing and nicotine-free snuff products. Mean values for three sites and ten subjects.

Three of the six nicotine-free products reduced the plaque pH values, with the final pH ranging from 5.7 to 6.0, which was almost identical to the results found with 10% sucrose. The other three nicotine-free products had only a minor effect on plaque pH. On the other hand, all four nicotine-containing products increased the pH during the whole 45-min test period, with the final pH varying between 6.8 and 7.2. A wide variation in AUC/AOC was found in the 11 tests. The control (10% sucrose) resulted in the largest AUC area, followed by three of the nicotine-free products. The statistical analyses showed that all four nicotine-containing products differed significantly from the six nicotine-free products and that all ten of these snus products (4+6) differed significantly from the sucrose control ($p < 0.001$). There were no statistical differences in the AOC among the four nicotine-containing products. When comparing the AUC for the six

nicotine-free products with each other, Energy, XQ's and Onico old recipe differed from the other three products ($p < 0.01$ or $p < 0.001$).

Table 8. Concentration (%) of starch, glucose, fructose and sucrose in the six nicotine-free and the four nicotine-containing products

| | Starch | Glucose | Fructose | Sucrose | Total sugar |
|-------------------------------------|--------|---------|----------|---------|-------------|
| Nicotine-free products | | | | | |
| <i>Choice Original Pepper</i> | 2.5 | 0.3 | 0.4 | 0.5 | 1.2 |
| <i>Energy</i> | 26.0 | 1.7 | 2.3 | 0.1 | 4.1 |
| <i>Jakobsson Classic</i> | 2.3 | 0.1 | 0 | 0.9 | 1.0 |
| <i>Onico (old recipe)</i> | 8.9 | 0.3 | 0.3 | 0.1 | 0.7 |
| <i>Onico</i> | 1.2 | 0 | 0 | 0 | 0 |
| <i>XQ's</i> | 26.0 | 0.5 | 0.1 | 5.9 | 6.5 |
| Nicotine-containing products | | | | | |
| <i>General Original Portion</i> | 1.5 | 0.2 | 0.3 | 0 | 0.5 |
| <i>Granit White</i> | 1.6 | 0.4 | 0.4 | 0.2 | 1.0 |
| <i>Gustavus Original Portion</i> | 1.5 | 0.3 | 0.3 | 0.1 | 0.7 |
| <i>Skruf Original Portion</i> | 1.5 | 0.2 | 0.3 | 0 | 0.5 |

The carbohydrate content of the 10 products is shown in Table 8. The nicotine-free snus (apart from the new formula of Onico) contained fermentable carbohydrates up to 6.5% sugars and 26% starch (XQ's). The four nicotine-containing products contained only traces of glucose, fructose and sucrose (varying from 0.5 to 1.0%).

Discussion

This thesis is based on four separate studies with the aim of studying some effects of the use of tobacco on oral health. The first (Study I) is an epidemiological survey, describing tobacco use in a Swedish population, changes over time and relationships with certain personality aspects and dental care habits. The second study (Study II) is based on the same population as Study I and had the aim to evaluate some intra-oral caries-associated variables and the effect of tobacco use on dental caries. According to Study I, there was a reduction in the number of smokers and an increase in the number of snus users over time and tobacco users stated that they did not visit a dentist as regularly as non-tobacco users. The toothbrushing frequency and inter-proximal cleaning was not as good in tobacco users as in non-users. The results of the epidemiological studies performed in 1993 and 2003 (Study II) indicated that daily smoking and snus use did not influence the prevalence of dental caries.

In Studies III and IV, the focus changed from epidemiology to a clinical approach in order to study the effect of etiological factors of snus use on caries in more detail. In Study III, no statistically significant difference in caries prevalence between snus users and non-users was found and there were only minor differences regarding other caries-associated factors between the two groups. Finally, in Study IV, the effect of nicotine-free and nicotine-containing snus on plaque pH in vivo was compared. The pH measurements showed that the four nicotine-containing products increased the pH, while three of the six nicotine-free products lowered it.

Data from epidemiological studies can be used to illustrate a number of important subjects, such as the incidence of dental disease, and to identify and analyse disease determinants and groups risking a deterioration in oral health. The results of the current two epidemiological studies (Studies I and II) have been used to provide an overall picture of the tobacco user as a person in relation to the non-tobacco user in terms of socio-economic conditions, personality aspects and some dental care habits, frequency of dental visits and oral status. The results are based on three cross-sectional epidemiological studies, in 1983, 1993 and 2003, of random selections of the population in Jönköping a medium-sized Swedish city. This comparison thus covers a time period

of 20 years and the cohorts represent the historical interplay of all the different events that occur during life. The non-response rate varied between 28% and 32% (15 to 70 years of age). The material consisted mainly of Swedish-born individuals and the percentage of immigrants was about 10%. There were many reasons for not taking part in the studies, none of which was likely to have a major influence on the results.

One important question that always arises is whether the study population can be regarded as representative of a population larger than that studied. These questions have already been discussed in detail (Hugoson et al., 2005a). The conclusion was that, when comparing the results of the Jönköping studies with those of other similar Swedish epidemiological studies, performed in about the same year of examination, no important differences in oral clinical variables were found.

There are always many inherent difficulties associated with performing epidemiological studies, not least handling several examiners and a large number of participants. It is also important to consider that, in epidemiological studies, where the variables are expressed as mean values, a minority of the population with a higher disease prevalence than the rest of the population will always be present and undetected. It is also important to remember that only associations between studied variables can be found and they should not be interpreted as evidence of a causal relationship.

A distinct trend was found in Study II where the total number of tobacco users decreased over time. However, it is especially important to note that this is a decrease among smokers. This is also in accordance with national surveys, where the same pattern has been shown. The smoking rates in Sweden have declined considerably during the last few decades. The prevalence of daily smokers in 2011 was 11% compared with 14% in 2004, which is among the lowest rates in Europe (Statens folkhälsoinstitut, 2012; Shafey et al., 2012). The reduction in the number of smokers can be due to different reasons. Many people are giving up smoking and fewer and fewer people are starting to smoke. Most smokers quit smoking without starting with another tobacco product such as snus, according to a Swedish follow-up study (Lundqvist et al., 2009).

Another reason could be that smoking is not accepted by society as it used to be and some smokers start to use snus instead of smoking, thereby simply changing to another product which produces a similar nicotine dependence. Approximately 50% of snus users are former smokers, according to a review (Foulds et al., 2003). There are some tobacco users who use both snus and cigarettes, so-called “dual users”. During the study years between 1990 and 2007, in a middle-aged population in the north of Sweden, the prevalence of dual users among men decreased. But among women, an increasing percentage could generally be seen (Norberg et al., 2011). However, dual users were excluded in Study II and were not represented at all among the participants in Study III in this thesis.

There is a trend in the community for the gaps between genders according to lifestyle to become smaller and smaller. The use of snus has increased, especially among men and in certain age groups, but also among women, from previously having been a traditionally male habit. The reason for this could be due to some smokers switching to snus. The prevalence of female smokers decreased from 19% in 2004 to 12% in 2011 (Statens folkhälsoinstitut, 2012). However, an increase among male and never-smokers with current snus use was shown. Among female snus users, former smoking dominated (Norberg et al., 2011). The reason why some choose snus instead of cigarettes could be that it has not been proved so clearly that snus affects the general health as much as smoking does (Lee, 2011). Another explanation could be that the use of snus is more accepted in the community from an environmental perspective. In 1993, the Swedish Tobacco Act was introduced and this might have affected the use of tobacco, especially the smoking prevalence (SFS 1993:581,). In 2003, the Swedish parliament adopted new national public health objectives, which might have influenced the use of tobacco in Sweden. Four targets relating to the use of tobacco have been defined: they are a tobacco-free start to life, reducing the start of tobacco use among individuals less than 18 years of age, reducing the number of smokers by 2014 and reducing the exposure to tobacco smoke. In the present epidemiological studies (Studies I and II), there were some changes between the study years in terms of gender. In 1983, there were no smokers among males in aged 15, but 9% of the females was smokers. Among 20-year-old women, 49% stated that they were smokers and 29% of males in the same age group were using snus. In 2003, 26% of males were using snus and the same

number (26%) of females were smoking in the 20-year age group. No statistical differences could be shown in terms of tobacco use and marital status in 1983 and 1993, but, in 2003, it was found that the use of tobacco was higher among single men.

The use of tobacco and certain food products, as well as oral hygiene, are examples of habits associated with lifestyle. Unfavourable lifestyle characteristics, such as a risky high consumption of alcohol and a low intake of fruit and vegetables, are known to be associated with smoking and the use of snus, according a public health survey from 2010 (Engström et al., 2010). That study also showed that overweight was related to snus use as well as dual use. These lifestyles habits are also confirmed in a study in which smokers did not visit a dentist as regularly as non-smokers and also had a low intake of vegetables and fruit (Mucci et al., 2001). This was also confirmed in Study III, where snus users did not visit a dental practice as often as non-users.

The different use of the health service suggests that smokers may have different attitudes to dental health compared with non-smokers and this could be translated as neglecting the prevention and treatment of dental caries. However, in Study III, data from the questioner showed that snus users had a less frequent intake between meals. This could be seen as contradicting to the results presented by other researchers, showing that snus users have a higher BMI than non-users (Hansson et al., 2009; Travier et al., 2009).

In Study I, there was a statistically significant difference, between tobacco users and non-tobacco users when it came to the frequency of daily toothbrushing. Oral hygiene habits were also studied in Study III, where there was no statistically significant difference in toothbrushing frequency between snus users and non-tobacco users, but the use of dental floss was more common among non-users. Moreover, Study I showed that tobacco users used toothpicks less frequently than non-users. Several studies have shown that good oral health habits, such as using dental floss or toothpicks, improve or maintain, oral health (Axelsson et al., 1978; Axelsson et al., 2004). In the present Study III, it was shown that snus users had statistically higher rates of GI, compared with non-users. The most probable reason for this is the use of snus.

Cross-sectional studies, like Study II, will only provide a snapshot of lifestyle and oral status and therefore have limitations. Information is missing about the length of time the different individuals have been using tobacco and the age at which they started their tobacco habits. Smokers smoked 10 cigarettes a day or more. Snus users used snus both as loose snus and as snus in portion bags. In Study II, the daily use of snus was a better way of describing the consumption of snus than the number of boxes. However, in Study III, the participants were asked how long they had been using snus and more than 50% of the individuals stated that they had been snus users for more than 20 years. Of these, approximately 60% had been using snus for more than 25 years, which shows that the participants were habitual snus users.

The prevalence of oral diseases, and dental caries in particular, has changed markedly during the last 40-50 years (Hugoson et al., 2005b). This decrease is very obvious in children and adolescents, but there is also a considerable improvement in oral health within the adult population. The present series of epidemiological studies, 1983, 1993 and 2003, from Jönköping, demonstrate a decrease in the number of edentulous individuals, an increase in the number of natural teeth and an improvement in oral hygiene and caries status. These results are also mirrored in the present description of non-tobacco users and tobacco users in terms of the number of teeth, PLI and DFS.

When it comes to dental caries in relation to the use of snus, only a few studies have so far been published (Vellappally et al., 2007; Skudutyte-Rysstad et al., 2009). Swedish snus is different from smokeless tobacco in other countries. Swedish nicotine-containing snus products generally contain negligible amounts of fermentable carbohydrates and have a high pH value and a low level of tobacco-specific nitrosamines (TSNA) (Foulds et al., 2003; Digard et al., 2009). One interesting observation in Study II was that snus users had a higher buffer capacity than smokers. When snus is used for many hours a day, as is the case for most snus users (Rolandsson et al., 2005), the pH in the oral cavity can be expected to be elevated. This may favour the remineralisation of the tooth surfaces and the inhibition of the cariogenic, acidic and aciduric microflora.

In Study III, there were only minor differences in caries-associated variables between the groups and there could be different explanations for this, such as high educational levels. The study was carried out at a dental clinic situated at the university and the recruitment partly took place at the university, which might influence the samples. On the other hand, the two groups (snus users and non-users) were as similar as possible in terms of educational level, gender and age. Another reason could be that all the subjects, but one stated that they used fluoride toothpaste every day and this could have masked the active and or protective effect of snus.

In the present Study III, the Cariogram computer program was used to mirror the caries risk profile of snus users and non-users. This is an interactive method for illustrating caries-related factors and the caries risk profile has been used in children, adults and the elderly. The program was introduced as a model to identify risk factors and it has been evaluated in two large longitudinal studies. The Cariogram can easily be used as part of the daily routine at the dental clinic and the model can also be seen as a tool for motivating the patient, as well as acting as a support for selecting preventive treatment for each individual. The program can also be downloaded free of charge in different languages. It does not, however, include social factors and these factors therefore need to be considered separately when constructing the Cariogram (Hänsel Petersson et al., 2002; Hänsel Petersson et al., 2003; Bratthall et al., 2005). In Study III, there were only minor differences in the mean value relating to the “chance of developing new carious lesions in the near future”, which goes hand in hand with the clinical data.

At the beginning of this millennium, nicotine-free snus was introduced in the Swedish market and one case showing root surface caries at the location where the product is placed was reported (Hansson et al., 2008). The nicotine-free and nicotine containing products which were tested in Study IV were all bought on the open market and they were stored in a refrigerator during the test period to keep them fresh. However, there were some difficulties regarding the recruitment of volunteers, all of whom had to be snus users for ethical reasons. Each participant had to make a total of eleven appointments. Moreover, they were asked not to brush their teeth for three days, which was unacceptable for some individuals. They also had refrain from eating, drinking and using snus two hours before the appointment, which was also was a reason for refusing to participate, especially among the snus users that used snus for most of their waking hours.

Plaque pH is an important variable in the caries process and the pH measurement in Study IV showed that the nicotine-containing snus raised the pH in plaque while some of the nicotine-free products lowered it. This was a single exposure to the plaque acidogenicity and, if this was tested in a longitudinal study, it might produce a different result. The pH method is well accepted in caries research for testing various food products and for evaluating the cariogenicity of the oral microflora (Lingström et al., 1993; Aranibar Quiroz et al., 2003).

New tobacco products are constantly appearing on the market and it is not easy for consumers to stay updated on the contents and the way these different products influence oral health or general health. The results of Study IV showed that three of the six nicotine-free products clearly reduced plaque pH but there are still nicotine-free products that do not change the plaque pH and do not harm the tooth surface from a cariological point of view. From a general health perspective, the nicotine-free snus could be a better choice when there is no nicotine content and thereby no dependence, as compared with nicotine-containing snus. Dental hygienists and dentists could be seen as a support or guide for the patients with their knowledge of these products from both a cariological and an oral health perspective.

Conclusions

The main conclusions from this thesis were:

Study I: During the 20-year study period (1983-2003), there was a reduction in the number of smokers and an increase in the number of snus users. Tobacco users had less frequent dental visits and poorer oral hygiene habits than non-tobacco users.

Study II: The results of the cross-sectional epidemiological studies, performed in 1983, 1993 and 2003, indicate that daily smoking or use of Swedish snus does not appear to increase the risk of dental caries.

Study III: This clinical study carried out in 2009-2011 showed only minor or no differences in caries and related factors between daily snus users and non-users.

Study IV: All tested nicotine-containing snus products increased the plaque pH, in contrast to three of the six nicotine-free products, which lowered the pH.

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