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EROSIVE TOOTH WEAR AND ASSOCIATED FACTORS IN NORTHERN FINLAND BIRTH COHORT 1966

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Abstract

The aim of this study was to investigate the status of erosive tooth wear (ETW) in Finnish middle-aged adults and its association with dental caries and sociodemographic and intrinsic factors. In addition, we performed a genome-wide association study (GWAS) to identify whether genetic polymorphism (single nucleotide polymorphism) could explain some of the individual variance in the ETW status. Another aim of the study was to validate the use of the erosion index, the Basic Erosive Wear Examination (BEWE), on 3D models.

Of the total Northern Finland Birth Cohort (NFBC1966), a convenience sample of 3,181 people was invited for an oral health examination in 2012–2013, of whom 1,962 participated, thus comprising the study group for the present study. ETW was assessed by sextants using the BEWE index. The clinical data was supplemented by information collected by means of postal questionnaires in 1997–1998 and 2012–2013, blood samples, and 3D models of the dentition. Of those clinically examined, 586 participants were randomly selected for the validation study of the BEWE index on 3D models.

ETW was a common finding among the Finnish adult population, and almost half of the population needed at least preventive measures against the condition, and almost one in ten had severe ETW. Male gender and restorative treatment need due to dental caries were associated with ETW, unlike sociodemographic factors. Of the intrinsic factors, daily reflux symptoms and hyposalivation were the most significantly associated with severe ETW. According to the results from the GWAS, susceptibility to ETW could be partly explained by genetic polymorphism. The BEWE index was found reliable for recording ETW clinically and on 3D models, and 3D models were especially sensitive in detecting initial ETW.

In conclusion, ETW seems to be common among Finnish adults, especially among males. In addition to risk factors, individual susceptibility should be kept in mind when assessing the risk for the condition. Early diagnosis of ETW is important in maintaining good oral health, and the BEWE seems to be a reliable index for that purpose both clinically and on 3D models.

Keywords: 3D models, adults, dental caries, dental erosion, epidemiology, gene research, sociodemographic factors, tooth wear, validation

Alaraudanjoki, Viivi, Erosiivinen hampaiden kuluminen ja siihen liittyvät taustatekijät Pohjois-Suomen vuoden 1966 syntymäkohortissa.
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Tiivistelmä

Tutkimuksen tavoitteena oli selvittää hampaiden erosiivisen kulumisen yleisyyttä ja vakavuutta suomalaisilla keski-ikäisillä aikuisilla, sekä erosiivisen kulumisen yhteyttä hampaiden reikiintymiseen sekä sosiodemografisiin ja sisäisiin tekijöihin. Lisäksi selvitettiin koko genomin kartoitusta hyödyntäen, voidaanko geneettisellä polymorfismilla (yksittäisillä emäsparin vaihdoksilla) selittää yksilön alttiutta erosiiviselle kulumiselle. Tutkimuksen tavoitteena oli myös validoida erosiivista kulumista arvioiva indeksi (The Basic Erosive Wear Examination, BEWE) 3D-malleilla.

Koko Pohjois-Suomen vuoden 1966 syntymäkohortista kutsuttiin 3 181 henkilöä suun terveystarkastukseen vuosina 2012–2013. Kutsutuista 1 962 osallistui tutkimukseen muodostaen lopullisen tutkimusjoukon. Erosiivista kulumista arvioitiin käyttäen BEWE-indeksiä. Kliinistä tutkimusta täydensivät vuosina 1997–1998 ja 2012–2013 tehdyt postikyselyt, verinäytteet ja hampaiston 3D-mallit. Kliinisesti tutkituista osallistujista yhteensä 586 henkilöä valittiin satunnaisesti BEWE-indeksin validointitutkimukseen 3D-malleilla.

Erosiivinen kuluminen oli yleistä suomalaisilla aikuisilla, ja lähes puolella oli erosiivista kulumista, joka vaatisi vähintään ennaltaehkäiseviä toimia. Vakavaa erosiivista kulumista on lähes joka kymmenennellä. Miessukupuoli ja korjaavan hoidon tarve hampaiden karioitumisen vuoksi olivat yhteydessä erosiiviseen kulumiseen, toisin kuin sosiodemografiset tekijät. Sisäisistä tekijöistä päivittäiset reflux-oireet ja vähäinen syljeneritys olivat vahvimmin yhteydessä vakavaan erosiiviseen kulumiseen. Koko genomikartoituksen perusteella vaikuttaa siltä, että alttius erosiiviselle kulumiselle saattaa selittyä osittain geneettisellä polymorfismilla. BEWE-indeksi näyttää olevan luotettava menetelmä niin kliinisessä arvioinnissa kuin arvioitaessa erosiivista kulumista 3D-malleilla. Alkava erosiivinen kuluminen oli helpommin havaittavissa 3D-malleilta kliiniseen arviointiin verrattuna.

Tutkimuksen perusteella voidaan sanoa, että erosiivinen kuluminen on yleistä suomalaisilla aikuisilla, etenkin miehillä. Jo tiedettyjen riskitekijöiden lisäksi yksilöllinen alttius erosiiviselle kulumiselle tulisi pitää mielessä riskikartoitusta tehdessä. Erosiivisen kulumisen aikainen diagnosointi on tärkeää hyvän suun terveyden ylläpitämiseksi, ja BEWE-indeksi vaikuttaa soveltuvan diagnosointiin niin kliinisesti kuin 3D malleillakin.

Asiasanat: 3D-mallit, aikuiset, epidemiologia, geenitutkimus, hampaiden eroosio, hampaiden kuluminen, karies, sosiodemografiset tekijät, validointi

To Eino, Veera and Ilkka

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26th November Viivi Alaraudanjoki

Abbreviations

AMBN	Ameloblastin
AMELX	Amelogenin, X-linked
BEWE	Basic Erosive Wear Examination
CAD-CAM	Computer-Aided Design and Computer-Aided Manufacturing
CDH4	Cadherin 4
CI	Confidence Interval
D	Number of Decayed Teeth
DCP1A	Decapping mRNA 1A
DE	Dental Erosion
DMFT	Number of Decayed, Missing and Filled Teeth
F	Number of Filled Teeth
F2R	Coagulation Factor II thrombin Receptor
FGF1	Fibroblast Growth Factor 1
FGFR1	Fibroblast Growth Factor Receptor 1
FBXO8	F-box protein 8
FOTI	Fiber-Optic Transillumination
GERD	Gastro-Esophageal Reflux Disease
GLRA3	Glycine Receptor Alpha 3
GWAS	Genome-Wide Association Study
ETW	Erosive Tooth Wear
ICDAS	International Caries Detection and Assessment System
ING1	Inhibitor of Growth family member 1
M	Number of Missing Permanent Teeth Due to Caries
MSX1	Msh homeobox 1
MYT1L	Myelin Transcription factor 1-like
NCBI	National Center for Biotechnology Information
NFBC	Northern Finland Birth Cohort
OR	Odds Ratio
PTG	Pantomography, a radiograph of the maxillary and mandibular dental arches and their contiguous structures
PXDN	Peroxidasin
RTN	Restorative Treatment Need
SCD5	Stearoyl-CoA Desaturase 5
SKAP2	Src Kinase Associated Phosphoprotein 2
SD	Standard Deviation

SNP	Single Nucleotide Polymorphism
TWI	Tooth Wear Index
VEDE	Visual Erosion Dental Examination
WHO	World Health Organization
ZWINT	ZW10 Interacting kinetochore protein
3D	Three-Dimensional

List of original publications

This study is based on the following publications, which are referred throughout the text by their Roman numerals:

I Alaraudanjoki V, Laitala ML, Tjäderhane L, Pesonen P, Lussi A, Anttonen V (2016) Association of erosive tooth wear and dental caries in Northern Finland Birth Cohort 1966 – an epidemiological cross-sectional study. BMC Oral Health 17(1):6: doi: 10.1186/s12903-016-0232-x

II Alaraudanjoki V, Laitala ML, Tjäderhane L, Pesonen P, Lussi A, Ronkainen J, Anttonen V (2016) Influence of intrinsic factors on erosive tooth wear in a large scale epidemiological study. Caries Research 50: 508-516.

III Alaraudanjoki V, Koivisto S, Männikkö M, Pesonen P, Laitala ML, Leinonen J, Tjäderhane L, Anttonen V (2018) Genome-wide association study on erosive tooth wear in Northern Finland Birth Cohort 1966, Manuscript

IV Alaraudanjoki V, Pesonen R, Saarela H, Laitala ML, Tjäderhane L, Kiviahde H, Lussi A, Anttonen V (2017) Is Basic Erosive Wear Examination (BEWE) suitable for recording erosive tooth wear on 3D models? Journal of Dentistry 59: 26-32.

Contents

1 Introduction

Erosive tooth wear (ETW) refers to the combined effect of chemical dissolution of tooth surface caused by acids and mechanical wear of the acid-softened surface (Ganss & Lussi 2014). As the teeth wear, dentine becomes eventually visible, causing not only aesthetic problems but also sensitivity and pain. Since the morphology of teeth changes, the condition may also predispose to occlusal imbalance and even temporomandibular disorders. Previously considered as a simple condition, the conception of ETW has recently been changing towards a complex interplay between patient-related (e.g. eating/drinking frequency, habits, saliva, bruxism, and general heath) and nutritional factors (e.g. acid type, pH, buffering capacity), thus contributing to a better understanding of the condition.

The prevalence of ETW seems to be increasing (Tschammler *et al.* 2016), and the concern and interest in the condition among researchers and clinicians has steadily increased during the last two decades. However, systematic prevalence data and incidence studies are scarce (Jaeggi & Lussi 2014). In Finland, current data on the prevalence or severity of ETW is vague (Bartlett *et al.* 2013).

Since erosive damage may compromise the health of the patient's dentition for the remaining lifetime and may require complex and expensive rehabilitation of the dentition, a proper diagnosis at an early stage of the condition is of outmost importance. A well-known fact, however, is that diagnosing mild erosive wear can be challenging because usually no clinically distinct, pathologic changes have yet occurred in that stage. Even though there are many indices developed for assessing (erosive) tooth wear, no internationally accepted index exists. To fulfill this need, the Basic Erosive Wear Examination (BEWE) index was developed by the best experts in the field a decade ago (Bartlett *et al.* 2008). The criteria have become an easily adapted tool for clinical practice. There are, however, only a few studies validating the index so far, and studies on its use on dental casts or 3D models do not exists. Furthermore, there are no studies on the use of 3D models as an additional tool in erosive wear diagnostics, although 3D models are widely used in modern clinical practice in prosthodontics, orthodontics, and implantology.

Even though pure erosion does not occur without extrinsic (diet) or intrinsic (regurgitated gastric acids) acids, the conception of the etiology is being expanded today. Susceptibility to the condition varies between individuals and is affected by a number of different variables, some of which remain unknown. Considering intrinsic and extrinsic factors, a study by Holbrook *et al.* (2009) found that only approximately 15% of the variance in the prevalence and severity of wear could be explained by intrinsic or extrinsic acidic challenges. Recently, it was discovered that not only the saliva and pellicle differ between individuals but the differences in the enamel may also play a role in the susceptibility to ETW (Uhlen *et al.* 2016a). Concerning this individual variation, it could be hypothesized that genetic polymorphism (single nucleotide polymorphism) may also play a role in the susceptibility to erosive wear.

The main aim of this study was to evaluate the prevalence and severity of ETW among the Northern Finland Birth Cohort 1966, as well as to analyze the role of associated factors (sociodemographic factors, dental caries, and conditions causing gastric acids entering the mouth). Part of the study focused on investigating genetic polymorphism in relation to ETW. Furthermore, the study also aimed to analyze the reliability of the BEWE index on 3D models and clinically, and the use of 3D models in ETW diagnostics.

2 Review of the literature

2.1 Definition of erosive tooth wear and the change in understanding

Tooth wear is conventionally categorized into dental erosion, attrition, and abrasion. Dental erosion refers to the loss of tooth surface due to chemical dissolution of enamel or dentine caused by acid insults, with the acid being mostly of nonpathological origin (Ganss 2014). The destructive effects have been thought to involve only the tooth surface, but recently it has been shown that erosive dissolution also occurs within the thin, partly demineralized and softened enamel layer (near-surface demineralization), thus leaving the tooth surface vulnerable to mechanical forces. These forces, mainly due to attrition or abrasion, remove the softened tooth surface, causing and exacerbating tooth wear (Shellis *et al.* 2013). In short, ETW is a combination of chemical and mechanical tooth wear; however, the initiation of wear is purely due to acids of intrinsic origin or acids from the diet or in some cases from the environment.

2.1.1 Change in understanding

ETW is becoming increasingly significant in the long-term management of the dentition. A few decades ago, erosive wear attracted little interest in dental research and clinical dental practice, and the studies concerning the condition were vague. However, when searching PubMed with words “dental erosion” OR “erosive tooth wear”, a total of 1,257 studies were published between the years 2010 and 2016, showing a significant increase in the interest towards ETW. The interest in tooth wear processes and erosion is not only scientific. As people live longer and their teeth persist for the entire life span, in addition to dental caries, tooth wear of different origin is becoming a daily concern in the clinical practice. Consequently, dental professionals need new methods to diagnose (erosive) tooth wear in its initial stages, as well as an overall understanding of the condition and new strategies to arrest its progress. On the other hand, skilled restorative techniques are needed to provide the best available treatment for patients with advanced tooth wear.

2.1.2 When is (erosive) tooth wear considered pathological?

Teeth are exposed to a variety of chemical and physical attacks during their life span, and therefore, tooth wear *per se* is mostly regarded as an inevitable, physiological condition (Ganss 2014). There have been several attempts to distinguish physiological and pathological tooth wear, but classifying the condition is ambiguous and currently open for debate. In some definitions, tooth wear is even considered an evolutionary adaptation and human dentition is described as "designed" on the premise that extensive wear will occur (Kaifu *et al.* 2003). It appears that the difference between physiological and pathological tooth wear is dependent on the concepts of health and disease. The classic definition in modern dentistry according to Smith and Knight (1984) is:

> *"Tooth wear can be regarded as pathological if the teeth become so worn that they do not function effectively or seriously mar the appearance before they are lost due to other causes or the patient dies. The distinction of acceptable and pathological wear at a given age is based upon the prediction of whether the tooth will survive the rate of wear."*

This definition is still considered valid and used in daily practice when assessing the need for treatment. Tooth wear of different origins is listed in the International Classification of Diseases (World Health Organization 2016)

2.2 Etiology of erosive tooth wear

Causes for dental erosion are traditionally categorized into acids of extrinsic or intrinsic origin. It has been reported that, in particular, the chemical properties of acids and the frequency of acidic challenges play an important role in the pure erosion process (Lussi & Hellwig 2014). It is important to note that there is no fixed critical pH value concerning the erosive tissue loss, but the critical pH varies according to the properties of the erosive solution itself (Lussi & Carvalho 2014). According to recent studies, risk indicators for ETW include frequent or high consumption of acidic drinks and dietary products (El Aidi *et al.* 2011, Søvik *et al.* 2015a), acidic reflux (Bartlett *et al.* 2013, Holbrook *et al.* 2009), and hyposalivation (Hara & Zero 2014). A recent meta-analysis examining extrinsic acids showed that carbonated drinks, acid snacks/sweets, and naturally acidic fruit juices increase the occurrence of erosion (Salas *et al.* 2015).

Considering the intrinsic factors, the most common general health condition in which intrinsic acids may enter the oral cavity is gastroesophageal reflux disease (GERD) (Moazzez & Bartlett 2014). The Montreal definition of GERD (Vakil *et al.* 2006) describes it as a condition that develops when the reflux of stomach contents causes troublesome symptoms and/or complications, and identifies dental erosion as one possible GERD complication. Other conditions that may cause gastric acids to enter the oral cavity include eating disorders, alcohol abuse, and pregnancy-related problems. A recent review (Schlueter & Tveit 2014) concluded that ETW appears to be more severe among individuals suffering from problems with GERD, alcohol abuse, or eating disorders than in healthy controls. However, the number of studies considering all of these conditions is limited, and the burden of intrinsic factors in erosive wear process is not well established epidemiologically.

To summarize, it seems unlikely that one or two isolated factors are responsible for a multifactorial condition like ETW. Instead, an interaction between chemical, mechanical, and biological processes seems crucial (Figure 1). For example, tooth grinding may have a bigger role in erosive lesions than previously has been suspected (El Aidi *et al.* 2011). In addition, there seems to be individual differences in susceptibility to ETW, for example, due to saliva secretion and its associated properties (Hara & Zero 2014, Hellwig *et al.* 2013, Moazzez *et al.* 2004).

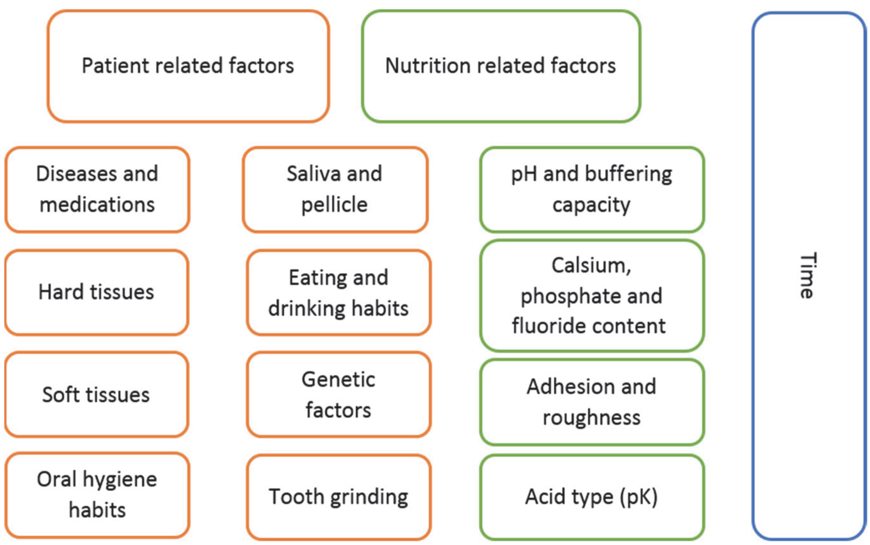

Fig. 1. Different factors playing a role in the onset and severity of the ETW process.

2.2.1 The association between erosive tooth wear and dental caries

Some of the risk indicators of ETW are consistent with dental caries, such as snacking habits, frequent use of sugar-containing soft drinks and acidic candies. Furthermore, individuals with erosion have been suggested to possess salivary characteristics similar to those of caries-active individuals (O'Sullivan & Curzon 2000). Since both processes are in the end caused by acids and comprise demineralization of dental hard tissues, it can be asked if ETW is associated with a higher need for caries treatment, or vice versa. The number of studies concerning the issue is scarce, and the findings in the literature on the issue remain equivocal. Many studies have reported a statistically significant association between ETW and dental caries (Isaksson 2013, Kazoullis *et al.* 2007, Mulic *et al.* 2013), while others have not (Auad *et al.* 2009, Truin *et al.* 2005).

2.3 Diagnosis of erosive tooth wear

> *"Diagnosis is the intellectual course that integrates information obtained by clinical examination of the teeth, use of diagnostic aids, conversation with the patient and biological knowledge."*

The above definition (Kidd *et al.* 2003, p. 111) was originally formed on the diagnosis of dental caries, but it is also valid for ETW. When considering erosion, the definition means that first, the clinician must know the normal variance in tooth anatomy and morphology to be able to do the comparison with the actual appearance. Second, the clinician must be able to use diagnostic measures to classify the disease, and then, based on a conversation with the patient, find out the possible symptoms and existence of etiological factors.

Clinically, in the initial stages of ETW, the tooth surface turns smooth, silky shining and dull. As the tissue loss continues, the morphology of the tooth changes: cusps become rounded and the appearance of the tooth seems melted, as is often described. Restorations can rise above the level of the adjacent tooth surface, and the incisal edges can turn almost transparent. In the end, the whole morphology of the tooth can disappear. On smooth surfaces, the width of the defect clearly exceeds the depth, which distinguishes erosive lesions from wedge-shaped defects. Apically from the smooth surface defect, there is often an intact border of enamel, likely due to plaque remnants or alkaline sulcular fluid. It has been proposed that the shallow defects coronal from the cemento-enamel junction are most likely pathognomonic for dental erosion, whereas the cupping of cusps and grooving of incisal surfaces can result from various chemical and physical influences (Ganss & Lussi 2014). However, "cupping" is traditionally one of the main findings in erosive dentitions.

It is important to distinguish erosive lesions from attrition and abrasion. Attrition is seen in patients with a tooth grinding habit, and it is often described as flat lesions with glossy areas and corresponding features in the antagonist teeth. Abrasion is due to extrinsic forces, such as excessive tooth brushing, and is often the main cause of buccal wedge-shaped defects. Considering occlusal surfaces, differentiation between erosion and abrasion is difficult since the lesions may be similar in shape (Ganss & Lussi 2014, Shellis & Addy 2014). In sum, as known from previous clinical and experimental studies, single individual wear mechanisms cause ETW extremely rarely, and instead, several mechanisms interact with each other (Shellis & Addy 2014).

2.3.1 Different indices of erosive tooth wear

From a clinical viewpoint, ETW is a surface phenomenon and requires a visual rather than a tactile approach. A huge number of different indices and their modifications have been developed, almost all of which are based on two indices, the Eccles index (Eccles 1979) and the Tooth Wear Index, (Smith & Knight 1984). Common for all these indices are the criteria used to differentiate erosive wear from other types of wear, as well as criteria used to describe the amount of hard tissue loss. In addition, many of the indices are based on dentine exposure, which has later been proven hard to assess correctly and reproducibly (Ganss *et al.* 2006) and which therefore does not give a full picture of the nature of ETW.

The Basic Erosive Wear Examination (BEWE) index was developed to avoid classifying lesions according to dentine exposure (Bartlett *et al.* 2008). In the four-step BEWE classification, the size of the lesion is quantified as an estimated proportion of the affected surface. Each surface of the tooth is examined and registered separately but recorded according to sextant. The most damaged tooth surface represents the BEWE score of the entire sextant (range 0 to 3). Score 0 means that there are no signs of erosive wear, whereas score 1 represents initial loss of surface texture. Score 2 means that erosive lesions are more pronounced and there is hard tissue loss in less than 50% of the surface area, and score 3 that hard tissue loss covers more than 50% of the surface area. The patient's BEWE score is the sum score of the sextants (range 0 to 18). The BEWE index has been developed by internationally recognized experts in the field, and it is intended to be an internationally accepted, standardized, and validated index and a simple tool for clinical practice (Bartlett *et al.* 2008). When BEWE is only used for assessing ETW, it is also important to list the criteria for erosion and for distinguishing erosion from abrasion and attrition.

In a recent in-depth analysis of the four most used indices (the Eccles index, the Tooth Wear Index, the Lussi index, and the BEWE index) (Wetselaar *et al.* 2016), it was argued that none of the indices met all the necessary qualifications for a hypothetical, broadly applicable tooth wear evaluation system. For BEWE, the shortcomings compared to a hypothetical index were that BEWE only evaluates chemical wear and may overestimate it, its suitability for assessing 3D scans has not been evaluated, and it has only been designed for partial assessment of dentition (sextants) and for cumulative use (BEWE sum score). It should be noted that almost all of these shortcomings are easy to overcome by making slight modifications to the index, and that BEWE is used (Bartlett *et al.* 2013) and is even validated (Olley *et al.* 2014) for tooth wear independent of origin. However, the authors concluded that it is not realistic that a single system is valid for all purposes.

To conclude, it appears that there is a need for standardization of indices, and especially previous indices should be reconsidered with respect to the validity of diagnostic criteria.

2.3.2 Reliability of the BEWE index in erosive tooth wear diagnostics

The BEWE index and the use of the cumulative BEWE score have been validated and shown to be acceptable for recording tooth wear and scoring severity in prevalence studies (Dixon *et al.* 2012, Mulic *et al.* 2010, Olley *et al.* 2014). Olley *et al.* (2014) have demonstrated that the sextant score and the BEWE sum score form a useful screening tool for assessing tooth wear without the need to record tooth wear on every tooth surface. It has been noted that the biggest problem when using BEWE is differentiating between intact enamel and initial loss of surface texture (Mulic *et al.* 2010), and therefore BEWE scores should be interpreted with some caution (Dixon *et al.* 2012). In addition, it has been suggested that the cut-off values need re-evaluation (Margaritis *et al.* 2011).

2.3.3 BEWE index applied to different methods

Concerning tooth wear assessment in general, conventional clinical examination has been suggested to be the least sensitive when compared to more sophisticated methods (Al-Omiri *et al.* 2013), including recording tooth wear on the basis of high-resolution 3D models. However, it has been shown that initial, non-occlusal wear is hard to distinguish, for example, on study casts (Wetselaar *et al.* 2009). The BEWE index was originally developed to be also suitable for grading dental casts (Bartlett *et al.* 2008), but to date, there are no studies evaluating the reliability of BEWE for assessing dental casts or digital 3D models. However, BEWE has been shown to be suitable for evaluating erosive wear on photographs (Hove *et al.* 2013). 3D models have been clinically used in orthodontics, oral surgery, and implant treatments for years, and they are expected to improve implementation of medical treatments and documentation of treatment outcomes (Baroudi & Ibraheem 2015). This technology could also be useful in tooth wear assessment and treatment planning, as well as monitoring. It has even been proposed that comparing sequential 3D models is the most accurate method for measuring tooth wear progression (DeLong 2006). However, no epidemiological studies concerning the use of 3D models in ETW assessment was found.

2.3.4 Assessment of progression rate

Assessment of the progression rate of ETW is important, as it determines whether current wear is active or inactive and whether there is a need especially for preventive treatment. Given that ETW usually develops over time, lifestyle factors present several years earlier may be responsible for the current state of ETW. Only a little information is available about clinical wear rates, and there is no scientific concept to assess the physiological wear rate, which both complicate the assessment of progression. Clinically, the signs for progression include frosty appearance and absence of staining (Ganss & Lussi 2014). In addition, absence of symptoms may indicate inactive tooth wear. Quantitative assessment of hard tissue loss over time is difficult without good quality study casts or photographs, which may be impractical in clinical use. For this reason, new diagnostic tools for chair-side use are needed. For example, metal markers bonded on tooth surface (Schlueter *et al.* 2005), applying ultrasound (Sindi *et al.* 2015), and optical coherence tomography (Wilder-Smith *et al.* 2009) have been introduced to help clinicians in decision making. It has been argued that intraoral scanning devices for digital impressions (3D models) could also be promising tools, however, these techniques have not yet been applied for monitoring ETW (Ganss & Lussi 2014).

2.4 Prevalence, incidence, and distribution of erosive tooth wear

Because of the numerous indices used for scoring ETW, comparison between prevalence studies is difficult. The number of epidemiological studies investigating the condition in adults is limited, and the study set-ups differ remarkably, complicating comparisons and drawing conclusions (Jaeggi & Lussi 2014). For long, it has been anticipated that the prevalence of ETW is increasing, but this tendency was only recently first shown among children (Tschammler *et al.* 2016). The most recent epidemiological studies in adult populations suggest a high prevalence of ETW (Table 2), however in many studies the severity of ETW is quite low (Chu *et al.* 2014, Fares *et al.* 2009, Struzycka *et al.* 2017, Vered *et al.* 2014). In Finland, the data on the prevalence of dental erosion dates back over two decades (Järvinen *et al.* 1988, Meurman *et al.* 1994), except for one recent multicenter tooth wear study (Bartlett *et al.* 2013). In that study, 17.7% of the Finnish participants had a maximum single BEWE score 2 or more, which was less than average (29.4%).

Table 1. Recent clinical studies among adolescents and adults, using (A) the BEWE index for analyzing ETW, (B) the BEWE index for analyzing tooth wear, or (C) some other index for analyzing ETW.

Authors	Country	Sample		ETW prevalence	Conclusion
		age	n	(BEWE>0)	
A					
Struzycka *et al.* (2017)	Poland	18	1,869	42.3%	Males had more ETW than females.
Provatenou *et al.* (2016)	Greece	14	263	79.0%	ETW was influenced by gender and diet.
Muller-Bolla *et al.* (2015)	France	14	339	56.8%	A consensus on the choice of index and age at assessment are needed.
Chu *et al.* (2015)	China	18–21	600	44.0%	Many had signs of DE, but very few signs of severe erosion.
Vered *et al.*(2014)	Israel	35–38	100 in	55.8%	The BEWE index is straightforward and comfortably accepted by the examinees.
		45–48	each	53.1%	
		55–60	group	61.9%	
Manaf *et al.* (2012)	Malaysia	19–24	150	68.0%	High prevalence of ETW.
B					
Wei *et al.* (2016)	China	35–49	720	67.5%	Prevalence of tooth wear and dentin exposure was high.
		50–74		100%	
Zhang *et al.* (2015)	China	15	360	89.4%	Tooth wear in adolescents should receive greater attention.
Bartlett *et al.* (2013)	[1]	18–35	3,187	57.1%	Major differences in the prevalence of ETW between countries.

Authors	Country	Sample		ETW prevalence	Conclusion
		age	n	(BEWE>0)	
C					
Mulic *et al.* (2016)	Norway	16	392	38%, VEDE	High prevalence and severity of DE among adolescents.
González-Aragón Pineda *et al.* (2016)	Mexico	14–19	417	31.7%, Lussi index	ETW was associated with age, high intake of sweet carbonated drinks, and xerostomia.
Kitasako *et al.* (2015)	Japan	15–29	191	30.9%	Dietary habits found to be frequently related to DE included acidic drinks for younger subjects and acidic fruits for older subjects.
		30–39	180	26.6%	
		40–49	191	23.6%	
		50–59	182	22.6%	
		60–69	186	25.8%	
		70–90	178	27.6%	
Søvik *et al.* (2014)	Norway	16–18	795	59.0%, VEDE	ETW was significantly more prevalent among males.
Isaksson *et al.* (2013)	Sweden	20	494	75.0%	A high prevalence of DE, but the level of severe erosion was low.
Mulic *et al.* (2013)	Norway	18	1,456	38.0%, VEDE	A high proportion of adolescents had ETW. Cuppings were a common finding.
Abu-Ghazaleh et al (2013)	Jordan	15–16	1,602	51.0%, modified TWI	Males had significantly more ETW than females.

[1]Estonia, Finland, Latvia, Spain, France, Italy, the UK

2.4.1 Gender differences in the prevalence of erosive tooth wear

With regard to gender differences, several studies have suggested that the prevalence of ETW is more frequent among males than among females (Al-Dlaigan *et al.* 2001, Bardolia *et al.* 2010, Fares *et al.* 2009, Mulic *et al.* 2012a, Søvik *et al.* 2014, Struzycka *et al.* 2017). Yet, there are still many studies that have not reported univocal gender differences in the prevalence of ETW (Bartlett *et al.* 2013, Provatenou *et al.* 2016, Vered *et al.* 2014). Differences between genders in terms of consumption of carbonated drinks and bite force have been suggested as an explanation (Hasselkvist *et al.* 2010); however, literature on the topic is scarce.

2.5 Susceptibility to erosive tooth wear

2.5.1 Saliva and pellicle in the susceptibility to erosive tooth wear

Susceptibility to ETW varies between individuals and is affected by a number of different variables, some of which still seem to remain unknown (Holbrook *et al.* 2009). Saliva is the most relevant factor in preventing the erosive wear of enamel (Hara & Zero 2014), and there is individual variance with respect to the level of prevention (Wetton *et al.* 2007). Especially salivary flow rate (Järvinen *et al.* 1991), buffering capacity (Humphrey & Williamson 2001, Van Nieuw Amerongen *et al.* 2004), the deposition of acquired salivary pellicle (Hannig & Hannig 2014) and the organic protein compound of saliva (Hara & Zero 2014, Hellwig *et al.* 2013) seem to affect the ETW process.

However, it is still not known which factors or components of saliva or pellicle are the most important ones concerning the level of protection, and study results on the topic are equivocal (Wang *et al.* 2011, Zwier *et al.* 2013). In an *in situ* study by Jager *et al.* (2011), it was found that several salivary parameters are associated with the susceptibility of hydroxyapatite to erosion, such as sodium, urea, total protein, albumin, pH and flow of unstimulated saliva, and sodium, potassium, urea, and phosphorus of stimulated saliva. Different proteins in acquired pellicle have also been shown to have an important role in protecting enamel from acid-induced demineralization (Hara & Zero 2014). Interestingly, it has been observed that the formation of acquired pellicle is reduced in patients with diet-induced erosion (Carpenter *et al.* 2014), thus extrinsic acids may also affect formation of pellicle.

2.5.2 Genes and erosive tooth wear

In many studies, males seem to dominate in the prevalence of (erosive) tooth wear with no clear explanation in the literature so far, which has increased interest on the topic. According to recent studies, enamel in males seems to be more prone to erosive wear when compared to females (Uhlen *et al.* 2016b), and individual susceptibility to enamel erosion is related to differences not only in the saliva but also in the enamel (Uhlen *et al.* 2016a). Furthermore, studies indicate that polymorphisms in enamel formation genes may be associated with the individual susceptibility to erosive wear, and it may also explain the gender difference (Søvik, 2015b, Uhlen *et al.* 2016b). However, this approach needs more extensive research, such as genome-wide association, candidate gene, and functional studies. Since Genome-Wide Association Studies (GWAS) are a hypothesis-free approach to scan the entire human genome to identify genetic loci that may be associated with diseases, new possibilities have opened up for gene research. Some GWAS have been performed on dental caries (Shaffer *et al.* 2013, Zeng *et al.* 2013), but to date, to our knowledge no GWAS have been established concerning ETW.

3 Aims of the study

The main aim of the study was to describe the prevalence, severity, and associated factors of ETW in adults in Finland, as well as to analyze new diagnostic possibilities.

The working hypothesis was that almost all of the examined adults have ETW of some degree, but the severity of ETW is low in general. Secondly, the hypothesis was that sociodemographic factors and dental caries are associated with ETW. Furthermore, the hypothesis was that ETW is more severe in patients with frequent symptoms of GERD, and that intrinsic factors are an important contributor in the ETW process. It was also hypothesized that there are loci in the genome which are associated with erosive wear; however, it is possible that they do not exceed the strict genome-wide association threshold. The BEWE index was hypothesized to be a reliable index both clinically and when applied to 3D models.

The specific objectives of this research were:

1. To assess the prevalence and severity of ETW in the NFBC1966 study population.
2. To analyze the association between ETW and sociodemographic factors and dental caries.
3. To determine the influence of intrinsic factors on ETW.
4. To perform a GWAS to analyze the possible contribution of genetics on the susceptibility to ETW.
5. To analyze the reproducibility of the BEWE index clinically and on 3D models.

4 Subjects and methods

4.1 Subjects

This work is composed of four individual studies that are based on the longitudinal research program Northern Finland Birth Cohort 1966 (NFBC1966), the main aim of which is to promote health and wellbeing of the population. Originally, the NFBC1966 comprised 12,058 children, whose expected time of delivery was in the year 1966. Ultimately, the birth cohort represented 96.3% of all such births (Northern Finland Birth Cohort Studies, 2016) (Figure 2).

Fig. 2. Picture published in the newspaper Kaleva of Prof. P. Rantakallio and Dr. Portin examining one of the cohort members in 1967. Reprinted with permission from Kaleva.

The entire NFBC1966 has been assessed regularly since birth by means of health questionnaires and clinical examinations. The original data have been supplemented by data in various hospital-specific and national registers. In connection with the 46-year follow-up survey (2012–2014), a subgroup comprising a total of 3,181 persons currently living in the city of Oulu, or within a distance of 100 km from Oulu, including rural areas, were asked to participate in a series of clinical investigations, including an oral and dental examination. Of the invitees, a total of 1,962 (61.7%) participated in the examinations, thus comprising the study population of this thesis. Of the study subjects, 903 were males and 1,041 females.

The first study (I) of the thesis assessed the prevalence and severity of ETW in the NFBC1966, and analyzed the association of ETW with dental caries. The second study (II) determined the influence of intrinsic factors on ETW on the basis of the 31- and 46-year surveys. For the third study, a GWAS on ETW was performed to analyze the possible role of genes in the susceptibility to erosive wear. The fourth study (IV) evaluated the reliability of the BEWE index in assessing ETW on 3D models.

All these studies are based on the clinical dental examinations of the cohort members and the supplementary data gathered during the NFBC1966 project, as well as on the discharge register of the Oulu University Hospital. Data used in the different studies are described in detail in Figure 3.

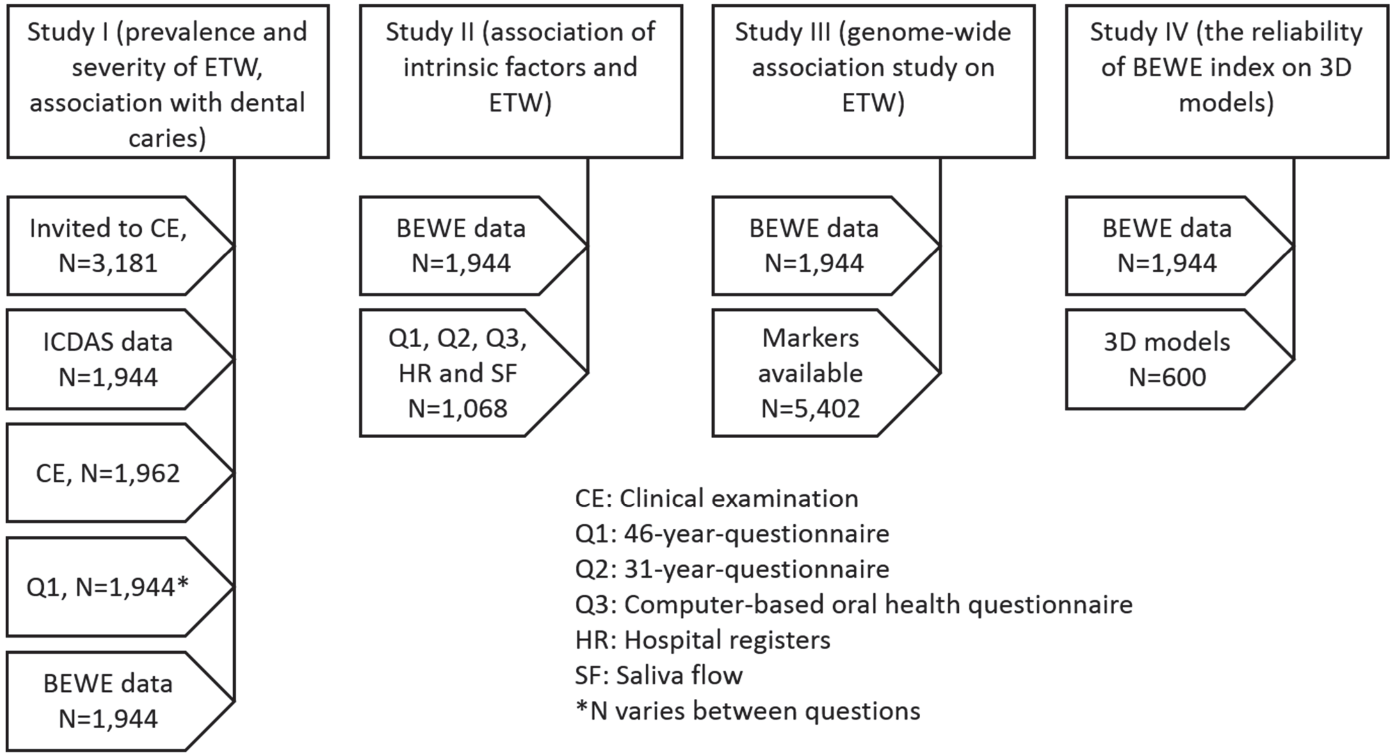

Fig. 3. Description of the population used in this study

4.2 Clinical dental examination protocol (I, II, III, IV)

Between April 2012 and June 2013, seven dentists performed the clinical oral and dental examinations on the study subjects by using a standardized clinical dental examination protocol. All the examinations were conducted in a dental chair of a modern dental unit equipped with optimal lighting at the Institute of Dentistry, Faculty of Medicine, University of Oulu, Finland. A probe, oral mirror, and WHO ball-pointed gingival probe were used to perform the examination. Before the assessment, the teeth were blow-dried by using a three-in-one syringe. No professional cleaning was done before the clinical examination, because the clinical examination also included periodontal measuring. A dental nurse registered the findings in an individual electronic patient file (by using software designed in the University of Oulu for the purposes of this study). In addition to the clinical examination, the salivary flow rate was assessed and a dental 3D modelling was performed. All clinically examined participants were asked to fill out a computer-based oral health questionnaire, which was a modified version of the questionnaire developed in the University of Oulu and used in several studies (Anttonen *et al.* 2011, Kämppi *et al.* 2014).

ETW—diagnostic criteria

ETW was assessed by using the BEWE index (Bartlett *et al.* 2008)] All tooth surfaces were assessed for erosive wear (range 0 to 3), and the highest score for each sextant was recorded. The scoring criteria is presented in Table 2. Then, the sum score of the sextant scores was calculated (range 0 to 18). The examiners were advised not to include wedge-shaped defects or incisal edges in the results if the role of abrasion or attrition was obvious. Furthermore, erosive wear could not be recorded when extensive restorations covering the entire tooth surface or prosthetic works were present.

Table 2. Scoring ETW by using the BEWE index.

Score	Clinical appearance
0	No erosive wear
1	Initial signs of erosive wear, such as loss of surface
2[1]	Distinct defect, hard tissue loss <50% of the surface
3[1]	Hard tissue loss >50% of the surface area

[1]Dentine is often involved in scores 2 and 3

Dental caries—diagnostic criteria

Caries lesions were detected by using the International Caries Detection and Assessment System ICDAS (Ismail *et al.* 2007). ICDAS protocol classifies caries lesions according to their appearance and histologic depth (codes 1-6). Scores 1 and 2 stand for early stage decays with visual changes in enamel, but without enamel breakdown. Scores 3 and 4 describe established decays, both with enamel breakdown. In score 4, an underlying dentine shadow is seen. Scores 5 and 6 describe severe decay. In this study, ICDAS score 4 was set as the cut-off value for restorative treatment decision, i.e. lesions of that depth or deeper represented manifested caries lesions. In borderline cases, especially to distinguish between ICDAS scores 3 and 4, the clinical examiners were advised to choose the more severe option. No radiographs for specific caries detection were obtained.

A dental nurse registered all clinical findings by using a laptop computer equipped with software designed in the University of Oulu for the purposes of this study.

Validation of the clinical examination

The dentists conducting the examinations were trained and calibrated by specialists and senior researchers in the field of cariology and periodontology. Lectures on the use of the different indices were given during the training to make sure that all the examiners would have similar knowledge about the different criteria. The criteria were presented by means of a PowerPoint presentation by using a PC and a data projector. Following the lectures, and in order to practice their diagnostic skills in cariology, the examiners determined the classification of 26 extracted teeth with a variety of caries lesions by using the ICDAS criteria (Ismail *et al.* 2007). A revision of the criteria and calibration of the examiners were repeated every three months. The criteria for the BEWE classification and the ICDAS were available online, as well as in written and graphical form in the examination room. An experienced clinician and senior researcher familiar with the study protocol and criteria acted as the gold standard and monitored that the examiners followed the study protocol precisely throughout the entire field study.

To assess the intra-examiner agreement, the examiners re-examined (including BEWE and dental caries) one quadrant of on average 10 patients approximately one month after the first examination. To assess the inter-examiner agreement, the gold standard re-examined one quadrant of on average 12 patients from each examiner. The validation was carried out according to the protocol followed in the national study (Health 2000, 2017).

4.2.1 Saliva samples

In addition to the clinical examinations, saliva samples were collected and unstimulated and stimulated salivary flow rates were determined. The flow rates were assessed in the morning before the clinical examination. The participants were asked to sit comfortably and were left undisturbed. For collecting rest saliva, the patients were asked to let saliva run into a plastic container for 15 minutes without any physical effort, and after that the flow rate was determined (ml/min). To determine the stimulated salivary flow rate (ml/min), the participants were given a piece of paraffin wax to chew and were asked to actively spit saliva into a plastic container for five minutes. All the participants were asked to fast for 12 hours prior to the saliva sampling. An oral hygienist assisted in the saliva sampling.

4.3 Questionnaires (I, II)

4.3.1 Postal questionnaires (I, II)

The cohort members have received several postal questionnaires during the NFBC1966 cohort studies. In 1997–1998, the 31-year questionnaire was sent to the cohort members whose address was known (n=11,541). Of those, 8,690 (75%) responded to the questionnaire and thus agreed to participate in the study. The questionnaire included questions about the general and oral health and health behaviors. Data based on this questionnaire were used in Study II.

In 2012–2013, a similar questionnaire, with some additional questions, was sent to the cohort members by mail as part of the 46-year follow-up survey (n=10,321); a total of 6,825 (66%) subjects responded. The sociodemographic data of the cohort members used in Study I were collected on the basis of this questionnaire. The specific variables obtained were gender (n=1,944), marital status (n=1,879), and education level (n=1,869).

The presence of gastro-esophageal reflux disease (GERD) and chronic alcoholism, as well as drinking habits concerning alcohol-containing drinks were assessed similarly in both postal questionnaires, and the data obtained were used in Study II. The presence of GERD was assessed by the question "*Have you ever had any of the following symptoms, sicknesses or injuries verified or treated by a doctor? Other sickness or injury, which?"* The presence of chronic alcoholism was assessed by the question *"Have you ever had any of the following symptoms, sicknesses or injuries verified or treated by a doctor: Alcohol abuse?" (Yes/no)* Drinking habits and alcohol consumption was questioned by the questions (all drinks separately) "*How often do you usually drink beer, cider or long drinks / light wine / wine / liquor?" (Never, less than once a year, a couple of times / three to four times a year, once in two months, once a month, two times a month, once a week, couple of times a week, daily),* "*How much beer, cider or long drinks / wine / liquor do you usually drink (1 bottle = 1/3 liter)?" (Less than one bottle, one bottle, two / three / four to five / 6 to 9 / 10 to 14 / over 15 bottles, not at all).*

The 46-year postal questionnaire also included several GERD-related questions, which were considered in Study II. The presence and severity of GERD symptoms were assessed by the following questions: "*How often have you had troublesome acid regurgitation and/or heartburn during the last year?" (Never/rarely than once a month/at least once a month/weekly/daily), "Do you wake up at night because of acid regurgitation and/or heartburn?" (Yes/no),* and *"At what age did you first have these symptoms?"* The number of pregnancies was investigated by the question *"If you have been pregnant, how many deliveries have you had?"*

4.3.2 Computer-based oral health questionnaire (II)

All the cohort members who were clinically examined were asked to fill out a computer-based oral health questionnaire (Anttonen *et al.* 2011, Kämppi *et al.* 2014) after their clinical dental examination. A dental nurse assisted with the questionnaire where needed. The questionnaire consisted of several oral and dental health-related questions, including questions about oral health behaviors, and the following questions were utilized in the analysis: "*Have you noticed or been told* *that you grind your teeth or clench while sleeping / during daytime?" (Yes/no), "What do you drink during meals?", and "What do you drink to quench thirst?" (Milk / juice / juice from concentrate / tap water / soft drink / light soft drink / bottled water / something else).* An enumerator assisted with responding the oral health questionnaire. No problems occurred, and no one refused answering.

4.4 Genetic data (III)

The genetic data used in the study were obtained from blood samples taken in connection with the 31-year examination. Blood samples were drawn after overnight fasting in the morning (between 08:00 and 11:00 a.m.). The genome-wide association genotyping is described in detail in Sabatti *et al.* (2009).

4.5 3D modelling (IV)

The three-dimensional models were obtained by an oral hygienist and clinical examiners trained by the manufacturer to use the iTero 3D scanner (Cadent, San Jose, CA, USA) on day of the clinical examination. All the models were scanned and stored unrevised in the server of the cohort.

3D model analysis protocol

In 2015, one-third (n=600) of the clinically examined cohort population with 3D models was randomly selected (the first 600 3D models from the list where they appear according to their project ID) to comprise the study population for Study IV. The 3D models were assessed on a computer screen by using the BEWE index and the same scoring criteria as in the clinical examination, as well as the 3Shape Ortho Analyzer™ software. The analyses were performed over one week at the same place and using the same lightning. The analyses were first timed to maximize the sample size in a limited period of time. The criteria for the BEWE classification were available to the examiners in written and graphical form in the analyzing room.

Validation of the 3D model analysis protocol

The gold standard examiner and the instructor of the clinical examination trained three examiners (two fourth-year dental students and the author) on the protocol by using the same instructions as in the clinical examination. The examiners were calibrated based on the coordinated analysis of the 3D models until agreement was reached on the severity of erosive changes. Two fourth-year dental students analyzed all the 600 3D models together, and a dentist (author) acted as the gold standard for the re-examinations and was available in the examination room throughout the study. To assess the intra-examiner agreement, every twentieth 3D model (n=19) was chosen to be re-examined after analyzing all the 600 3D models. To assess the inter-examiner agreement, 11 of these randomly chosen 3D models were re-examined also by the gold standard. The re-examinations were performed at the same time and in the same space. During the assessment of the inter-examiner agreement between the students and the gold standard, the students were allowed to discuss with each other while deciding the score.

4.6 Statistics

To analyze the prevalence and severity of ETW, the BEWE sum scores were classified in three categories and interpreted as presented in Table 3. The data were also analyzed based on the highest BEWE score recorded per individual (range 0 to 3) (Study I). Wisdom teeth were excluded from all the analyses. P-values <0.05 were considered statistically significant. The data on Studies I, II and IV were analyzed by using SPSS (version 22.0, SPSS, Inc., Chicago, IL, USA) and on study III by using R (version 3.2.5 patched, a language and environment for statistical computing, R Foundation for Statistical Computing, Vienna, Austria).

Table 3. Severity of ETW according to BEWE sum scores.

BEWE sum score	ETW severity
0–2	No or only mild ETW, no treatment need
3–8	Moderate ETW, at least preventative strategies needed
9–18	Severe ETW, treatment needed

4.6.1 Study I

To analyze the role of sociodemographic factors in ETW, the variables were categorized as follows: marital status was dichotomized into persons married, cohabiting or living in a registered relationship and the rest (unmarried, divorced and widowed). Education level was categorized into basic education (nine years of comprehensive school) and upper comprehensive school education (total 12 years plus matriculation examination).

The ICDAS score 4 was set as the cut-off point for decayed surface (D). Restorative treatment need (RTN) was dichotomized for surface, tooth and sextant level, and both dichotomized and trichotomized for dentition level (Table 4). This was done to be able to analyze the association between RTN and ETW on sextant and dentition level. To analyze the association between past and present caries experience and erosive wear, DMFT indices were calculated and categorized. In calculating the DMFT values, the ICDAS score (D, ICDAS ≥4) and the presence of fillings (F) or extractions (M) were considered. The M value consisted of all missing teeth regardless of the cause of extraction, since no data about previous dental treatments were available. Based on the mean DMFT value among the population, DMFT ≤14 was considered as low and DMFT >14 as high caries experience. The association between past and present caries experience and erosive wear was analyzed by comparing the BEWE sum scores and categorized DMFT values and the present restorative treatment need due to caries.

Table 4. Restorative treatment need on different levels according to D and ICDAS scores.

Levels	Restorative treatment need (RTN)
Surface	
D=0	No RTN (ICDAS score 0–3)
D=1	RTN (ICDAS score 4–6)
Tooth	
D=0	No RTN
D≥1	At least one surface in a tooth with RTN
Sextant	
D=0	No RTN
D=1	At least one tooth in a sextant with RTN
Dentition dichotomized	
D=0	No RTN
D≥1	At least one tooth in a dentition with RTN
Dentition trichotomized	
D=0	No RTN
D=1–3	1 to 3 teeth with RTN, moderate RTN
D≥4	More than 4 teeth with RTN, high RTN

Descriptive statistics, including frequencies, distributions, means, and standard deviations, were used. Cross-tabulation and chi-square tests were used for analyzing the distribution of different variables according to gender. The Mann-Whitney U-test was used to compare the number of decayed teeth and BEWE sum scores between genders. An independent sample t-test was used to compare the mean DMFT values between genders. Logistic regression models were used to analyze the association between severe ETW and different variables, and OR values and 95% confidence intervals (CI) were calculated. All the variables (gender, education, marital status, restorative treatment need, DMFT) were initially included in the bivariate analyses (chi-square test and unadjusted logistic regression model), and the variables that were statistically significantly associated with ETW were included in the adjusted logistic regression model.

4.6.2 Study II

To study the association between ETW and intrinsic factors, the variables in the questionnaires were classified as presented in Table 5. Questions about GERD and tooth-grinding were only included in the 46-year follow-up questionnaire. On the basis of the Montreal definition of GERD (Vakil *et al.* 2006), self-reported troublesome acid regurgitation and/or heartburn were considered as symptoms of GERD.

To evaluate the outcome of heavy use of alcohol, the consumption of alcohol (g/day) was determined on the basis of the 31- and 46-year postal questionnaires. For heavy use of alcohol, 30 g/day was set as the cut-off point for men and 20 g/day for women (Foster & Marriott 2006). To evaluate the long-term effect of heavy alcohol consumption on ETW, the study population was divided into four categories: those who were heavy users of alcohol at the age of 31 and 46, those who were heavy users of alcohol only at the age of 31 or 46, and those who were not heavy users of alcohol either at the age of 31 or 46. Bruxism was considered "possible bruxism", in accordance with a recent consensus report (Lobbezoo *et al.* 2013), if the answer for the bruxism question at the age of 46 years was "yes". With respect to non-alcoholic drinking habits, consuming "something else" during meals or when thirsty was considered a missing value.

A history of eating disorders in the study population was assessed based on the Oulu University Hospital Discharge Register. To analyze the saliva secretion, the cut-off value for hyposalivation was set to < 0.1 ml/min and for reduced salivary flow to 0.1–0.25 ml/min for unstimulated saliva. For stimulated saliva, the cut-off values were <0.7 ml/min and 0.7–1.0 ml/min respectively (2014).

Table 5. Classification of self-reported variables obtained from the questionnaires.

Self-reported data	Interpretation
Gastro-esophageal reflux disease[a]	
GERD symptoms daily	Severe GERD
GERD symptoms weekly	Moderate GERD
GERD symptoms less frequently	Mild GERD
Symptoms of GERD at the age of 20 or earlier	Early-onset GERD
Symptoms of GERD at the age of 21–30	Mid-onset GERD
Symptoms of GERD at the age of 31 or later	Late-onset GERD
No reported GERD symptoms	No GERD
Alcohol abuse verified by a doctor	
Alcohol abuse in both questionnaires	Long-term alcohol abuse
Alcohol abuse only in the 46-year questionnaire	Short-term alcohol abuse
No alcohol abuse	No alcohol abuse
Heavy use of alcohol	
Heavy user of alcohol in both questionnaires	Long-term heavy use of alcohol
Heavy user of alcohol in either questionnaire	Heavy use of alcohol
Not a heavy user of alcohol in either questionnaire	Not a heavy user of alcohol
Pregnancies	
No deliveries	No deliveries
1–2 deliveries	1–2 deliveries
3–4 deliveries	3–4 deliveries
At least 5 deliveries	At least 5 deliveries
Tooth grinding daytime / at sleep[a]	
Yes	Possible bruxism
No	No bruxism
Non-alcoholic drinking habits	
Consuming milk, tap water or bottled water during meals	No erosive drinking habits during meals
Consuming milk, tap water or bottled water when thirsty	No erosive drinking habits when thirsty
Consuming juices, soft drinks or light soft drinks during meals	Erosive drinking habits during meals
Consuming juices, soft drinks or light soft drinks when thirsty	Erosive drinking habits when thirsty

[a]Asked only in the 46-year questionnaire

Cross-tabulation was used to analyze the association between the different variables (gender, symptoms of GERD, onset of GERD symptoms, alcohol abuse, heavy use of alcohol, pregnancies, sleep bruxism and hyposalivation, non-alcoholic drinking habits), and chi-square tests were used to investigate the statistical significance of the differences between the groups. A logistic regression analysis was used to calculate the unadjusted and adjusted odds ratios (OR), 95% confidence intervals (CI), and Coefficient of Determination. The variables that were statistically significant risk factors for ETW were included in the logistic regression model. P-values <0.05 were considered statistically significant.

4.6.3 Study III

To reduce the probability of over-registration, and considering the age of this population, severe erosive wear was selected as the cut-off point for the GWA analysis, and the members affected by severe erosive wear were considered erosion cases. All the cohort members with BEWE ≤8 were considered controls.

The following quality control steps were applied: single nucleotide polymorphisms (SNPs) with call rate <95% or minor allele frequency <0.05 were excluded from the study, along with individuals with a genotyping success rate <95%. Imputation success was set at 0.4. In addition, statistically estimated HapMap2- and 1000 Genomes-imputed markers were available for 5,402 individuals. The GWAS was performed on the full sample and stratified by gender (male n=903 and female n=1,041) and by using the extreme opposite approach (comparing those with BEWE score 0–2 to those with BEWE score 9–18). The genetic analyses were performed with SNPTEST 2.5, using the frequentist association models. For every SNP meeting statistical (p-value≤5x10E-8) or suggestive significance (p-value≤5x10E-5), we explored any nearby genes and whether they had known biological functions relevant to erosive wear by using the NCBI Variation Viewer. The associated markers were not expected to be causal, but were assumed to be physically proximal and in linkage disequilibrium with an unobserved causal variant.

4.6.4 Study IV

The kappa coefficient and percentage agreement were calculated to analyze and demonstrate the inter- and intra-examiner, as well as inter-method agreements. Different cut-off points were used to analyze their impact on the outcomes. Kappa values were interpreted as suggested by Landis and Koch (Landis & Koch 1977). The inter-method difference between the mean BEWE outcomes per sextant was analyzed by using the Wilcoxon signed rank test, and the mean BEWE sum score by using a paired sample t-test. The differences of the BEWE sum score distributions between the methods were tested by using the chi-square test and Fisher’s exact test.

4.7 Ethical considerations

The data were disclosed to investigators without personal identification numbers, meaning that individual investigators were not aware of any subject's identity. The Ethical Committee of the Northern Ostrobothnia Hospital District approved the full cohort study (74/2011), which was performed according to the Helsinki Declaration of 2008, as well as the present subgroup study (227/2012).

5 Results

5.1 Prevalence and severity of erosive tooth wear (I)

The study sample comprised 1,962 44–46-year-old adults, and was slightly dominated by the female gender (46.5% men and 53.5% women). BEWE index data were available for a total of 1,944 subjects. Of those, 75% had ETW of some degree, and about half had erosive wear needing at least preventive measures, or even operative care (Figure 4). The highest BEWE score was 1 for 54.7%, 2 for 18.2%, and 3 for 2.1% of the participants. Males dominated with respect to both the prevalence and the severity of ETW (Figure 4). Anterior sextants were most affected (15%) by distinct ETW (BEWE score 2 or 3). In posterior sextants, the prevalence of distinct tissue loss was 6%.

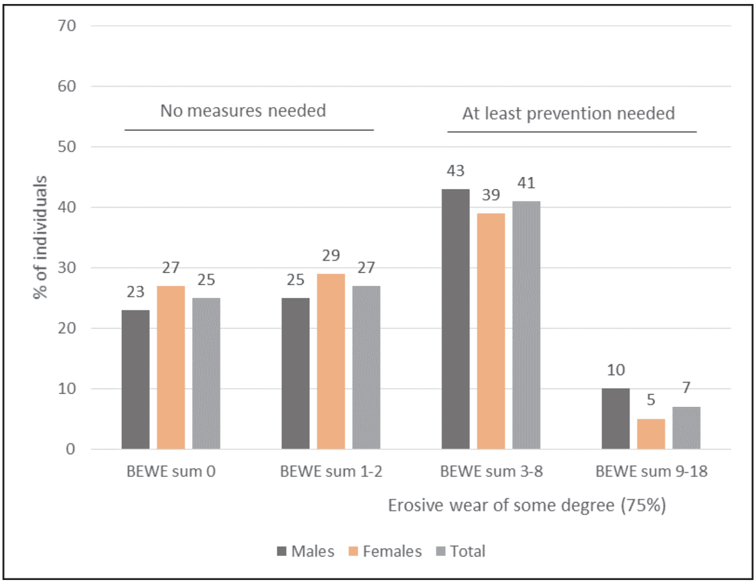

Fig. 4. The distribution of ETW according to gender.

5.2 Associating factors of erosive tooth wear (I, II)

5.2.1 Sociodemographic factors (I)

In an adjusted regression model with ETW as the dependent variable, the OR for the male gender was 1.8 (95% CI 1.26 to 2.68) compared to women. Severe ETW was the most prevalent among males with only basic education (53.6%) and the least prevalent among women who had completed the matriculation examination (40.2%) ($p<0.001$). In the adjusted regression model, neither education nor marital status was statistically significantly associated with ETW.

5.2.2 Caries experience and restorative treatment need due to caries (I)

The mean DMFT score in the entire study population was 14.9 (SD 5.15). The proportion of adults in need for restorative treatment was 39.8%, and the mean value for D was 1.0 (SD 1.84). Males had more restorative treatment need due to caries (mean D 1.2, SD 2.0) compared to females (mean D 0.8, SD 1.7), and the difference between the genders was statistically significant ($p<0.001$). There was no statistically significant difference between the genders concerning DMFT.

The association between restorative treatment need due to caries (D) and severe ETW was statistically significant, whereas the association between severe ETW and DMFT was not (Table 6). The association between ETW and DMFT was, however, statistically significant when comparing those with BEWE sum score 0–2 to those with BEWE sum score ≥3. In an adjusted regression model computed on the basis of dichotomized restorative treatment need (no/yes), the OR for having severe ETW was 2.1 for those with restorative treatment need. As seen in Table 6, the more restorative treatment need, the more odds for severe ETW.

Table 6. The association between severe erosive wear and restorative treatment need (RTN) and DMFT. Unadjusted and adjusted logistic regression models.

Variable	Severe erosive wear (BEWE≥8)				
	No n (%)	Yes n (%)	p-value	Unadjusted OR (95% CI)	Adjusted OR[1] (95% CI)
No RTN[2]	1,113 (95.1)	57 (4.9)	<0.01	1	Not incl.
RTN	695 (89.8)	79 (10.2)		2.2 (1.56–3.16)	
No RTN[2]	1,113 (95.1)	57 (4.9)	<0.01	1	1
Moderate[3] RTN	551 (90.3)	59 (9.7)		2.1 (1.43–3.05)	2.0 (1.34–2.94)
High[4] RTN	144 (87.8)	20 (12.2)		2.7 (1.58–4.64)	2.7 (1.56–4.75)
DMFT≤14	885 (93.6)	61 (6.4)	0.36	1	Not incl.
DMFT>14	923 (92.5)	75 (7.5)		1.2 (0.83–1.67)	

[1]Logistic regression model computed using trichotomized restorative treatment need

[2]D=0

[3]D=1–3

[4]D≥4

5.2.3 Intrinsic factors (II)

Intrinsic factors explained 5.9% of the variance in the prevalence and severity of ETW in this population. Gender (p<0.001), daily GERD symptoms (p=0.047), early onset of GERD (p=0.048), diagnosed alcoholism either at the age of 31 or 45 years (p<0.001), long-term heavy use of alcohol (p=0.006), sleep bruxism (p=0.003), and hyposalivation (unstimulated saliva) (p<0.001) were all statistically significantly associated with severe ETW. The association between consuming juices or soft drinks to quench thirst and moderate ETW was statistically significant (p=0.045). These variables were included in a logistic regression model, and the outcome was that the strongest risk factors for ETW were hyposalivation and daily symptoms of GERD (Table 7).

When considering the questions concerning waking up at night or taking medicines daily because of GERD, daytime tooth grinding, or stimulated salivary flow, no statistically significant association with ETW was found. The number of pregnancies or drinking juices or soft drinks during meals were neither associated with severe ETW (Table 7). There were no subjects with diagnosed eating disorders among the study population.

Table 7. Logistic regression model; unadjusted and adjusted ORs and 95% CIs for severe ETW.

Variable	Unadjusted OR (95% CI)	Adjusted OR (95% CI)
Gender		
Male	2.1 (1.45–2.99)	2.0 (1.16–3.53)
Female	1	1
GERD Symptoms		
Daily	2.5 (1.13–5.71)	3.8 (1.20–12.01)
Weekly	1.4 (0.77–2.48)	not incl.
Rarely	1.1 (0.75–1.68)	not incl.
Never	1	1
Onset of GERD symptoms		
Age ≤20	1.9 (1.13–3.46)	1.1 (0.45–2.61)
Age 21–30	1.2 (0.71–1.90)	not incl.
Age 31–46	1.0 (0.60–1.58)	not incl.
No GERD	1	1
Diagnosed alcoholism		
In 31-year survey	9.0 (2.61–31.45)	not incl.
In 46-year survey	4.8 (1.87–12.09)	not incl.
In either survey		2.5 (0.65–9.70)
No diagnosed alcoholism	1	1
Heavy alcohol consumer		
In both surveys	3.0 (1.37–6.59)	2.0 (0.63–6.24)
Only in 46-year survey	1.6 (0.86–3.06)	not incl.
Only in 31-year survey	1.1 (0.40–3.21)	not incl.
Never	1	1
Possible sleep bruxism		
Yes	1.7 (1.20–2.43)	1.3 (0.74–2.17)
No	1	1
Unstimulated saliva secretion		
Hyposalivation	5.5 (2.35–12.82)	3.8 (1.20–11.84)
Lowered secretion	0.9 (0.51–1.45)	not incl.
Normal secretion	1	1
Juices or soft drinks when thirsty		
Yes	1.1 (0.69–1.87)	not incl.
No	1	not incl.

We also determined the smallest amount of alcohol consumed per day that was positively associated with the prevalence and severity of ETW at the age of 46 years (Table 8). The difference in the prevalence and severity of ETW was statistically significant between those who, at the age of 46 years, reported consuming at least

7 g of alcohol (half of the standard drink in Finland) per day and those who reported consuming less than 7 g of alcohol per day. The difference in the prevalence and severity of ETW at 46 years of age was also statistically significantly higher for those who at the age of 31 years consumed alcohol at least 3 g per day.

Table 8. The smallest amount of alcohol per day and the risk of ETW.

Amount of alcohol/day	BEWE sum ≤8 N (%)	BEWE sum >8	p-value
46-year questionnaire			
<7 g of alcohol/day	975 (94%)	62 (6%)	0.01
≥7 g of alcohol/day	701 (91%)	68 (9%)	
31-year questionnaire			
<3 g of alcohol/day	584 (95%)	29 (5%)	0.02
≥3 g of alcohol/day	881 (92%)	75 (8%)	

5.3 Genome-wide association study

The GWAS was performed by using 136 cases with severe erosive wear (BEWE sum score >8) and 1,808 controls (BEWE sum score ≤8). Since severe erosive wear cases among males were twice to that of females, we also conducted a GWAS stratified by gender. The performed GWAS scans implicated several loci and SNPs putatively associated with severe erosive wear, as seen in the Manhattan plots on the whole study population and separately on males and females (Figure 1 in Study III). The genomic inflation factor, λ, was 1.006, 1.014, and 1.024 for the three analyses, respectively, indicating negligible p-value inflation.

5.3.1 Genome-wide association study on the whole sample

The SNP exhibiting the strongest signal in the whole sample was rs11681214 located in the chromosomal region 2p24 (p-value=3.99E-08), with two suggestively associated SNPs locating nearby. Additionally, there were 117 suggestively significant SNPs. The SNPs with the strongest signal and the biggest clusters of suggestively significant SNPs are listed in Table 9. Figure 5 illustrates the regional plot of rs11681214.

Table 9. Loci with the strongest signals in the GWAS on ETW, whole sample.

Region	SNP	p-value	Related gene symbol
2p25	rs11681214[a]	3.99E-08	*PXDN, MYT1L*
4q34	Chr4:174962625, rs72698178	2.13E-07, 8.83E-06	*FBXO8*
5q31	chr5:141405828, rs66756037	8.16E-06, 7.36E-06	*FGF1*
8p11.2	rs11993596, rs112007639	5.62E-06, 6.23E-06	*FGFR1*

[a]Statistically significant

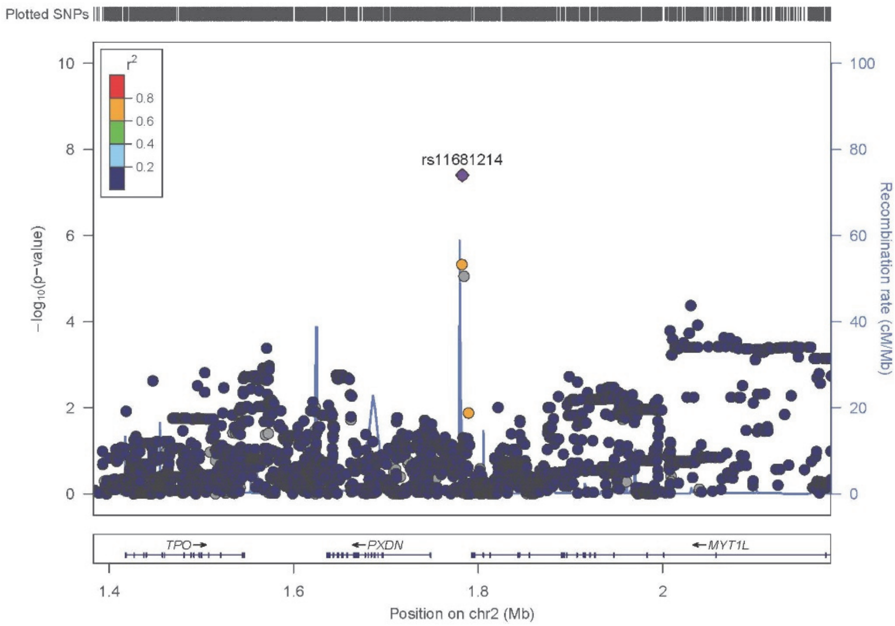

Fig. 5. Regional plot of rs11681214 in the chromosome 2. The SNP rs11681214 is located between the genes *PXDN* and *MYT1L.*

5.3.2 Genome-wide association study using extreme opposite approach

Using this approach, two statistically significant SNPs met the statistically significant threshold: rs118188573 on chromosome 7p15.2 (p-value 6.38E—08) near the gene *SKAP2* and Homeobox A-gene family, and rs142118483 on chromosome 10 (p-value 7.62E-08) near the gene *ZWINT*. Close to these two SNPs, two and 17 SNPs, respectively, reached the suggestive significance. Both SNPs were also suggestively significant in the whole-sample GWAS.

On chromosome 4, two interesting clusters of suggestive SNPs existed: one in the chromosomal region 4p16 near the gene *MSX1* gene and another in the gene *GLRA3* in the chromosomal region 4q34 near the genes *FBXO8* and *SCRG1*. The latter was suggestive also in the whole-sample GWAS.

5.3.3 Genome-wide association study on males

In males (n=903), 14 SNPs exceeded the genome-wide significance level, and 214 SNPs attained the level of suggestive significance. Many of the SNPs associated with ETW were near or in the genes that have a known or plausible role in tooth development or that are otherwise related to the oral environment, such as *FGF1* and *FGFR1*. The loci with the strongest signals and related genes are listed in Table 10.

Table 10. Loci with the strongest signals in the male GWAS.

Region	SNP	p-value	Related gene symbol
3p21.1	rs189767158, chr3:53325260	3.81E-08, 3.30E-08	DCP1A
5q31.1	rs12108935, chr5:141405828, rs66756037	6.19E-08…2.4E-08	FGF1
8p11.2	rs11993596, rs112007639 rs12546327, rs2461333	8.38E-09…1.52E-08	FGFR1
20q13.3	rs2426986, rs16984837 rs6101273	9.66E-08…3.35E-08	CDH4

5.3.4 Genome-wide association study on females

In the female GWAS (n=1,041), altogether 9 SNPs met the criteria of statistical significance, and 317 SNPs were found suggestively significant. Also among females, many of the SNPs associated with ETW were near or in the genes that are putatively related to tooth development or the oral environment, such as *F2R* and *SCD5*. The loci with the strongest signals and related genes are listed in Table 11.

Table 11. Loci with the strongest signals in the female GWAS.

Region	SNP	p-value	Related gene symbol
4q21.2	chr4:82684912	7.13E-10	*SCD5*
5q13.3	chr5:76795786	7.97E-10	*F2R*
13q34	rs61969071, rs9559864 rs61969075	9.47E-08…8.43E-08	*ING1*

5.4 3D models for erosive tooth wear assessment (IV)

ETW was recorded as more severe based on the 3D models than the clinical examination (Figure 6), and the inter-method differences were statistically significant in all sum score categories.

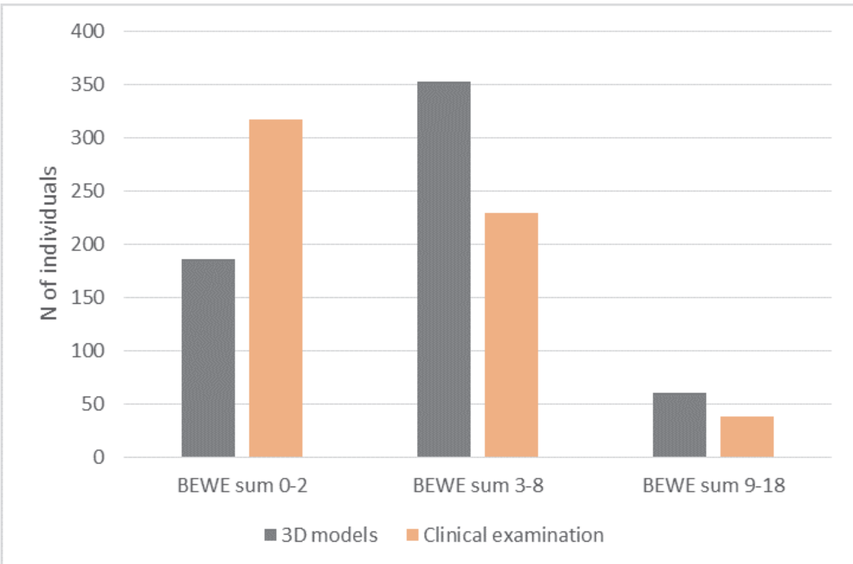

Fig. 6. Distribution of BEWE sum scores according to 3D models and the clinical examination.

5.5 Reproducibility of the BEWE index (I, IV)

5.5.1 Clinical examination (I, IV)

When considering the agreement on distinct erosive wear (BEWE score 2 or 3), both the intra- and the inter-examiner agreement were excellent. Considering ETW of any degree, the intra-examiner agreement was moderate and the inter-examiner agreement was fair (Table 12). Intra- and inter-examiner agreement could not be calculated for BEWE sum scores since only quarter of jaw per patient was re-examined, and thus ETW was reassessed only for one whole sextant/patient.

Table 12. Reproducibility of the two methods and inter-method agreement based on the mean kappa values.

Method	Mean kappa values (sextant)	
	Distinct ETW	ETW of any degree
Clinical examination		
Intra-examiner	0.98	0.46
Inter-examiner	0.81	0.30
3D models		
Intra-examiner	0.89	0.71
Inter-examiner	0.87	0.72
Inter-method	0.31	-

5.5.2 3D models (IV)

The intra- and inter-examiner agreements between sextants were excellent considering distinct ETW, and fair-to-good when considering erosive wear of any degree. With respect to BEWE sum scores, the intra-examiner agreement was 0.64, the percentage agreement reaching 95%. The inter-examiner agreement was 1.

5.5.3 Inter-method (IV)

On a sextant level, the mean inter-method agreement was fair (Table 12). The lowest level of agreement concerned the lower anterior sextant. With respect to BEWE sum scores, the inter-method agreement was 0.41 (cut-off BEWE sum >8), the percentage agreement being 91%.

5.6 Main findings (I–IV)

- ETW was common among the Finnish adult population: 75% had erosive wear, and almost 50% needed at least preventive measures to arrest the progress of the condition.
- Male gender and restorative treatment need due to dental caries significantly increased the odds for ETW, whereas sociodemographic factors were not associated with ETW according to the adjusted model.
- The studied intrinsic factors were relatively uncommon among the study population.
  - Of the intrinsic factors, daily symptoms of GERD and hyposalivation were the most significantly associated with severe ETW.
- There were statistically significant signals in the GWAS on ETW, implying a genetic contribution to the ETW process.
  - There were differences in GWAS signals between males and females concerning ETW.
- The BEWE index was reliable for recording ETW clinically and on 3D models.
  - 3D models were especially sensitive in detecting initial erosive wear.
  - The biggest inter-method differences between clinical and 3D detection were in the upper posterior sextants.

6 Discussion

The NFBC1966, which includes data on individuals even before their birth, provides a unique opportunity to conduct epidemiological studies by investigating the impact of behavioral, biological, and genetic risk indicators and factors on diseases. Originally, the cohort comprised 96.3% (12,068) of all births during 1966 in Northern Finland, thus representing this age group in Northern Finland in an excellent manner. The present study comprised a total of 1,962 individuals from the NFBC1966, thus representing 19.0% of the cohort members who were alive in 2012–2013 and whose address was known, and 62.3% of those were invited for the clinical examination. Considering this, participation rate was relatively high, and the study population, which was not limited to any subgroup, is one of the main strengths of this study.

The study population represents the NFBC1966 well. The results here are in line with the national study on caries experience (Health 2000, 2017), which has a similar study population of similar age. Due to the multifactorial etiology of ETW, it can be speculated that the prevalence of ETW among those who did not participate is not expected to differ from those who did. It seems that our findings represent the ETW and caries status of an average Northern Finnish population well.

6.1 Main findings

Among the study group, the overall prevalence of ETW was high, which was expected based on the average age of 46 years of the cohort members. Additionally, almost half of the now middle-aged cohort members were found to be in need of at least preventive measures to arrest further progression of the condition. Of the individuals with erosive lesions, 15% had severe ETW. The most recent studies using the BEWE index in other countries are in line with the prevalence figures presented here (Bartlett *et al.* 2013, Vered *et al.* 2014). However, ETW studies on adults are scarce, and due to the variations in scoring systems and target populations, comparison between the studies is difficult if not impossible. Yet, many studies using different scoring systems than BEWE (Fares *et al.* 2009, Isaksson 2013) have reported similar prevalence values as the present study.

ETW was associated with restorative treatment need due to caries, whereas there was not an association between ETW and sociodemographic factors other than gender. As reported before, our results also showed that the male gender seems to predispose individuals to ETW (Bardolia *et al.* 2010, Fares *et al.* 2009). Intrinsic factors related to erosive wear were rare and explained only 6% of the difference in the prevalence and severity of erosive wear in the study population. This might suggest that intrinsic factors are of minor importance epidemiologically with respect to the ETW process, at least when assessed by using questionnaires. However, on an individual basis, daily problems with GERD and reduced saliva secretion or hyposalivation seemed to be clear risk indicators for severe ETW. Considering individual variation, GWAS on ETW supported the assumption that susceptibility to ETW may be influenced by genetic variance. The genome-wide study revealed several loci in genome that were statistically or putatively associated with severe ETW.

Based on the observed kappa values, the BEWE index was assessed as acceptable for recording ETW clinically. When considering the agreement on distinct ETW on the basis of the BEWE criteria, both the intra- and the inter-examiner agreement were excellent. However, with respect to determining initial wear, there were difficulties, especially in differentiating between intact enamel and initial loss of surface texture. The age of the cohort and the fact that the examiners were not experienced in scoring initial erosive wear and that the clinical examination included numerous factors to be assessed may all explain this finding. On 3D models, assessing ETW by using the BEWE index appeared to be reliable and the method was especially sensitive in finding initial ETW.

6.2 Study population

The age of the cohort complicates the assessment of ETW, which must be kept in mind when considering the outcome of this study. Erosive wear usually co-exists simultaneously with other types of tooth wear like attrition and abrasion, which are often present especially in ageing dentitions. In general, the prevalence and size of erosive lesions tend to increase with age (Jaeggi & Lussi 2014). Due to limited experience of the examiners combined with the challenges due to the age of the cohort members, it seems more likely that the prevalence of ETW is rather underestimated than overestimated in this study. In a study of young European adults by Bartlett *et al.* (2013), in Finland, the maximum BEWE score per sextant was at least 2 for almost 20% of the examined. This is in line with the findings of this study. Due to the increasing consumption of acidic beverages and changes in nutritional habits and taste preferences, it is anticipated that the prevalence of ETW can be even worse in younger age groups.

The population in Northern Finland is considered to be genetically isolated and homogenous (Sabatti *et al.* 2009), which increases the value of this study. In our study, the subjects were invited to participate all around the Oulu region, including rural areas. Furthermore, as Oulu is one of the cities that gain population from Northern Finland and countryside, it was expected that many of those invited do not originate from the city of Oulu. Overall, geographically, the sample can be considered representative, and genetically even close to ideal.

In terms of sociodemographics, education level and marital status of the participants were not found to be statistically significant variables for severe ETW, which is in line with many studies (Bartlett *et al.* 2013, Mulic *et al.* 2013, Vered *et al.* 2014). Since both healthy and unhealthy diets contain acidic items, potential differences in eating and drinking behaviors between people with different socioeconomic backgrounds seems not to be significantly associated with the risk of ETW epidemiologically.

The data about this study population also enables studying the association between long-term effects of dietary habits and the effect of individual susceptibility and ETW. ETW often develops over time, and lifestyle factors present for years may be responsible for the current ETW. On the other hand, it also takes time to be able to clinically observe the effect of biological factors, such as saliva properties, on ETW, and biological factors become more important as the individual ages. In this respect, the age of the cohort may even be considered beneficial.

6.3 Methods

6.3.1 Questionnaire

The most common method for analyzing health and health-related behaviors in large epidemiological studies is extensive questionnaires. Although the 31- and 46-year questionnaires of the NFBC1966 have some discrepancies, the aim has been to maintain them as identical as possible to allow monitoring of behaviors over time. Unfortunately, GERD-related questions were not included in the 31-year questionnaire at all, which made analyzing the long-term effects of GERD on ETW challenging. In addition, the original questionnaire included only one question on perceived oral health. The only question in it referring to ETW concerned sensitivity of teeth, which is also among problems related to dental caries. This question alone was considered too imprecise concerning the aims of the present study.

Considering questionnaires in general, the responders' ability to recall, for example, consumed food items, alcohol products, and their quantities may cause bias in the results. It is also known that people tend to give answers to questionnaires more according to a social norm that to the actual situation - a phenomenon known as social desirability (Sjöstrom & Holst 2002). However, considering the large size of the target population, questionnaires can be regarded as a reliable and an efficient manner to assess health and health-related behaviors.

All the clinically examined participants responded to a separate computer-based oral health questionnaire in connection with the clinical oral examination. This questionnaire was a modified version of the computer-based questionnaire developed in the University of Oulu and used in several studies (Anttonen *et al.* 2011, Kämppi *et al.* 2014). A dental nurse was available to assist with the questionnaire where needed. People in this age group are familiar with using the computer, and there were no problems with answering the questionnaire, as reported earlier (Anttonen *et al.* 2012). This increases the validity of the questionnaire.

Since neither the clinical examination nor the separate oral-heath questionnaire were used in previous years, causal interpretation of the observed associations was not possible. However, long-term effects of some surveyed variables, such as alcohol consumption, could be studied by using the previous and present postal questionnaires. This helped to reduce the problems related to cross-sectional studies.

6.3.2 Clinical examination

The clinical examination was planned carefully, and all the examiners were trained and calibrated according to the Health 2000 and the Oral Health of the Conscripts 2011 study protocols (Anttonen *et al.* 2012, Tanner *et al.* 2013). Prior to the study, the examiners were not familiar with the BEWE index, but they received training on the index and on ETW diagnostics in general. Differential diagnosis of tooth wear is difficult, and diagnosing early signs and symptoms of ETW is challenging even in young dentitions. Even though the examiners assessed and registered only erosion, distinguishing between erosion, attrition and abrasion is a challenge.

So far, erosion or even other types of tooth wear have not been recorded systematically in clinical practice. Inexperience in recording tooth wear and using a specific protocol may have produced some bias in the study. Some of the examiners had work experience from several decades, while some had less experience and one had only recently graduated. Overall, the experienced dentists had better inter- and intra-examiner agreements on BEWE scores when compared to the unexperienced dentists.

Considering the re-examination protocol as a whole, it was not possible to analyze reproducibility with respect to BEWE sum scores since only one jaw quadrant was re-examined. This is a shortcoming in the clinical examination protocol, which however affects only ETW studies, and was due to the strict time limits in the re-examination protocol.

As for clinical findings, with respect to caries diagnostics, the ICDAS protocol states that the teeth should be cleaned with at least a toothbrush and floss before conducting an examination (Ismail *et al.* 2007). The clinical examination in this study did not include professional cleaning; however, it is a common habit in Finland to brush your teeth before seeing a dentist. Therefore, the outcome can be considered reliable.

With respect to missing teeth, the cause of extractions could not be defined, which may cause some bias in the DMFT score. However, orthodontic extractions and tooth loss due to trauma are usually known by the person, and in this group tooth loss due to periodontal causes is anticipated to be very rare. It is also well acknowledged that radiography considerably facilitates the diagnosis of interdental and occlusal caries. However, only Fiber-Optic Transillumination (FOTI) device was used in addition to the clinical examination. Dental pantomographs (PTG) were taken of all the participants, but not bitewing radiographs. Because information offered by PTGs may not be reliable in caries diagnostics (Molander 1996), they were not used for that purpose in this study. Thus, it is likely that current restorative treatment need due to dental caries is higher than presented here.

6.3.3 BEWE

Using a proper and validated index and clear criteria to assess ETW is likely to increase the reliability of the examination. Using specific criteria also helps in finding out the right diagnosis. There are some studies that have validated the BEWE index and shown it to be suitable for recording and scoring (erosive) tooth wear in prevalence studies (Dixon *et al.* 2012, Mulic *et al.* 2010, Olley *et al.* 2014). A study on the validity and reliability of the BEWE index shows only moderate inter- and intra-examiner agreement (Dixon *et al.* 2012) suggesting that the BEWE scores should be interpreted with some caution. Our results confirm this suggestion, as the clinical examiners had some difficulty in differentiating between intact enamel and initial loss of surface texture, and consequently there was also variation in the outcomes between the examiners. The same tendency also exists in other studies, especially those with multiple examiners (Larsen *et al.* 2005, Mulic *et al.* 2010).

It can be argued that the criteria for determining the different stages of ETW should be described more precisely, or even illustrated in the BEWE classification, since the word "initial" and "distinct" can be understood in different ways. This would most likely increase the clinical usability of the BEWE index, and would standardize the interpretation of the initial findings even worldwide. Our study indicates that these interpretational problems do not seem to exist when considering more severe ETW. When using BEWE score 2 or 3 as the cut-off point in sextant agreement analysis, an excellent intra- and inter-examiner agreement was found. Thus, using the BEWE sum score and the cut-off point ≥9 for all the association analyses in Studies I, II and III is likely to give reliable results on ETW.

While the BEWE index does not cover distinguishing between enamel and dentinal lesions, we hypothesized that BEWE could also be used in ETW assessment on 3D models. According to Study IV, the use of the BEWE index on 3D models was reproducible, fast, and easy to adapt, and the BEWE index appeared to be reliable for ETW assessment on 3D models. Study IV also confirms that ETW is challenging to detect clinically, especially in its very early stages and in locations difficult to see clinically. The grading scale itself is probably not a key factor, since for example BEWE score 1 includes all forms of initial erosion.

It can be questioned if the same BEWE sum score cut-off values are valid for 3D model-assessed and clinically assessed scores, since BEWE is initially designed for clinical use. Considering that much more erosive lesions are likely to be found when using 3D modelling, it is hard to say whether there is a risk for over-treatment or a chance to prevent the lesions at the very right time. In any case, 3D models offer a modern instrument for recording and monitoring the condition.

6.3.4 Genetic data and genome-wide association study

Genome-wide association studies provide a hypothesis-free approach to scanning the entire human genome to identify genes or genetic loci that have even modest effects on complex human diseases. Since there are only a few genes that have been suggested to be associated with ETW so far (Søvik *et al.* 2015b), GWAS can be considered as a good starting point in the genetic research on ETW.

There are many advantageous features that characterize the genetic data concerning the NFBC1966. First, all the individuals in the NFBC1966 derive from a genetic isolate which is comparatively homogeneous in its genetic background and environmental exposures, and which has more extensive linkage disequilibrium than most other populations. Additionally, when a founder population has expanded recently from severe bottlenecks, as in Finland, it is known that many variants may disappear from the population while others increase rapidly due to subsampling and genetic drift. Therefore, some variants are probably enriched in Finland compared to other populations, and thus we may identify associations that may be unfeasible to detect in comparable sized samples from other populations (Peltonen *et al.* 2000). Secondly, all the participants were born in the same year, eliminating age as a potential confounder.

An independent study sample for replication is highly recommended in GWAS and would have also been beneficial in our study. The lack of replication is, however, partly counteracted by the fact that many of the associations identified in our study were supported by the literature, as they were involved in oral and tooth-related biological processes. Additionally, GWASs are ideally conducted in huge study populations, and our sample size of nearly 2,000 is in this regard small. Because of this, some of our suggestive and statistically significant loci may be true reflections of genetic risk, whereas some of them may be false positives. Likewise, we anticipate missing a large number of false negatives. We could have only used the "extreme opposite model" (comparing those with severe ETW to those with no ETW at all), but we did not want to compromise the small sample size any further. However, we performed our GWAS by also using the extreme opposite approach. Overall, bearing in mind the small sample size and the lack of replication of our GWAS, our study sample is an excellent population for a genetic study.

6.3.5 3D modelling

3D models and the CAD-CAM technique are widely used for creating imprints for prosthetic crowns or inlay and onlay restorations, but are recently also used for diagnostics. 3D technology is considered to have large potential in tooth wear studies (Van't Spijker *et al.* 2012), as well as in clinical use (Ganss & Lussi 2014). However, this study is the first one to compare ETW assessment between 3D models and clinical examination, as well as to assess the reliability of the BEWE index on 3D models.

In our study, a trained dental hygienist performed the intraoral 3D scanning after the clinical examination, and the scanning was not considered difficult or time-consuming by the operator. Considering the design of Study IV, using the same examiners to assess both the clinical appearance and 3D models could have been beneficial for the study; however, this was not possible due to timeframe of five to six years. To avoid shortcomings because of this, the 3D model examiners (two fourth-year dental students and a dentist) were calibrated by using the same protocol and the same expert as in the training and calibration of the clinical examiners. In addition, the gold standard of the clinical examination was also involved in the training and calibration process.

Having dental students as examiners of 3D model shows how easy it is to adopt the method and criteria. During the calibration process, the dental students had some problems in differentiating between normal morphologic variation and erosive lesions. The calibration was continued so long that an agreement on the findings was achieved. The two students also worked together. The intra- and inter-examiner agreements were excellent for distinct ETW, and good for ETW of any degree, respectively. The high agreement shows that even with little experience, dentists can use 3D models, for example, for monitoring ETW.

The magnification of 3D models and the ability to rotate models in the analysis program with no time limit probably facilitated the detection of ETW. In addition, the absence of saliva and other patient-related factors most likely simplified the detection of ETW on 3D models compared to the clinical examination. Initially, it was hypothesized that ETW assessment on 3D models is challenging due to the unicolor design of the model. On the contrary, even the smallest morphologic changes of surfaces were relatively easy to detect. Indeed, a 3D scanner is capable of detecting volume loss even after 1 min exposure to acid (Meireles *et al.* 2016). On the other hand, it must be remembered that although 3D models seem to be extremely precise, there may be flaws with the stitching and smoothing data gathering processes, which are not yet fully explored. However, considering all the applications for 3D models nowadays, it is anticipated that the preciseness demanded for ETW assessment meets the requirements. Additionally, it is impossible to assess the optical properties of the enamel on 3D models. Overall, the lack of a proper golden standard produces problems in studies like this, which must be kept in mind in further studies.

However, the hypothesis that lack of true colors in 3D models would make diagnostics difficult was shown not to be true. Recently, scanners that can produce 3D models with true colors have been introduced, and these are very likely to facilitate the assessment of ETW even more in the future. In addition, the use of these kinds of imprints offers a wide range of possibilities from monitoring the progression of wear to motivating patients and assessing the oral situation even with remote access.

6.4 Studied background factors associated with erosive tooth wear

6.4.1 Gender

In this population, men had severe ETW twice as frequently as females. This is in line with many previous studies (Bardolia *et al.* 2010, Fares *et al.* 2009, Mulic *et al.* 2012b), but there are still many studies that have reported no differences between the genders (Bartlett *et al.* 2013, Vered *et al.* 2014). Higher consumption of carbonated drinks and higher bite strength have been suggested as an explanation for males being more prone to ETW (Bardsley *et al.* 2004, Smith *et al.* 2006), but this has not yet been confirmed. It has also been stated that boys need about 10% more sourness and 20% more sweetness than girls to be able to recognize the taste. Boys also tend to like more extreme flavors, such as sourest sweets or super-sweet soft drinks (Gambon *et al.* 2012). Thus, also taste preferences for sour foods and drinks may play a role in the development of dental diseases such as erosion or dental caries. In our study, men also had statistically significantly more restorative treatment need due to dental caries than females, but there was no difference between the genders concerning DMFT, which in line with the Health 2000 study (2017).

Recently, Uhlen *et al.* (2016b) showed that enamel in males seems more prone to ETW than that of females, and that susceptibility to enamel erosion may be influenced by genetic variation in enamel formation genes. This is truly an interesting topic for future research as it changes our perspective on the susceptibility to dental diseases and is also highly relevant in terms of future clinical dental practice. In our Study III, genetic variation also seemed to influence ETW susceptibility.

Furthermore, males and females had different loci in the genome significantly associated with ETW. Among males, the most interesting SNPs were near the genes known to be associated with tooth development. Among females, many of the most interesting SNPs were close to the regions suggested to be associated with dental caries (Eckert *et al.* 2017, Shaffer *et al.* 2013). To make it even more complicated, in studies concerning susceptibility to dental caries, it is females that usually exhibit a higher prevalence and severity of dental caries across all ages than males (Lukacs & Largaespada 2006). The reason for this is not fully understood, even though earlier tooth eruption in females, differences in dietary behaviors, utilization of oral care, hormones, and characteristics of saliva have been proposed (Lukacs & Largaespada 2006). In a study by Shaffer *et al.* (2015), gene-by-sex interactions for caries experience were observed for the first time, suggesting that different genes seem to play important roles between the genders. Thus, it can be cautiously suggested that the structure of enamel and susceptibility to dental diseases may indeed be influenced by gender and genetics, even though the mechanisms still need extensive research.

6.4.2 Dental caries

In this study, restorative treatment need due to caries increased the odds for severe ETW, and extensive restorative treatment need even more. Additionally, past caries experience was positively associated with present ETW. Since both caries and ETW have common etiological factors, such as snacking, high or constant intake of sweetened soft drinks, and low saliva secretion, this association is understandable.

With respect to DMFT, in the analyses, all missing teeth were coded as missing due to caries, since it is difficult to find out the real cause of extraction in a cross-sectional study. In this age group, however, it is likely that most of the extractions are due to dental caries. In the literature, the association between caries and ETW is not clear, and the number of studies on the subject is limited. Some studies report statistically significant association between ETW and dental caries (Isaksson 2013, Mulic *et al.* 2013) while others do not (Auad *et al.* 2009, Truin *et al.* 2005). At first, it was assumed that this association is true only for ETW of extrinsic origin and can be explained by only eating and drinking habits. However, individuals with erosion have been suggested to possess similar salivary characteristics to caries-active individuals (O'Sullivan & Curzon 2000). Additionally, it seems that an acidic environment changes the acquired pellicle so that it becomes more sensitive to erosive attacks (Carpenter *et al.* 2014), and thus altered pellicle due to intrinsic acids may also increase the risk for ETW. The role of this altered pellicle with respect to dental caries is not known.

Recently, it was also reported that some of the genes involved in the enamel development (*AMELX* and *AMBN*) are associated with calcium and phosphorus levels in saliva (Küchler *et al.* 2017). In fact, this is the first study to report genetic variations contributing to calcium and phosphorous levels in saliva. Such variations may increase individual susceptibility to dental caries. Considering this, it is reasonable to assume that this genetic variance in saliva also plays a role in ETW susceptibility. Interestingly, in our candidate gene study, *AMBN* was the only enamel formation gene showing association, even though weak, with ETW. Furthermore, some of the significantly ETW-associated loci in the genome in Study III have also been suggested to be involved in the dental caries process (Eckert *et al.* 2017, Küchler *et al.* 2013, Shaffer *et al.* 2013). As both the ETW and dental caries processes eventually cause destruction of dental hard tissue as a consequence of an acidic environment, this suggested association is intriguing

Clinically this might imply that those prone to dental caries may also have an increased risk for ETW, and vice versa. Epidemiologically it also seems that those already needing restorative treatment for dental caries could be regarded as possessing a risk for ETW, and vice versa. Evidence is still needed.

6.4.3 Gastro-esophageal reflux-disease

Symptoms of GERD have been identified as one of the most important risk indicators for ETW (Bartlett *et al.* 2011, Bartlett *et al.* 2013, Moazzez & Bartlett 2014). In this study, daily symptoms of GERD increased the odds for ETW almost four-fold, supporting these previous studies. According to the literature, the association between diagnosed GERD and ETW seems even clearer (Holbrook *et al.* 2009, Moazzez & Bartlett 2014, Pace *et al.* 2008), even if contradictory results have been presented as well (Di Fede *et al.* 2008, Jensdottir *et al.* 2004). Once gastric acids have caused heartburn symptoms, the gastric acids may or may not progress into the mouth, which probably explains the difference between studies. Although one in eight reported symptoms of GERD daily or weekly here, self-reported gastric problems appear to be a minor factor in the ETW process epidemiologically. Even though frequent or long-term symptoms of GERD seem to increase the risk for ETW on an individual basis, self-reporting alone may not be reliable enough when assessing the etiology of ETW. For instance, silent reflux, i.e. GERD without symptoms, may mislead the clinician and cause bias in studies based on symptoms, such as the present one. Our hypothesis that ETW is more severe among people with frequent symptoms of GERD turned out to be valid, but the low prevalence of diagnosed GERD was surprising. Self-reporting and the fact that participants were only 46-years old may partly explain this. Overall, GERD remained to be one of the most important intrinsic factors of ETW, even though it seems that heartburn from time to time does not play an important role in the etiology of ETW.

6.4.4 Eating disorders

Eating disorders affect the regulation of food intake, for example, due to restricted dietary choices and induced vomiting, which is why these conditions can also impact oral health. In general, there are only a few studies on the prevalence of ETW in persons having eating disorders (Schlueter & Tveit 2014). However, people with eating disorders seem to be at high risk for both exogenous-caused erosion due to restrictive diet and endogenous-caused erosion due to episodes of vomiting (Johansson *et al.* 2012, Schlueter & Tveit 2014). In our study sample, there were no diagnosed eating disorders according to hospital registers, and only a few cohort members reported having had eating disorders in the questionnaire. Consequently, eating disorders were not investigated further in our study.

6.4.5 Alcohol consumption

Alcohol consumption was classified among intrinsic factors in this study, even though many alcohol products, such as wines and long-drinks are erosive itself. Alcohol consumption (g/day) was determined on the basis of several questions concerning the amount of different alcohol products consumed and drinking habits. However, it is known that people are likely to underestimate the consumption of unhealthy products (Simpura 1987), and thus it is possible that the consumption of alcohol is even higher than presented here. In this study, we were able to analyze the long-term effect of consuming alcohol, since the questions about alcohol use were also present in the 1997–1998 questionnaire.

People with chronic alcoholism, as well as those who reported heavy use of alcohol, especially already in the 31-year questionnaire, had more ETW in this study population than the rest, but the association between the conditions was not statistically significant in the regression model. Only a few studies have investigated the association between ETW and alcohol abuse, and our findings are in agreement with them (Dukic *et al.* 2010, Robb & Smith 1990, Teixeira *et al.* 2017).

The multifactorial etiology of ETW complicates finding a clear association between alcohol consumption and ETW. Alcoholics and heavy alcohol users may also have a reduced salivary flow rate, and they might suffer from GERD and bruxism—all of which may lead to susceptibility to erosive wear. Additionally, many alcohol products itself are erosive (Lussi *et al.* 2012). Especially wine, both red and white, is known for its erosive potential; however, in Finland the consumption of wine has increased only recently and wine is used less than other alcoholic products (THL, 2017).

In this study, no differences were observed between alcoholics and non-alcoholics with respect to saliva secretion or possible sleep bruxism, but daily symptoms of GERD were three times higher among alcoholics than the rest, supporting the idea that gastric acids play an important role. This is in line with the literature, since it has been proposed that the main source of acids causing tooth wear among alcoholics is regurgitated or refluxed stomach juice (Moazzez & Bartlett 2014, Smith & Robb 1989).

Long-term alcohol consumption seemed to have a cumulative effect on ETW, even with low doses, which is an interesting finding and has not been reported before. However, the reason for this can only be speculated. Probably this influence is also of extrinsic origin. Consuming alcohol might also refer to some life-style habits that people not consuming alcohol do not have. Understanding the effect of heavy, or low but long-term, alcohol consumption on tooth wear could be beneficial in future preventative alcohol programs, but more research is needed to confirm these findings.

6.4.6 Saliva

Saliva composition and flow rate, along with acquired salivary pellicle, play a major role in the ETW process (Buzalaf *et al.* 2012) and seem to be one of the key factors in susceptibility to erosive wear. The flow rate of stimulated saliva was not associated with ETW in this study, but instead, low unstimulated saliva flow rate was a considerable risk factor for severe ETW (OR 3.8), the finding being in line with other studies in the field (Hara & Zero 2014, Järvinen *et al.* 1991, Rytömaa *et al.* 1998). As this cohort ages and medications become more prevalent, the role of hyposalivation is likely to increase along with the risk for further progress of ETW.

In the future, it would be interesting to conduct a more in-depth analysis of the components of the saliva of the cohort members, since the protein content of the saliva seems to play a crucial role in the development of dental erosion (Hellwig *et al.* 2013). It is still not known which components are most important. Changes in the components of the saliva in late pregnancy and during lactation, for instance, have been suggested to temporarily predispose women to dental erosion (Laine

2002). However, based on our results, it seems unlikely that pregnancy-related changes in the salivary composition would be substantial enough to cause increased susceptibility to erosive wear. However, the multifactorial etiology of ETW complicates drawing conclusions on the issue. Since the protein content in saliva varies, and is likely related to genetic inter-individual variance, saliva may also be a factor to explain the genetic findings in Study III. Interestingly, the only statistically significant SNP in the whole sample was located near a gene coding peroxidasin, which again is one of the main salivary antioxidants. Considering the individual susceptibility to ETW, it has even been stated that saliva factors and tooth surface composition may be at least as important as the frequency of acid attacks (Schlueter & Tveit 2014).

6.5 Genetic component of erosive tooth wear

The interest in understanding the mechanisms underlying the individual susceptibility to oral diseases coincides with the development of feasible approaches to study complex human diseases such as cardiovascular diseases and cancers to gain a better understand of genetic susceptibility. There has been much interest and progress recently in identifying genetic contribution to caries (Vieira *et al.* 2014), but ETW has just started to attract interest and no GWAS on ETW had been performed before our study.

In our GWAS, the strongest evidence of the association with ETW in the whole sample was found in the SNP rs11681214 with two suggestively associated SNPs located nearby. This SNP is located near the gene peroxidasin (*PXDN*) which is known to encode a proteolytic enzyme, peroxidase, which is the main salivary antioxidant and is found in the organic matrix of enamel pellicle (Hannig *et al.* 2008). Although the function of this enzyme in relation to ETW is not known, the possible link is interesting and would deserve more studies. Interestingly, close to the region where this SNP exists, a suggestively associated polymorphism was observed in a GWAS on dental caries (Shaffer *et al.* 2013), which suggests an interesting link between the GWASs on ETW and dental caries. This link was further supported by our findings in the GWAS on females, where the SNPs in the regions 4q21, 5q13.3 and 13q34 reached the statistical threshold. All these regions are near the ones proposed by Shaffer *et al.* (2013) in their GWAS on dental caries. Regions 4q21 and 5q13.3 have even been studied further (Eckert *et al.* 2017, Shimizu *et al.* 2013).

Based on all the GWASs performed in this study, it seems that individual susceptibility is influenced by genetic variance as well. All the GWASs exhibited SNPs that were statistically significantly associated with erosive wear. However, this approach needs further studies and serves more as a launching pad for ETW gene research.

6.5.1 Genome-wide association study using the extreme opposite approach

Two SNPs met the statistically significant threshold: one near the *Homeobox A* gene family and another near the gene *ZWINT*. Both SNPs were found suggestively significant also in the whole-sample GWAS. *Homeobox* genes are known to be involved in tooth morphogenesis (Suryadeva & Khan 2015), however, specific data on the *Homeobox A* gene family with respect to tooth development is not yet available. *ZWINT* is involved in the kinetochore function, although its exact role is not known (Woo Seo *et al.* 2015). This locus was also close to a region reported to be associated with dental caries (Shaffer *et al.* 2013).

Additionally, two interesting clusters of suggestive SNPs existed, one near the gene *MSX1* and another in the gene *GLRA3* and near the genes *FBXO8* and *SCRG1*. The cluster near *MSX1* deserves special attention since *MSX1* is one of the most important genes involved in tooth development (Suryadeva & Khan 2015). This can be considered one of the most interesting findings made by using this approach and would deserve future research. *GLRA3* encodes a member of the ligand-gated ion channel protein family, but there is no data on it considering the oral environment. However, *FBXO8* has been reported to be involved in the development of bilateral cleft lip (Calcia *et al.* 2013) and *SCRG1* in vertical tooth movement (Suda 2008).

6.5.2 Genome-wide association study on males

The top SNPs in the GWA study on males were located near the gene *FGFR1*, which has been proposed to be the one of the important receptors in human tooth development (Huang *et al.* 2015) and to be involved in molar-incisal hypoplasia (Jeremias *et al.* 2016). Additionally, this gene has been shown to participate in the regulation of differentiation and secretory functions of odontoblasts and ameloblasts (Kettunen *et al.* 1998), and its role has been found to be critical in the formation of proper enamel (Takamori *et al.* 2008).

In chromosome 20, there was a cluster of SNPs, including three significantly associated ones located near the gene *CDH4*. Recently, it was shown that *CDH4* promotes the expression of E-cadherin (Xie *et al.* 2016), which plays a crucial role in enamel development (Guan *et al.* 2016, Li *et al.* 2012). Overall, according to our study, it seems that some of the genes related to enamel development may be associated with severe ETW among males.

6.5.3 Genome-wide association study on females

In the GWAS on females, the loci exhibiting the strongest evidence of association were in the gene *SCD5* and near *F2R* and *ING1*. *SCD5* impairs cathepsin B secretion (Bellenghi *et al.* 2015), which is expressed during the maturation stage of the enamel (Tye *et al.* 2009), and also apparently has a role in dentin degradation (Vidal *et al.* 2014). *F2R* has recently been suggested to be highly expressed in secretory stage ameloblasts (Simmer *et al.* 2014). This locus was also suggestive in the whole-sample GWAS and has been further studied in a fine-mapping study concerning dental caries (Shimizu *et al.* 2013). ING1 regulates cell proliferation, apoptosis, and differentiation by multiple pathways (Zhang *et al.* 2016), but there are no studies on it in relation to the oral environment.

6.6 3D models in erosive tooth wear assessment

3D models were found convenient to analyze, and the assessment using the BEWE index was reproducible. Overall, ETW was scored more severe on 3D models when compared to clinical examination, and there was a moderate agreement between the outcomes of 3D models and clinical examinations. Similar agreements between the clinical examination and indirect methods on dental erosion have been reported previously (Hove *et al.* 2013, Wetselaar *et al.* 2009). Sensitivity for severe erosive wear was relatively low on 3D models. This may be explained by the fact that at times it was hard to distinguish between eroded tooth surface and a worn filling. Additionally, the increased sensitivity in early wear detection by using 3D models is possibly one reason for the lower levels of agreement in the clinical examination. It seems that when considering the assessment of ETW, 3D models and clinical examination are not fully comparable, and 3D models are not to replace the clinical assessment of ETW.

Upper posterior sextants exhibited significantly more erosive wear in 3D models when compared to lower posterior sextants, but in clinical examinations no such difference was found. We speculate that this inter-method difference concerning posterior sextants may be due to the poorer visibility and light when clinically assessing upper posterior teeth compared to lower ones and 3D models. Additionally, in this age group, the first lower molars are often restored due to caries, and signs of ETW are not easy to detect. This may also partly explain the lower intra-method reproducibility of the lower posterior sextants. The predominance of the upper posterior sextant has not been reported previously. Based on these observations, detecting erosive wear clinically in upper posterior sextants must be emphasized.

As a conclusion, study IV shows that 3D models could serve as an additional assessment tool and offer substantial advantage compared to the clinical examination alone when assessing and monitoring ETW, especially in the initial stages of the condition.

6.7 Future studies

As the research on ETW is attracting more attention, data on ETW from different perspectives is under continuing research. Especially longitudinal studies on the etiology of ETW with questionnaires precise enough are needed. Our postal and computer-based oral health questionnaires were utilized only partially in this study and would have more potential for future studies. They would also enable studying the role of extrinsic factors related to ETW more in detail, somewhat longitudinally as well.

On the other hand, one of the most intriguing future research lines could be the individual differences related to saliva and pellicle, as well as the genetic variance related to ETW. A lot of research currently focuses on products preventing erosive wear, and as the role of, for example, different proteins in saliva or in pellicle are revealed, we might be closer to finding the best means to prevent ETW as well. Extensive research is, however, needed before this can be reality. One of our aims in the future is to study the saliva of the examined cohort members more in-depth.

Diagnosing ETW is still a challenge. A validated questionnaire planned especially for assessing the etiology of ETW would be most valuable and is another topic for future study. It has been proposed that the BEWE index would benefit from this kind of an additional questionnaire (Margaritis *et al.* 2011). Future studies are also needed on the use of 3D models in ETW assessment, and especially in the assessment of the progression of erosive wear, since the method seems to show promise for that as well.

It would also be interesting to find out how knowledgeable Finnish dentists are on ETW, as we anticipate that there is probably a need to train dentists more on ETW diagnostics and on different indices.

7 Summary and conclusions

This study offers unique data on ETW among middle-aged adults from Northern Finland. According to the study, ETW is common among middle-aged people, although severe forms of ETW are quite rare. The prevalence of those in need for preventive treatment need was almost 50% according to the BEWE index protocol. The prevalence of untreated dental caries is also still high, and it seems that the pressure towards restorative treatment need and more complicated treatments will increase in the future.

Furthermore, there seems to be an association between ETW and dental caries occurrence. Considering the lifestyle and eating and drinking habits of today's young people, it can be assumed that ETW will become even more common in the future, and preventing and treating the condition may even demand new strategies from dentists and public dental care providers.

ETW is a multifactorial condition, and of its intrinsic factors, daily problems related to GERD and hyposalivation were among the strongest risk factors for ETW in our study population. A bit surprisingly, long-term alcohol consumption seems to be associated with ETW even with low doses. However, the influence of alcohol consumption on ETW status is likely to be also of extrinsic origin. This new finding needs more research, and at this stage, it could at least encourage dentists to go through all dietary and drinking habits of the patient when assessing the etiology of erosive wear.

Over a hundred years ago, G. V. Black hypothesized that the origin of erosive wear might be hereditary, but he also named other possible factors of systemic or extrinsic origin (Black 1908). Even though a lot has changed over the years, he was surprisingly right with his thoughts on the etiology of wear. When just a few decades ago the existence of "poor teeth" was denied, now there is increasing evidence of the individual variance in the susceptibility to oral diseases. Yet, further evidence is still necessary. In addition to intrinsic and extrinsic risk factors influencing ETW, it seems that the genetic polymorphism also plays a role in the erosion process. Even though there is still a long way to go, eventually the field of dentistry might be shifting towards customized dentistry along with the developments in gene testing and analyzing.

In the end, the very next step should be to encourage dentists to diagnose ETW even at the early stage and start preventing the condition before it progresses and makes treatment more complicated. According to our results, the BEWE index suits well for the purpose and it is easy to use and easy to learn. It also guides through the treatment planning. The BEWE criteria are also reliable when used on 3D models, and could even be used for monitoring ETW, as well as, for example, in tele-dentistry in the future.

It would also be of great value to inform the general public on ETW process, the etiological factors related to it, and the consequences ETW may cause. By the means of disseminating the information on ETW for dentists, for individuals and for general public, our study could accomplish the final aim of the NFBC studies of promoting health and wellbeing among the population.

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Original papers

I Alaraudanjoki V, Laitala ML, Tjäderhane L, Pesonen P, Lussi A, Anttonen V (2016) Association of erosive tooth wear and dental caries in Northern Finland Birth Cohort 1966 – an epidemiological cross-sectional study. BMC Oral Health 17(6): doi: 10.1186/s12903-016-0232-x

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IV Alaraudanjoki V, Pesonen R, Saarela H, Laitala ML, Tjäderhane L, Kiviahde H, Lussi A, Anttonen V (2017) Is Basic Erosive Wear Examination (BEWE) suitable for recording erosive tooth wear on 3D models? Journal of Dentistry 59: 26-32.

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