Bonding Strength to Teeth with Amelogenesis Imperfecta

In partial fulfillment of the requirement for the degree of Dental Doctorate in Paediatric Dentistry

Paediatric Dentistry Department

Eastman Dental Institute

University College London

2018 - 2021

Submitted by:

Husam Hashil Hamed Al-Siyabi

BDS (OMAN), MFD – RCSI (Ireland)

Project supervisors:

Prof. Paul Ashley (Department of Paediatric Dentistry, Eastman Dental Institute-University College London)

Dr. Susan Parekh (Department of Paediatric Dentistry, Eastman Dental Institute-University College London)

DECLARATION OF WORK

I, Husam Hashil Hamed Al-Siyabi confirm that the work presented in this thesis is my own. Where information has been derived from other sources, I confirm that this has been acknowledged and indicated in the thesis.

Husam Hashil Hamed Al-Siyabi

Eastman Dental Institute, University of College London

23 July 2021

Abstract

Background

Amelogenesis Imperfecta (AI) is an inherited dental condition of enamel, which can cause increased tooth sensitivity, difficulties maintaining oral hygiene, rapid tooth loss or enamel fractures, as well as defects in enamel thickness, colour, and shape. All these factors may impair aesthetic appearance and the masticatory ability, requiring dental treatment for a lifetime which may affect their overall quality of life.

Aim:

1. To assess the burden of care for children with AI attending the Paediatric Dental Department at the Eastman Dental Hospital (EDH).
2. To review the available scientific evidence on the adhesive interface between AI affected teeth and restorative materials.
3. To analyze different AI classifications quoted in the literature, to determine the consistency and standardization of reporting on AI classifications.

Material and method:

A service evaluation of AI patients being treated in the department from 2002-2019.

Two systematic searches were conducted using search terms in both electronic and hand search journals.

Results:

The burden and impact of care audit showed the average number of appointments per year was 5, (SD=2.5). The average distance travelled to the hospital was 33.7 miles (SD =30 miles) and the treatment provided included:

- Extractions - majority in hypoplastic group (78%, n=10).
- Composite restorations - hypoplastic (66%, n=18), mixed (16%, n=4).
- Indirect coronal restorations – hypoplastic (67%, n=17), mixed (12%, n=3).
- Bleaching and microabrasion - most performed in hypomature group (56%, n= 8 and 67%, n=5 respectively).

- More failed composite restorations occurred in hypocalcified (25%, n=4) and mixed type (23%, n=40) with debonding being the most common reason.

In the systematic review of bonding strength to AI affected teeth, studies showed a lower bonding strength of AI affected teeth in comparison to sound teeth. Bonding strength of composite was not significantly different when using self-etch compared with etch and rinse adhesives and deproteinization with sodium hypochlorite had no effect on shear bond strength, but chlorine dioxide and sodium fluoride showed better values in enhancing bonding strength.

In the review of classification of AI papers did not use or cite a classification (n=12, 36%) and 43% only described the phenotype with no information regarding the basic genetic information if known. Those results are elaborated ineach chapter in details.

Conclusion:

The service evaluation provides data on the burden of care for children with AI. The high number of appointments, treatment needs, and miles travelled illustrate the scope ofcomplications that can occur and stress the need for comprehensive management of this condition. Lower bonding strength values and durability of restorations to AI affected teeth which requires further laboratory studies. There are variations and inconsistency of classification used for studies published from 2015.

Impact statement

This study aims to look at the available evidence on bond strength to teeth with Amelogenesis Imperfecta which will contribute to the wider discussion on AI affected teeth. With the current need of further understanding of the condition and the problems associated with it and therefore, to take appropriate actions for conducting laboratory and clinical trials which will bring the desired change and improvement of the quality and durability of restorative treatment provided.

This study also explores the burden and impact of care in AI treatment on children and their families. AI has a high burden of care for both patients and caregivers, and further efforts to reduce this burden must be carefully considered. It provides an insight on how critical to provide more accessible treatment pathways for patients to obtain timely care, which will minimize the psychosocial impacts, enhance their quality of life and self-confidence. This study was presented and published in the European Academy of Paediatric Dentistry, where researchers from all around the world gather to interact and exchange knowledge.

Another significant impact of the figures showed in this study for the clinicians and researchers when reporting AI condition, which might help to lead to the development of a standardized form for reporting AI and its types and thus, improve our knowledge and ability to compare types and classification of AI affected teeth more easily.

Overall, it provides an insight for the need for continuing of research and development to prevent and manage this condition which will enhance the care provided for patients affected with AI.

ACKNOWLEDGEMENT

After three years of hard efforts and sacrifice, Alhamdulillah, I was able to finish and submit my thesis. First and foremost, I would like to express my deepest appreciation and gratitude to my family. Especially my father, for his unwavering support, constant advise, and encouragement during my entire journey. I would like to dedicate this work to my mother who passed away on the 4th of April 2021. “There are no words of appreciation that will ever fulfil your worth”, thank you for being always with me.

A warm appreciation to my great mentors and supervisors, Dr Susan Parekh, and Professor Paul Ashley for their exceptional guidance and support.

To all the staff I have met and worked with in the unit of Paediatric Department in Eastman Dental Hospital / Institute and in Great Ormond Street Hospital, thank you so much for your lovely companionship.

To my dear colleagues, Hasan Jamal & Violeta Kakiora, who were supportive during our DDent journey that we experienced together. We had together moments of happiness and despair for the last three years. I would like also to thank my other DDent colleagues for their support.

Contents

Table of Contents

List of Tables

List of Figures

List of Abbreviations

ADAI	Autosomal Dominant AI
AI	Amelogenesis Imperfecta
ARAI	Autosomal recessive AI
ASA	American Society of Anesthesiologists
CDR	Cone Rod dystrophy
CEJ	Cemento - Enamel Junction
DDE	Developmental defect of enamel
EB	Epidermolysis Bullosa
EDH	Eastman Dental Hospital
EDI	Eastman Dental Institute
EDI	Enamel Defect Index
EDJ	Enamel Dentinal junction
ER	Etch and Rinse
FGF	Fibroblast growth factor
GA	General Anaesthetic
GDP	General Dental practitioner
GOSH	Great Ormond Street Hospital
HSB	Hunter Schreger bands
IEE	Inner enamel epithelium
IU	Intrauterine
KHN	Knoop hardness number
LA	Local Anaesthetic
OEE	Outer enamel epithelium
PEB	Post eruptive breakdown
PMC	Preformed Metal Crown
REE	Reduced enamel epithelium
SD	Standard Deviation
SE	Self-Etch
SEM	Scanning Electron Microscopy
VHN	Vickers hardness number
NOS	Newcastle Ottawa Scale

Statement of problem

Amelogenesis Imperfecta (AI) is a rare genetic disorder of enamel, patients may have increased tooth sensitivity, difficulty keeping good oral hygiene, loss of tooth structure, in addition, abnormal in the enamel thickness, colour and shape; variables that may affect aesthetic appearance and masticatory efficiency (Lundgren et al., 2016) which necessitates lifelong dental care which may affect their overall quality of life. Patients with AI have had a detrimental effect on quality of life which involve area such as phycological distress, social and physical disabilities (Hashem et al., 2013). The Eastman Dental Hospital (EDH) has one of the largest cohorts of AI patients in the United Kingdom and therefore, it is well placed to investigate dental care for this population. As a result, we conducted a burden of care audit to determine and assess the impact.

Treatment with AI is based on individual diagnosis and phenotype. Optimum care of patients considers the developmental stage of the patient. A retrospective review of restorative treatment in AI affected patients, found that all AI affected patients are favorably influenced by their restorative treatment, and almost half of the patients preferred treatment to be carried at an earlier age (Lindunger and Smedberg, 2005).

The burden and impact of AI treatment on children and their families have not been well studied. Understanding what treatment in different types of AI regularly require, the commitment and time investment of families and patients, the travel distance and the time required to attend a single appointment should form the basis for advancing treatment strategies and aim to improve AI management with focus on the standard of care provided to patients. The burden of care service evaluation was done in conjunction with Leeds University, to increase our sample size and compare between the two geographical sites. One of the findings of the study was that the type of AI had an impact on restoration failure and the need for repeated treatment, so it was important to look at the available evidence for bonding strength to AI affected teeth.

Bonded restorations intended to improve the poor aesthetic in patients with AI. However, high failure rates of bonding have been reported for bonding to AI affected teeth (Lindunger and Smedberg, 2005). The bond between enamel and restoration is extremely reliant on the nature of enamel surface. Hypocalcified, Hypomature and hypoplastic AI have abnormal matrix formation. Differences in morphology and micro morphology compared to normal enamel and irregular etching patterns. The higher organic content in the hypocalcified and hypomature AI more than the hypoplastic AI is the cause of higher failure rate of bonded restorations.

The basic concept of bonding to teeth is dependent on the micromechanical interlocking of adhesive resin with enamel and dentine. Contemporary resin bonding systems are classified into two groups; self-etching (SE) adhesive systems which include etching and priming in one step, whereas etch and rinse (ER) adhesive systems involve etching with phosphoric acid (Sarr et al., 2010). In etch and rinse adhesives, a phosphoric acid conditioner usually used to degrade the hydroxyapatite crystals and produces gaps for penetration. This has proven a successful method of bonding to sound enamel, because of the high mineral content (Erickson et al., 2009). As the substrate structure influences the effectiveness of adhesive agents, the decrease mineral content of AI affected enamel could be detrimental to the bonding process (Şaroğlu et al., 2006).

The different classifications descriptions have been used to describe AI in our systematic review and the classification quoted in the clinical records by the clinicians in the audit making it difficult to compare studies. Therefore, we decided to look at the standardization and different classifications quoted in the past years.

1. Background

Amelogenesis imperfecta (AI) is a hereditary development disorder of structural defects in enamel caused by ectodermal disruptions. As a consequence of impaired amelogenesis, both primary and permanent teeth are affected (Chan et al., 2010). Based on the stage of enamel formation that is impaired by the genetic mutation, AI can be categorized into hypoplastic, hypocalcified or hypomature and mixed forms (Aldred et al., 2003). The clinical appearance of hypoplastic AI affected teeth is usually thin enamel, surface pitting, or vertical grooving, whereas the forms of hypocalcified and hypomature AI are distinguished by the presence of normal quantities of deficiently mineralized enamel matrix. The decreased hardness of hypocalcified enamel means it is more likely to wear away, exposing dentine which results in a modified structure and mineralization (Hyun et al., 2009).

The clinical consequence of AI is the cosmetic concern with the appearance of the teeth. The causes of poor aesthetics are due to the abnormal shape of the crown because of enamel loss, surface roughness and discoloration (Canger et al., 2010). Several treatment modalities have been suggested including extraction of affected teeth and fitting of a removal or fixed prosthesis. However, this method is sometimes extremely invasive and has a significant risk of consequences. Therefore, the use of bonded composite restorations is a far more conservative option (Nathwani and Kelleher, 2010).

Bonding to enamel and dentine is based on micromechanical interlocking of adhesive resin with dental hard tissues. Although enamel bonding is dependent on the micromechanical retention to the etched substrate, the adhesion of dentine occurs by hybridization with the exposed collagen network (Van Meerbeek et al., 2003). A conditioner is usually used to dissolve the hydroxyapatite crystals which creates spaces to allow the resin to infiltrate, this has proven a successful method of bonding to normal healthy teeth due to the high mineral content of enamel. However, bonding to AI affected teeth has been reported to have a high rate of

failure and the durability of dental restorations is unknown because of the substrate structure influencing the efficacy of adhesive material, the decreased mineral content of AI affected teeth could be detrimental to the bonding quality to varied degrees depending on the type of AI.

1.1. Mechanism of tooth development

The tooth germ is formed from an aggregation of cells which are derived from the oral surface ectoderm of the first branchial arch, the underlying mesenchyme of the neural crest and the frontonasal prominence which form teeth in human embryo (Ten Cate and Nanci, 2013). The tooth germ comprises of three separate components: the enamel organ, dental papilla, and the follicle or dental sac. The enamel organ consists of the outer enamel epithelium (OEE), inner enamel epithelium (IEE), the stellate reticulum and stratum intermedium. The function of the enamel organ is not limited only to the formation of enamel, but they also play a significant part during dentine formation and the dento-gingival junction establishment. However, it becomes part of the reduced enamel epithelium (REE) well after enamel maturation as shown in Figure 1-1 (Ten Cate and Nanci, 2013).

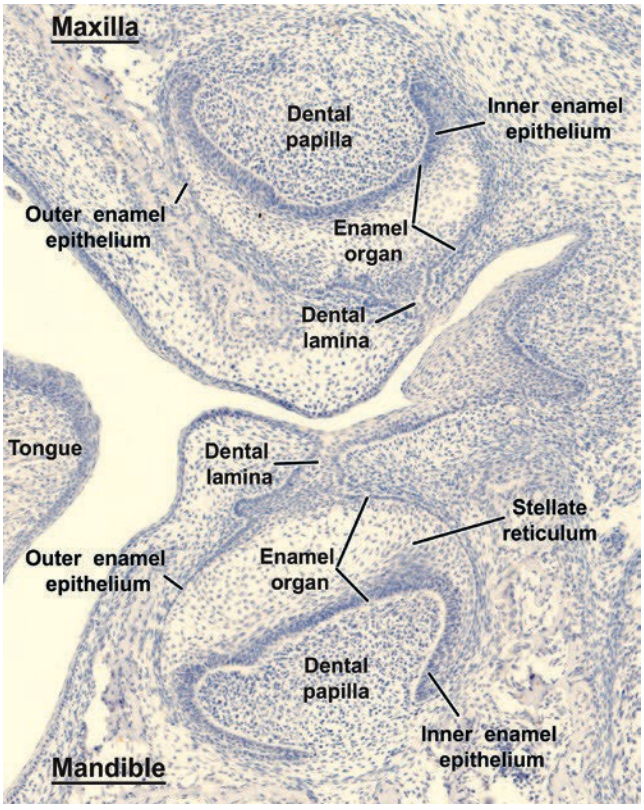


Figure 1-1 showing the different parts of enamel organ during cap stage of tooth development (Nanci, 2017).

The stages of tooth development are described as bud, cap, and bell stages. The crown and root development are shaped by these stages respectively as described in Figure 1-2 (Lacruz et al., 2017). Mesenchymal cells of the dental papilla that lies below the enamel organ forms the dentine through odontoblasts, whilst the middle part of the papilla create the pulp. Epithelial cells which lie beside odontoblast develop into ameloblasts, that give rise to enamel (Bailleul-Forestier et al., 2008).

The cap phase starts by the 11th week of intrauterine life. Morphogenesis is the biological process which causes organisms to evolve their shape progresses, in this case with the invagination of the enamel organ to create a cap-shaped structure. The size and shape of each tooth becomes clearer at this stage, this mechanism is controlled by the enamel knot. The impulses from the enamel knot control the development and dictates the locations of the epithelial folds corresponding to the cup pattern. Subsequently, the cap-shaped structure becomes the enamel organ that covers the dental papilla and produces enamel, dentine, and pulp respectively (Tummers and Thesleff, 2008).

The bell phase is also known as the phase of ‘histodifferentiation’ and ‘morphodifferentiation’. It is divided into two phases: early and late bell phase, as it begins at the 14th week of intrauterine life. Most cells are termed stellate reticulum due to their star shaped appearance. The enamel organ is bell shaped at this point. The primary role of this cell is to safeguard the dental tissue underneath against physical damage and to preserve the form of the tooth (Ten Cate and Nanci, 2013).

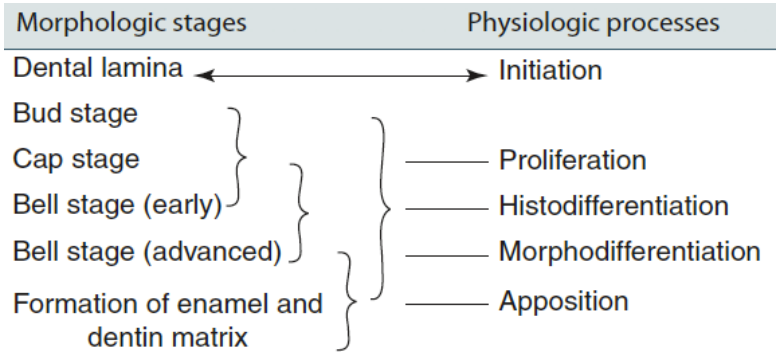


Figure 1-2 stages of tooth development displaying overlap different physiological processes and morphological stages of tooth formation, with the exclusion of the initiation stage (Soxman et al., 2019).

The outer enamel epithelium (OEE) is thought to be involved in maintaining the shape of the enamel organ. The cervical loop, located at the boundary of the enamel organ. This is where the OEE is linked with the IEE, has a high cell division activity. During the bell phase, the IEE is first seen, when the cells distinguish into ameloblasts and generate the enamel matrix. After the ameloblast cells are created, the dental papilla starts to distinguish into odontoblasts. For the enamel to form, the dentinal layer must be present. However, the existence of ameloblasts is also crucial if dentinogenesis is to proceed (Thesleff and Juuri, 2015). The ameloblast and odontoblast cells are accountable for the future development of enamel and dentine. Dentine formation begins when the basement membrane of the IEE is thickened. After dentine development begins, the inner enamel epithelium cells release an organic matrix that protects the dentine. During the oppositional phase the formation of dental hard tissue begins which is also known as the late bell phase. It happens at about 18th week of IU life. At this point, the dental lamina begins to dissolve as the developing tooth descends into the oral cavity (Ten Cate and Nanci, 2013). Nevertheless, when remnant of dental lamina called gland of Serres if not resorbed that may form in eruption cyst (Berkovitz et al., 2017).

The growth of permanent dentition is launched by the lingual expansion of the OEE in the primary teeth, which subsequently forms the tooth germ of the permanent successors. For the permanent molars that do not replace the primary teeth, the epithelial tissue of primary second molar grows backwards to bud off effectively the first, second and third molars teeth. The development of tooth buds for the first permanent molar appears in utero at about 4 months, the second permanent molar appears about 6 months after birth, whereas the third permanent molar appears 4-6 years of life (Berkovitz et al., 2009).

1.2. Enamel

Enamel is a distinctive mineralized tissue because it is acellular and is composed of crystallites that are bigger and more directed than other mineralized tissues. Due to its mineralized complicated composition, enamel is adjusted throughout its lifetime to absorb mechanical and abrasive stresses (Simmer and Fincham, 1995). As it is the hardest tissue of the body, in contrast to the bone it cannot be remodeled once its mineralized. Enamel is made up of about 95% mineral of its weight, 1-2% organic material, and about 2-4% of water (Lacruz et al., 2017). The inorganic content comprises of crystalline calcium phosphate recognized as hydroxyapatite crystals (Robinson et al., 2017). The densely packed carbonated hydroxyapatite is organized into a woven framework called rods or prims (White et al., 2001). which is more fracture resistance (Margolis et al., 2006, Ruan and Moradian-Oldak, 2015).

Unlike bone or dentine, enamel does not contain collagen. Nonetheless, enamel does contain distinctive proteins termed amelogenin and enamelin (Moradian-Oldak, 2012). Enamelin is a minor element of the matrix that regulates the mineralization of enamel crystals; amelogenin protein is the main element of the continually secreted enamel extracellular matrix (Iijima et al 2010). Other proteins involved in the formation of dental enamel include ameloblastin, tuftelin, amelotin and sialo-phosphoprotein dentin (Crawford et al., 2007b). Research has shown the existence of several other proteins in the extracellular matrix of enamel just like albumin, serine proteases, glycoconjugates and calcium dependent proteases (Fincham et al., 2000). The

importance of these proteins is not fully understood, but it is assumed that they promote the growth of enamel by serving as a foundation for minerals to build on.

In individuals, enamel differs in density over the tooth surface, mostly thickest at the cusp up to 2.5 mm and thinnest at the boundaries around 1.3 mm with cementum at the CEJ (Ten Cate and Nanci, 2013).

1.2.1. Amelogenesis

Amelogenesis is the process of enamel formation of teeth. This method happens simultaneously with the formation of dentine (dentinogenesis) but as a completely different process. While dentine must be present to form enamel, ameloblasts must be available as well to initiate the formation of dentine. The newly differentiated odontoblasts send a signal to the IEE, which further differentiates the epithelial cells into active secretory ameloblasts. In turn, dentinogenesis depends on triggers from the differentiating IEE for the continuation of the process Figure 1-3 (Moradian-Oldak, 2012).

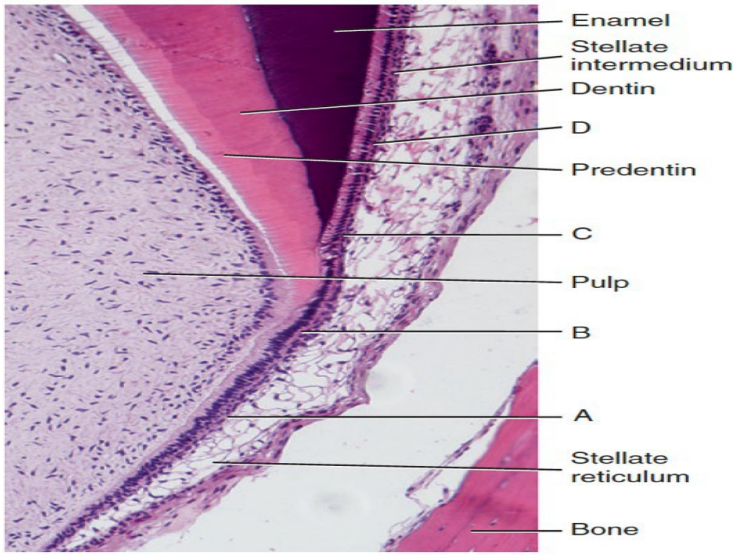


Figure 1-3 Amelogenesis features as seen by a light microscope. At A, the inner enamel epithelium (IEE) consists of short, columnar undifferentiated cells. At B, these cells elongate and differentiate into ameloblasts that face differentiating odontoblasts and then begin secreting the enamel matrix. At D, enamel matrix is constantly deposited by ameloblasts (Nanci, 2017).

The interaction in both the IEE and odontoblasts is an example of biological principal known as reciprocal induction among mesenchymal and epithelial cells. The enamel is initially synthesized as a soft mineralized organic matrix, which consist of only 30 percent mineral with the rest being organic material and water. This proportion is gradually inverted, and the mature tissue retains more minerals than the organic matter (Robinson et al., 2003, Moradian-Oldak, 2012). Enamel formation can be split into three separate developmental phases: first, an extracellular matrix is created, followed by the mineralization of this matrix and lastly the removal of the matrix to allow crystallite development or maturation of enamel (Ayers et al., 2004). The process of enamel formation involves a series of extremely controlled cellular operations and protein-controlled mineralization. The main developmental phases of enamel are pre-secretory, secretory, transition and maturation characterized by ameloblast morphology and function (Ruan and Moradian-Oldak, 2015, Nanci, 2008). Lacruz et al suggested a further seven-stage subdivision: pre-secretory, early secretory, late secretory, transition, pre-absorptive, early maturation, and late maturation stages as shown in Figure 1-4 (Lacruz et al., 2017).

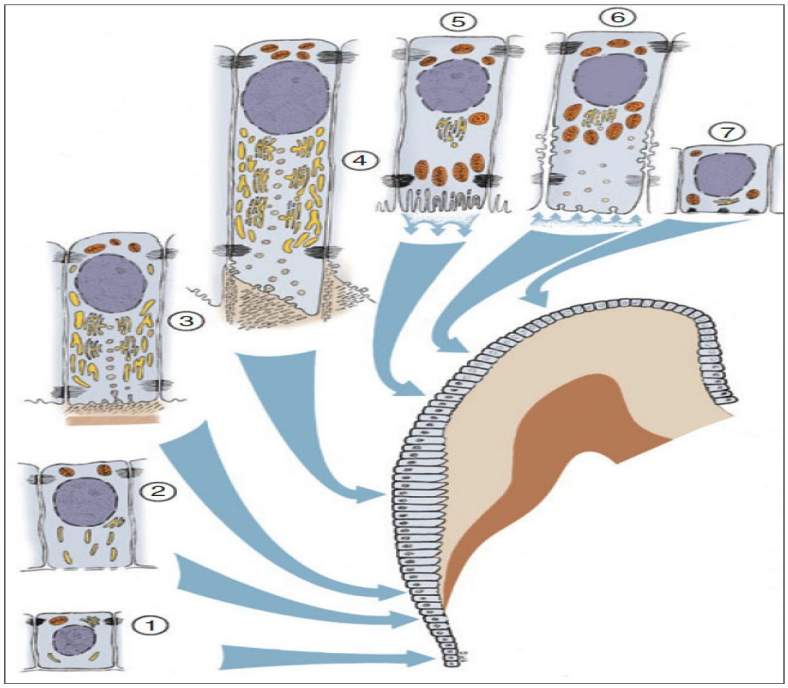


Figure 1-4 Schematic illustration in the development cycle of ameloblasts of the various functional stages as would occur in a human tooth.1, Morphogenetic stage; 2, histodifferentiation stage; 3, initial secretory stage (no Tomes' process); 4, secretory stage (Tomes' process); 5, ruffle-ended ameloblast of the maturation stage; 6, smooth-ended ameloblast of the maturation stage; 7, protective stage (Nanci, 2017).

1.2.1.1. Pre secretory stage

This phase is also recognized as the inductive phase where ameloblast morphodifferentiation occurs. The IEE differentiates during the bell phase of tooth formation to form the enamel forming cell identified as the ameloblast. The ameloblast cells became first cuboidal then differentiate into columnar cells which are basically the pre-ameloblasts. After the modification in the IEE, the dental papilla nearby the mesenchymal cells begin to distinguish into odontoblasts. Next, the basal lamina dividing the two cells which then collapses when the first dentine layer is laid down and generates a signal for the ameloblast to start secretion (Berkovitz et al., 2017).

1.2.1.2. Secretory stage

The method of amelogenesis starts first at the secretory phase, which includes the secretion of enamel matrix proteins such as amelogenin, ameloblastin and enamel crystals through ameloblasts. The secretory ameloblasts developed from the pre-ameloblasts by tall columnar cells and Tome's process. At this point, most of the enamel's density and quantity is finished (Lacruz et al., 2017). "Tome's process" forms at the apical secretory end of the ameloblast after the deposition of the first, thin and a prismatic tissue, which is a cone shaped or pyramidal shaped process. The Tomes processes form is essential for the development of the enamel prisms framework (Berkovitz et al., 2017).

The initial hydroxyapatite crystallites are thin needle like and smaller than the crystallites found in mature enamel. Enamel crystallites grow parallel to the ameloblast's distal surface, with each crystallite emerging as flattened hexagons when examined in cross section. A single prism is formed by four ameloblasts, and every ameloblast is part of the four prisms development (Berkovitz et al., 2017).

1.2.1.3. Transition stage

The first enamel matrix laid down during the secretory phase has elevated water and protein level, but low mineral content which is more permeable (Slavkin and Bavetta, 2017). In the transition stage, the ameloblasts decrease the secretion of their enamel proteins and generate a protease that deteriorates and promotes the removal of the organic matrix from the extracellular compartment. These modifications speed up the development of enamel crystallites in size and end their length development as part of the maturation phase which makes a significant contribution to enamel hardness (Hu et al., 2007, Lacruz et al., 2017). The transition of this immature enamel to the well mineralized enamel is considered maturation (Robinson et al., 1995).

1.2.1.4. Maturation stage

The transition and maturation start after the enamel layer's complete thickness is determined (Ruan and Moradian-Oldak, 2015). Enamel crystallites continue to develop and expand in width and thickness. The ameloblast exhibits ultrastructural alterations as the Tomes process collapses and the inner structure is completely reorganized and the length decreased byhalf (Ruan and Moradian-Oldak, 2015). These cells are striated or have a ruffled boundary under the microscope. These indications that the ameloblastshave altered their role from manufacturing to transportation, as in the secretoryphase. Ameloblast transfer calcium, phosphate, and carbonate ions into the matrix and extract water and debased enamel matrix protein. Generally, mineral content of enamel increases suddenly at the start of maturation owingto the absorption of mineral ions by quantity of up to 95 percent mineral (Robinson et al., 1995).

Mineralization of enamel does not occur evenly; in denser enamel, the outer layer is often more mineralized particularly in comparison to the inner layer. Once development is completed, ameloblast with remnant of the enamel organ composes the Reduced enamel epithelium (REE). Enamel mineralization occurs only once, when the tooth erupts into the oral cavity the ameloblasts are shed in the REE; therefore, the formation of enamel is finalized after amelogenesis. The ameloblast cells have limited repair capacity, so any disruption during the mineralization of enamel may lead to permanent discoloration or inadequate formation of the enamel layer as the mineralization mechanism of enamel is delicate and takes place over a long period of time (Pinkham et al., 2005).

1.3. Properties of Enamel

1.3.1. Physical properties

1.3.1.1. Colour

The ordinary enamel colour ranges from light yellow to greyish white that emits photons in varying angles because of the crystalline material of enamel. Immature enamel hue is white, even though light penetrates it easily, with almost all of it reflected without absorption. This tends to result in minimum subsurface scattering and white colour. Tooth colour is influenced by the incorporation of intrinsic shade and the existence of extrinsic stains which may build up on the surface of the tooth (Joiner and Luo, 2017). Dispersion and absorption of light within enamel and dentine result in the inherent colour of the teeth and since the enamel is comparatively transparent, the characteristics of dentine can play a significant part in ascertaining the general colour of the tooth (Joiner, 2004).

The colour does have a slightly blue tone at the edges of the teeth in which there is no dentine surrounding the enamel. Because enamel is semi translucent, the dentine colour and any material beneath enamel heavily influences a tooth's appearance. The white colour of enamel in the primary teeth as it has a much opaquer crystalline shape than permanent teeth. Enamel translucency rises with era and some of the underlying dentine's colour is then transferred which results in a yellow appearance (Berkovitz et al., 2009). Kim et al. used a Vita Easyshade spectrophotometer of 604 teeth of U.S. children aged 2 to 5 years and he concluded the most common shades are A1 (46%), A2 (25%) and B1 (11%) for the primary teeth (Kim et al., 2007).

The gradient of mineral density in hypomineralized enamel lesions with maximum density at the dentino-enamel junction (DEJ) and poorest density towards the surface, which is the reverse of normal enamel, explains why the colour is changed by mineralized enamel (Garot et al., 2017).

1.3.1.2. Hardness

Enamel is the body's hardest and most mineralized biological tissue. It can resist the forces of shear and shock when two or more bodies collide over a short period of time. In the harsh environment of the oral cavity, enamel also suffers from intense PH and temperature fluctuations within the human body, It can withstand 100s of masticatory cycles with biting forces of up to 770 N (Gordon et al., 2015, Beniash et al., 2019). It also has a high resistance to abrasion to restrict wear. These characteristics are very significant because enamel cannot be repaired or replaced (Berkovitz et al., 2009). Hardness of enamel has been one of the mechanical characteristics most commonly studied. Knoop hardness number (KHN) or `Vickers hardness number (VHN) recorded the values of enamel hardness based on the technique used (there were no significant variations in the value acquired from both techniques (Gutierrez-Salazar and Reyes-Gasga, 2001)).

Normal values of enamel hardness in permanent teeth ranges between 3.1 – 4.7 gigapascals (GPa) with an elasticity module between 62.06 – 95.77 GPa (Mahoney et al., 2004). One research used atomic force microscopy (AFM) and nano-indentation methods to check the composition of a single enamel prism / rod in third permanent molars. The average hardness was 3.9 and 3.3 GPa when analyzed perpendicular to the glass plates (Habelitz et al., 2001).

The outer surface of enamel to the enamel dentinal junction enamel hardness remains constant (Gutiérrez-Salazar and Reyes-Gasga, 2003). Research combining Vickers indentation and SEM showed the variations in the mechanical characteristics of enamel effectively (Xu et al., 1998). Fragrell et al. used the electronic micro hardness tester equipped with a Vickers diamond to assess the enamel hardness of normal and hypomineralized teeth on the surface of enamel. The study revealed that the average value of the enamel hardness in ordinary teeth was more than two times greater than the VHN 350.7 and 144.3 in hypomineralized enamel (Fagrell et al., 2010).

Reduced mineral content and/or increased organic content cause hypomineralized enamel to show reduction in hardness and elasticity, resulting from increased porosity and a continuously disorganized prism structure when compared to normal enamel (Mahoney et al., 2004, Farah et al., 2008).

Classic crystals in mature enamel are approximately 50 nm wide (26 nm x 63 nm as shown by Daculsi and Kere bel 18) and longer than 10 µm (Beniash et al., 2019). The mechanical characteristics of single enamel rods could not be measured by any of the above techniques (Habelitz et al., 2001).

1.3.1.3. Microstructure

Calcium hydroxyapatite is the main mineral element of enamel. It is made up of cross section crystallites that are hexagonal in shape. Hundreds of thousands of crystallites make up the enamel rod, the fundamental enamel structural unit. The crystallite size is approximately 70 ñm in width, 25 ñm dense, and extends to the entire tissue length. Enamel rods extend from the EDJ to the surface. The enamel rods appear as keyhole structures in cross section, with head pointing occlusally and the tail cervically. The crystallites run parallel to the prim's long axis in the rod head (Berkovitz et al., 2009).

1.3.2. Chemical properties

Enamel is the body's most mineralized tissue, creating a very strong, thin, transparent layer of calcified tissue covering the surface of the tooth's anatomical crown. It is known that carbonate ions in hydroxyapatite can substitute phosphate or hydroxyl ions. The proportion of magnesium and carbonate is comparatively large during the secretory phase of amelogenesis. It may have to do with less ordered immature crystallites, and with more foreign products. The incorporation of "carbonatoapatites" in the carbonate structure tends to generate less stable apatite (Kunin et al., 2015).

Fluoride is also included in fluorapatite forming hydroxyapatite crystallites. Fluorides can improve the composition of crystallite to be more orderly (Kunin et al., 2015). As secretion progresses, carbonate, magnesium, and fluoride

concentrations fall into the transitional phase. The crystallites are growing and there is clear evidence that the mineral content is increasing. The inorganic content of the enamel increases at the maturation phase relative to the matrix structure (Robinson et al., 1995). The remaining tissues are water and organic material. Water presence relates to tissue porosity that can appear between the crystal and surround the organic compounds.

1.3.3. Developmental defects of enamel

Visible deviation from the standard translucent appearance of tooth enamel due to enamel organ dysfunction can affect the primary and permanent dentition (Correa-Faria et al., 2013). Enamel defects have been correlated with a wide range of aetiologies including systemic, local, and environmental variables. Studies have shown that systemic conditions such as perinatal, prenatal, postnatal disease, low birth weight, periodic antibiotic use, coeliac disease, and respiratory illnesses are linked to enamel defects (Visweswar et al., 2012). Narang et al. noted high correlation between asthma and developmental enamel defect in permanent dentition (Narang et al., 2003).

Developmental defects of enamel (DDE) may be categorized into two main types: hypomineralization or hypoplasia of enamel. Hypomineralization (also called opacity) is a qualitative enamel defect that arises as a defect in enamel's translucency which is clinically presented as enamel opacity, either demarcated or diffuse. hypoplasia is quantitative resulting in deficient enamel density in both primary and permanent dentitions (Jälevik et al., 2001). Epidemiological studies indicate that the prevalence of these enamel defect appears to be increasing in practically all demographics, highlighting their clinical significance and value for public health projects. Studies have found that DDE incidence in developed countries and ranges from 24-49% in primary and 9-63% in the permanent dentitions (Seow et al., 2011)

Different indices have been suggested over the previous years to measure enamel defects including fluorosis. These indices can be split into two primary classifications: specifically, fluorosis indices and descriptive indices that include

all forms of defects (Clarkson and O'mullane, 1989). Dean's Index, Tooth Surface fluorosis Index and Thylstrup and Fejeskov Fluorosis Index are the most popular indices to define enamel fluorosis. However, because of the difficulty in distinguishing between fluoride and non-fluoride enamel defects this led to the design of a second set of indices that include all kinds of enamel defects. These indicators, however, were descriptive in origin, which caused further confusion. A world Dental Federation Commission Working Group on Oral Health Research and Epidemiology was created to overcome these disadvantages. A descriptive index entitled "The Developmental Defect of Enamel" (DDE) index was suggested by the group. The modified index defines the enamel defect in terms of type, number, demarcation, and location of the defects Table 1-1 (Clarkson and O'mullane, 1989).

Categories	**Code**
Normal	0
Demarcated opacities:	
white/cream	1
yellow/brown	2
Diffuse opacities	
" — Lines	3
"— Patchy	4
"— Confluent	5
Confluent/patchy + staining + loss of enamel	6
Hypoplasia	
Pits	7
Missing enamel	8
Any other defects	9
Extent of Defect	
Normal	0
< 1/3	1
at least 1/3 >2/3	2
at least 2/3	3

Table 1-1 modified DDE index (Clarkson, 1989)

1.4. Amelogenesis Imperfecta

Amelogenesis Imperfecta (AI) is a group of conditions, genomic in origin, that affect the enamel structure and clinical appearance of all or almost all the teeth and can be associated with morphological and biochemical changes elsewhere in the body (Crawford et al., 2007b). Almost 85 inherited conditions may affect the formation of enamel, but AI is the most prevalent condition affecting the quantity and quality of enamel in the absence of other developmental characteristics (Wright et al., 2011).

1.4.1. Epidemiology of AI

Prevalence rates vary widely from 1:14,000 in the US to 1:700 in Sweden depending on the population (Smith et al., 2017). A Swedish study found that 63% of patients were autosomal dominant, while another Middle East study found that autosomal recessive AI was the most common type (Ranganath et al., 2010).

1.4.2. Aetiology of AI

AI is an enamel developmental defect that may be autosomal, autosomal recessive, x-lined or irregular during inheritance (Crawford et al., 2007b). Mutation in the following genes was associated with AI, amelogenin (AMELX), kallikrein (KLK4), sequence like family 83, member H (FAM83H), enamelysin (MMP20), WD 72 (WDR72), and three other possible genes (ANABN), tuftelin (TUFT1), enamelin (ENAM) and homeobox protein DLX-3 (DLX3). Although such genes are known to code for proteins released in the enamel matrix, around 25% of AI patients have only found a condition that causes mutation. The aetiology of AI includes more genes that need to be identified and studied (Hu and Simmer, 2007). While there is an understandingof the genes involved in AI, the way the phenotypic manifests remain limited. At least 18 genes have been identified as having mutations that can trigger AI, and of 192 distinct variants of AI genes have been published in the literature (Smith et al., 2017).

The expression of Ameloblastin (AMBN: MIM *601259) for every inherited condition described in Mendelian Inheritance in Man (MIM); is assigned a 6-digit number, clinical geneticist across the globe have given these numbersas numerical taxonomic system for hereditary disease (Amberger et al., 2015).This gene was first observed in the enamel organ and, after amelogenin, is the second most abundant matrix protein of enamel known to play a key role in enamel formation (Smith, 1998). By mediating cell growth, migration, differentiation, apoptosis, and gene expression, the extracellular matrix plays an important role in tissue growth and homoeostasis (Damsky and Werb, 1992). AMBN is believed to be involved in the enamel extracellular matrix (ameloblast attachment) to the underlying enamel matrix and in enamel crystal growth modulation and it is already known that mutations in this gene cause AI (Rajpar et al., 2001).

Disturbances in the development of enamel can occur at different stages, resulting in a different clinical presentation from one individual to another. Hypoplastic enamel is caused by faults in the amelogenesis secretory stage where there is a lack of appositional development. Hypomature enamel appears when there is no complete removal of the organic matrix that separates enamel crystallites and typically breaks down during enamel formation. There are more prominent errors in the mineralization of the enamel in the hypocalcified form of enamel, resulting in a much softer surface (Hu et al., 2007).

1.4.3. Classification of AI

AI was classified into four groups with at least 15 subtypes based on the normal amelogenesis process and its clinical manifestations (Patel et al., 2013). The history of classification of AI since 1945 to 2003 is shown in Table1-2. Some were solely based on phenotypes, while others used phenotype asa primary and mode of inheritance as a secondary discriminant. The variety of classification based only on phenotype could be perplexing, and it is not usually feasible to cross reference among the different subtypes employed (Crawford et al., 2007a). The three major types of AI are associated with

defects in the process of enamel synthesis as illustrated in Figure 1-5. When there is a defect in the enamel matrix in the hypoplastic form, the enamel does not have the usual thickness; it is thin, or cavities are found in localized or generalized areas due to apposition defects. The enamel's radio-opacity is considerably higher than the dentin (Anitha et al., 2018, Roma and Hegde, 2016).

Enamel is of normal thickness in the hypocalcified form, but there is defective matrix mineralization, with irregular enucleation and mineralization of prism or rod crystals, resulting in soft enamel that is easily removed with an instrument. In these situations, the enamel has lower transparency than the dentin (Anitha et al., 2018, Roma and Hegde, 2016).

While the enamel is of normal thickness in the hypomature form, it's the hardness and opacity that are unusual. During the maturation phase, there is a deficiency in the growth of crystals because proteins are not entirely removed. In these conditions, the enamel radio opacity is about the same as the dentin (Urzúa et al., 2011, Anitha et al., 2018, Roma and Hegde, 2016).

Figure 1-5 illustration of three basic types of Amelogenesis Imperfecta

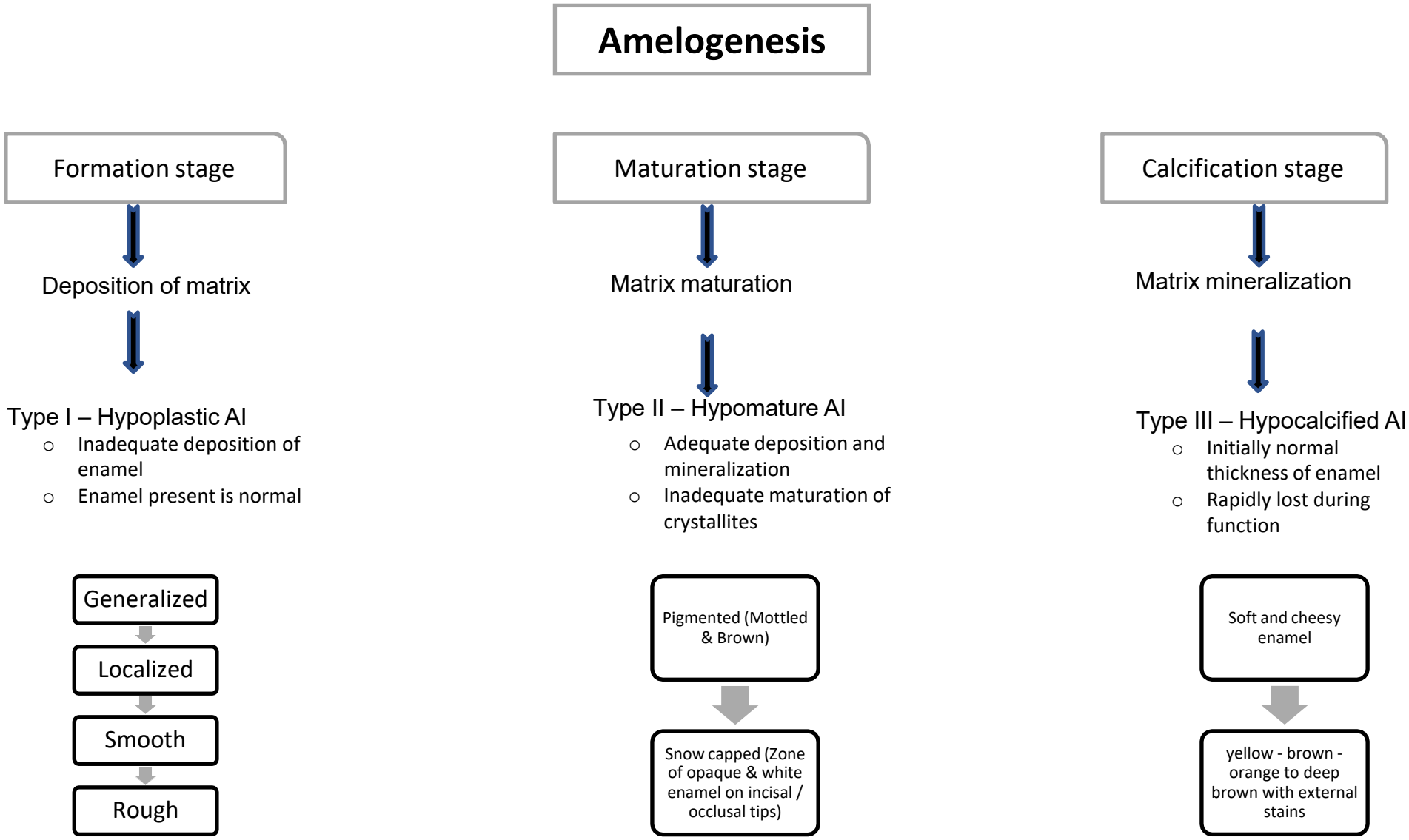


Table 1-2 Classification systems applied to amelogenesis Imperfecta (AIdred et al., 2003, Crawford et al., 2007a)

Weinman *et al.*, 1945	*Two types based solely on phenotype: hypoplastic and hypocalcified*
Darling, 1956	*Five phenotypes based on clinical, micro-radiographic and histopathological findings*.Hypoplastic Group 1 – generalized pitting Group2 – vertical grooves (now known to be X-linked AI) Group 3 – Generalized hypoplasia Hypocalcified Type 4A – chalky, yellow, brown enamel Type 4B – marked enamel discoloration and softness with post-eruptive loss of enamel Type 5 – generalized or localized discoloration and chipping of enamel
Witkop, 1957	*Classification based primarily on phenotype. 5 types:* 1. Hypoplastic 2. Hypocalcification 3. Hypomaturation 4. Pigmented hypomaturation 5. Local hypoplasia Added mode of inheritance as further means of delineating cases.
Schulze, 1970	*Classification based on phenotype and mode of inheritance*.
Witkop and Rao, 1971	*Classification based on phenotype and mode of inheritance. Three broad categories: hypoplastic, hypocalcified, hypomaturation*. **a.** Hypoplastic Autosomal dominant hypoplastic-hypomaturation with taurodontism (subdivided into a and b according to author) Autosomal dominant smooth hypoplastic with eruption defect and resorption of teeth Autosomal dominant rough hypoplastic Autosomal dominant pitted hypoplastic Autosomal dominant local hypoplastic X-linked dominant rough hypoplastic **b.** Hypocalcified

	Autosomal dominant hypocalcified **c.** Hypomaturation X-linked recessive hypomaturation Autosomal recessive pigmented hypomaturation Autosomal dominant snow-capped teeth White hypomature spots

Winter and Brook, 1975	*Classification based primarily on phenotype. Four main categories: hypoplasia, hypocalcification, hypomaturation, hypomaturation-hypoplasia with taurodontism, with mode of inheritance as a secondary means of sub-classification.* **a.** Hypoplasia Type I. Autosomal dominant thin and smooth hypoplasia with eruption defect and resorption of teeth Type II. Autosomal dominant thin and rough hypoplasia Type III. Autosomal dominant randomly pitted hypoplasia Type IV. Autosomal dominant localized hypoplasia Type V. X-linked dominant rough hypoplasia **b.** Hypocalcification Autosomal dominant hypocalcification **c.** Hypomaturation Type I. X-linked recessive hypomaturation Type II. Autosomal recessive pigmented hypomaturation Type III. Snow-capped teeth **d.** Hypomaturation-hypoplasia with taurodontism Type I. Autosomal dominant smooth hypomaturation with occasional hypoplastic pits and taurodontism Type II. Autosomal dominant smooth hypomaturation with thin hypoplasia and taurodontism
Witkop and Sauk, 1976	*Classification based on phenotype and mode of inheritance, similar to classification of Witkop and Rao (1971)*
Sundell and Koch, 1985	*Classification based solely on phenotype* 1. Hypoplastic 1.1 Rough 1.1.1 Basic form 1.12 Thin enamel 1.1.3 Pitted basic form 1.1.3.1 pitted thin 1.1.3.2 pitted with horizontal grooves 1.1.3.3 pitted with vertical grooves 1.1.5 Unspecified appearance

	1.2 Smooth 1.2.1 Thin enamel 2. Hypomineralized 2.1 Hypomaturated 2.1.1 localized opacities 2.1.2 Generalized opacities 2.2 Hypocalcified 2.2.1 Localized or generalized
Witkop, 1988	*Four major categories based primarily on phenotype (hypoplastic, hypomaturation, hypocalcified, hypomaturation-hypoplastic with taurodontism) subdivided into 15 subtypes by phenotype and secondarily by mode of inheritance*. Type I. Hypoplastic Type IA. Hypoplastic pitted autosomal dominant Type IB. Hypoplastic, local autosomal dominant Type IC. Hypoplastic, local autosomal recessive Type ID. Hypoplastic, smooth autosomal dominant Type IE. Hypoplastic, smooth X-linked dominant Type IF. Hypoplastic, rough autosomal dominant Type IG. Enamel agenesis, autosomal recessive Type II. Hypomaturation Type IIA. Hypomaturation, pigmented autosomal recessive Type IIB. Hypomaturation, X-linked recessive Type IIC. Hypomaturation, snow-capped teeth, X-linked Type IID. Hypomaturation, snow-capped teeth, autosomal dominant? Type III. Hypocalcified Type IIIA. Autosomal dominant Type IIIB. Autosomal recessive Type IV. Hypomaturation-hypoplastic with taurodontism Type IVA. Hypomaturation-hypoplastic with taurodontism, autosomal dominant Type IVB. Hypoplastic-hypomaturation with taurodontism, autosomal dominant
Aldred and Crawford, 1995	*Classification based on:* Molecular defect (when known) Biochemical result (when known) Mode of inheritance Phenotype

Hart *et al*., 2002	*Proposed a molecular defect sub classification of the AMELX conditions* 1.1 Genomic DNA sequence 1.2 cDNA sequence 1.3 Amino acid sequence 1.4 Nucleotide and amino-acid sequences 1.5 *AMELX* mutations described to date
Aldred *et al*., 2003	*Classification based on:* Mode of inheritance Phenotype – Clinical and Radiographic Molecular defect (when known) Biochemical result (when known)

Table 1-3 Classification systems applied to amelogenesis Imperfecta continued (Aldred et al., 2003, Crawford et al., 2007a)

Phenotypic classification has been shown to inaccurate and recent genetic studies have shown that many people within a particular family have common genetic mutation, categorized according to their phenotype (Aldred et al., 2003).

In 2003, Aldred proposed a new classification according to inheritance mode as a primary mode and phenotype as the secondary mode. It is supposed to lead to a better understanding of how genotype contributes to the actual "functional genomic "phenotype (Aldred et al., 2003).

1.4.4. Types of AI

During amelogenesis phase, ameloblasts depends on various genes found also in other tissues, such as Laminin- 332 and type XVII collagen. Those genes generally cause syndromes that involve enamel defects. Non syndromic AI is caused by genes that are functionally specific for enamel tooth formation such as AMELX, ENAM, FAM83H, WDR72, KLK4 and MMP20 which make up roughly 50% of all AI cases (Chan et al., 2011). The following are description of mode of inheritance as primary and basic phenotypes as secondary discriminant:

1.4.4.1. Autosomal Dominant AI (ADAI)

In every family generation, ADAI usually affects one or more individuals (Wright et al., 2011). The most reported form of AI is autosomal dominant AI. The most direct gene is ENAM (4q13), the highest and least abundant protein in the enamel matrix, which is 3-5%, also, mutation in FAM83H and the remaining are of unknown aetiology (Urzúa et al., 2011). Autosomal dominant hypocalcified AI (OMIM: #130900) is caused by FAM83H mutations and has a unique enamel phenotype. The enamel is poorly mineralized and cheesy brown in colour at the time of eruption. On the other hand, mutations in ENAM gene are dose dependent which result in local hypoplastic AI (OMIM: # 104500) showing partially thin enamel or complete absence of enamel when both alleles are defected as shown in Figure 1-6 (Simmer et al., 2013). Even though there is only one ENAM allele, the hypoplastic enamel sometimes

shows a horizontal groove as a result of stopping the synthesis of protein from the allele (Wright, 2006).

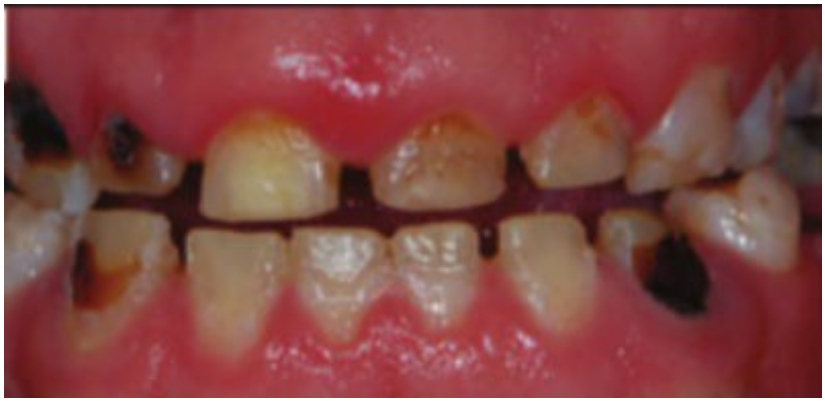


Figure 1-6 Autosomal dominant Hypoplastic AI characteristics of ENAM mutation in 6 years old boy (Simmer et al., 2013).

In the ENAM gene, nine mutations were identified. Evidence of one DLX3 mutation also correlated with taurodontism, and 6 mutations are identified with the FAM83H gene (Kim et al., 2008). They belong to different ethnic backgrounds and such mutations are found in the gene exon5 (Urzúa et al., 2011).

hypocalcified AI that may acquire autosomal dominant and/or autosomal recessive. In Caucasian and Negroid races, hypocalcified AI can be seen. North America is the most prevalent hypocalcified AI nation (Wright et al., 2011).

1.4.4.2. Autosomal recessive AI (ARAI)

Autosomal recessive AI (ARAI) is considered if it was known consanguineous marriage in family with patient affected with AI. Autosomal recessive AI is caused by a mutation in several genes such as enamelysin (MMP20, 11q22.3), Kallikrein 4 (KLK4, 19q13.41), ENAM, and WRD72. Clinically known to be hypomature pigmented AI phenotypes (Urzúa et al., 2011, Volodarsky etal., 2015).

1.4.4.3. X-Linked AI

X-linked AI occurs in five percent of all AI cases due to the X-chromosome mutation of the amelogenin gene found at the Xp22.1-p22.3 chromosome corresponding to the amelogenin locus (AMELX) Figure 1-6. The X chromosome in the Xq24-q27.1 zone has genetic evidence for a second AI locus (Urzua et al, 2011). The Y chromosome (AMELY) contains a second amelogenin gene, but this gene is typically expressed at a low level and does not seem to stimulate AI. There is also a family case report showing a substantial relationship to another X linked related -Xq22- 28 (Wright, 2015).

In early AI studies, the clinical variability in X-linked AI was noted. The heterogeneous AI phenotypes were associated with the position of the amelogenin protein mutations (i.e., signal peptide region, N-terminal region, or C-terminal region) and the function and expression effects of mutations (Duan et al., 2019). In the development of enamel, AMELX accounts for about 80-90 percent of the protein and is the main extracellular protein matrix. It serves as an organic scaffold important for regulating enamel crystallite order and directional growth. Proteins of amelogenin are normally extracted to encourage normal enamel crystallites to develop and provide enamel that exceeds 95% in mineral content (Wright, 2015).

The correlation of enamel defects with AMELX mutations in humans may clearly demonstrate the essential role of AMELX. Such mutations may lead to two separate phenotypes that most often overlap: hypoplasia due to enamel quantity deficiency or mineralization (hypomaturation) deficiencies. The broad range of AMELX related enamel phenotypes are caused by large deletions, signal peptide mutation or changes in different biological domains. In other words, the mutations of AMELX are complex, including mutations of deletion, violence, and nonsense. The presentations and extent of males and females affected dentition differ as affected males express the mutant X allele and females display a pattern of mosaic due to inactivation of the X chromosome. Affected males form a hypomature, yellowish, rough enamel from normal to very thin layers of enamel of different thicknesses. Because of X chromosome

inactivation, females with AMELX defects will have wither discolorations or vertical bands of hypoplasia, and lyonization is established (Wright, 2006, Berkman and Singer, 1971, Witkop Jr, 1967). The concept of lyonization states that one of the two X chromosomes is spontaneously inactivated early in development in female somatic cells. Heterozygous females will have mosaic patterns of different cell ratios in which only one of a common pair of alleles is involved. This will result in a phenotypic variation in x linked syndrome clinical expression (Patel et al., 1991, Witkop Jr, 1967, Berkman and Singer, 1971).

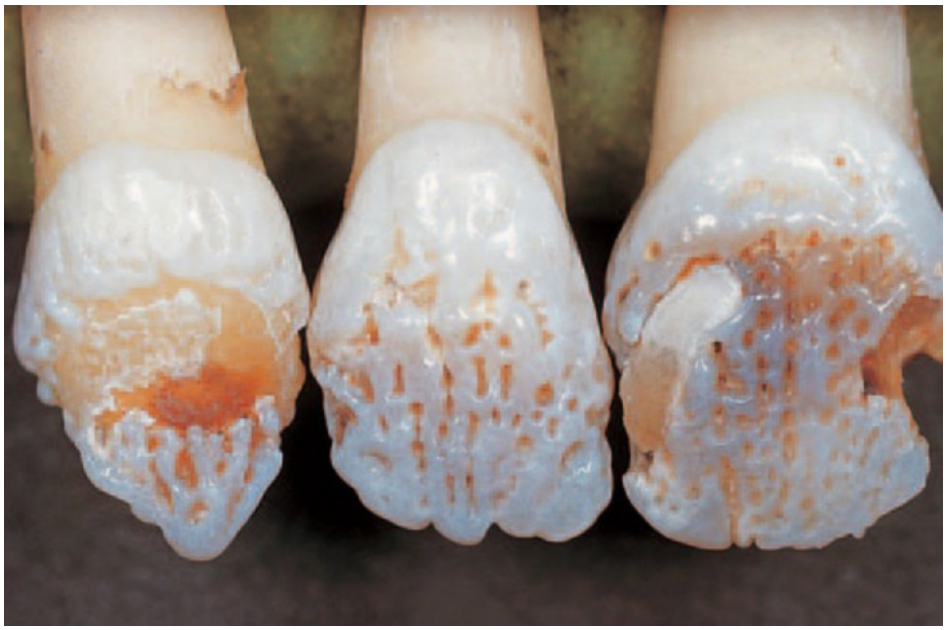


Figure 1-7 Amelogenesis Imperfecta, X-linked dominant hypoplastic type (Odell, 2017)

The clinical presentation and extent of AI can vary considerably between patients, and it is often difficult to diagnose the phenotype from clinical examination alone (Patel et al., 2013).

1.4.4.4. Hypoplastic AI

Hypoplastic AI occurs when there is inadequate enamel matrix and thus the thickness of the enamel surface is decreased (Cogulu et al., 2009, Adorno-Farias et al., 2019). The enamel that is produced is well mineralized, as some

areas of the enamel organ lack IEE cells, resulting in a deficiency of differentiation into ameloblast (Pinkham et al., 2005, Adorno-Farias et al., 2019). Many forms of hypoplastic enamel involving, smooth, pitted, and rough (Figure). The enamel has a thin, hard, and rough surface in the rough hypoplastic form Figure 1-7. Teeth tend to be smaller in size without proximal contact and very thin or non-existence enamel areas resulting in sensitivity. In 60% of cases, anterior open bite was also recorded (Rowley et al., 1982).

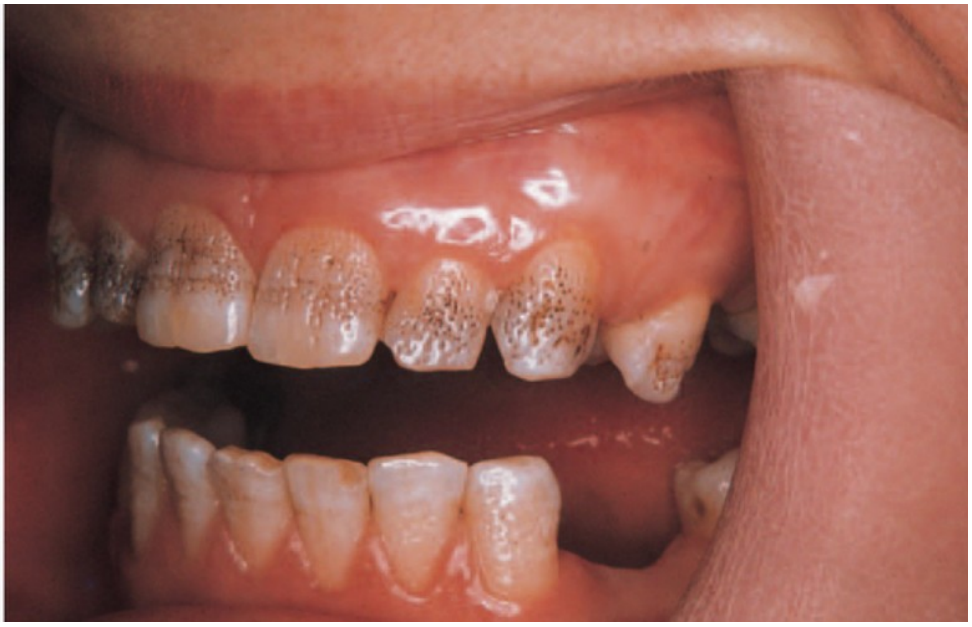


Figure 1-8 Amelogenesis Imperfecta, hypoplastic pitted type (Odell, 2017)

1.4.4.5. Hypocalcified AI

Hypocalcified AI is attributed to a defect in the calcification stage of the formation of enamel, marked by regular enamel thickness but poor calcification of the matrix Figure 1-8 (Adorno-Farias et al., 2019). Clinically, in the incisal region the enamel is delicate and fragile and may be lost shortly after eruption, to leave the underlying dentine exposed, resulting in an unsightly appearance. The enamel has a soft texture and easily can be scraped away. Large quantities of supragingival calculus are accumulated on the teeth, often correlated with severe gingivitis or periodontitis (Winter and Brook, 1975, Roma and Hegde, 2016).

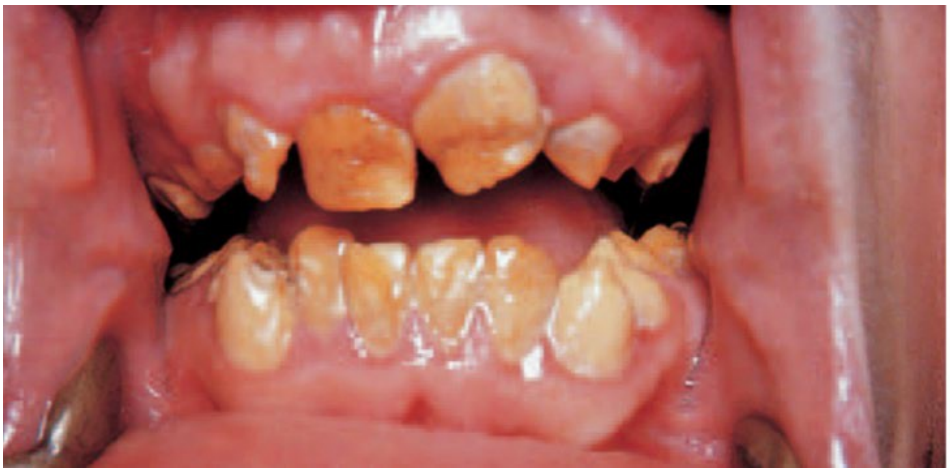


Figure 1-9 Amelogenesis Imperfecta, hypocalcified type (Odell, 2017)

1.4.4.6. Hypomaturation AI

Hypomature AI is identified by a normal thickness enamel but a reduced radiodensity and mineral content because of a mineralization deficiency in the amelogenesis maturation stage Figure 1-9 (Pinkham et al., 2005, Adorno-Farias et al., 2019). Clinically, the shape and size of the crowns is different, with a rougher, duller and less refractive surface than normal enamel, and they are more fragile because they tend to break or chip but do not seem more susceptible to caries (Kim et al., 2005, Roma and Hegde, 2016). KLK4 mutations with affected teeth seem to have a homogeneous dark yellow colour, whereas teeth with MMP20 defects have uneven greyish brown discoloration and are shinier (Kim et al., 2005).

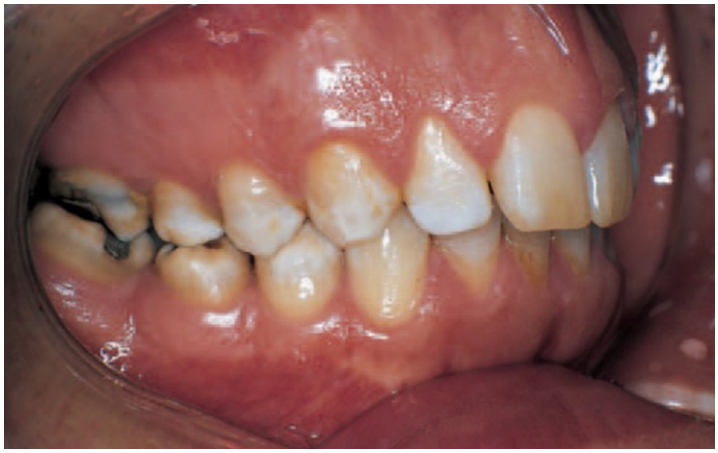


Figure 1-10 Amelogenesis Imperfecta, hypomaturation type (Odell, 2017)

1.4.4.7. Hypoplastic / Hypomature AI with taurodontism

The enamel with a yellow brown discoloration along with the pitting of the surfaces which appears mottled (Pinkham et al., 2005, Roma and Hegde, 2016). Taurodontism can be described as variations in the shape of the tooth caused by Hertwig's epithelial sheath diaphragm failure to invaginate at the correct horizontal point (Benazzi et al., 2015). The characteristic features are an expanded pulp space, apical displacement of the pulpal surface, and no restriction at the cement-enamel junction point. While permanent molars teeth are most frequently affected, this alteration is often seen, unilaterally or bilaterally, in both permanent or deciduous dentition and in any combination of teeth or quadrants (Dineshshankar et al., 2014)

1.4.5. AI in primary dentition

The clinical description of primary teeth may be equivalent to permanent dentition, although these teeth appear to be less affected. A study was conducted to investigate an AI family member of five generations. They noted that the primary exfoliated teeth also had a lack of enamel density, rough texture, and decreased consistency with enamel dentine and post eruptive breakdown (PEB) Figure 1-10 (Gjørup et al., 2009).

Former research on the primary structure and composition of enamel affected by local hypoplastic autosomal dominant AI caused by an ENAM mutation, showed the presence of a 'glass- like' enamel rod with reduced antibody to ENAM protein (Shore et al., 2010).

Another report revealed that a 4-year-old child had hypersensitivity to the teeth and was confirmed with AI. To eliminate the sensitivity and restore normal occlusion, full mouth rehabilitation of the primary molars with stainlesssteel crown and resin filled celluloid for the incisors resulted in better eating habits (de Souza-e-Silva et al., 2010).

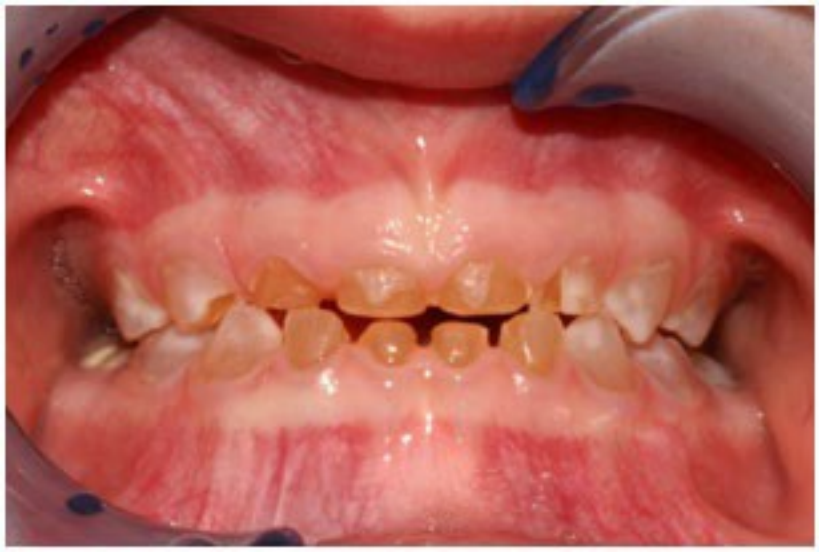


Figure 1-11 The early stage of hypoplastic AI in primary dentition of 3-year-old patient (Balian et al., 2017)

1.4.6. Syndromes associated with AI

There are multiple AI related syndromes because of changes in other parts of the body for example Trico – dento – osseous syndrome, Koklschutter (TONZ) syndrome, Nephrocalcinosis, Epidermolysis Bullosa (EB) and Cone red dystrophy (Jalili syndrome).

In 1988, Jalili and Smith first documented the combination of recessively inherited cone rod dystrophy (CDR) and amelogenesis imperfecta (AI) (Parry et al., 2009, Jalili and Smith, 1988). (OMIM 120970) usually occurs predominant or equivalent loss of cone in childhood or early adulthood relative to rod photoreceptors, poor visual acuity, defects in colour vision, photophobia, and loss of visual field (Michaelides et al., 2006). CDR related mutation can be inherited as an autosomal dominant, autosomal recessive, or X-linked mutation. It is characterized clinically by irregular enamel for both primary and permanent dentitions, taurodont permanent molar teeth indicating deformity in dentine, vision impairment in infancy or early childhood, and vision loss in advanced age Figure 1-11. Nystagmus is the first medical symptom of an abnormal vision in the first few months of life (Parry et al., 2009).

The affected enamel is mineralized by only 50 percent in comparison to ordinary enamel and the enamel prisms have been concealed by an amorphous organic material close to the teeth affected with MMP20 and KLK4 mutations in hypomaturation AI (Parry et al., 2009).

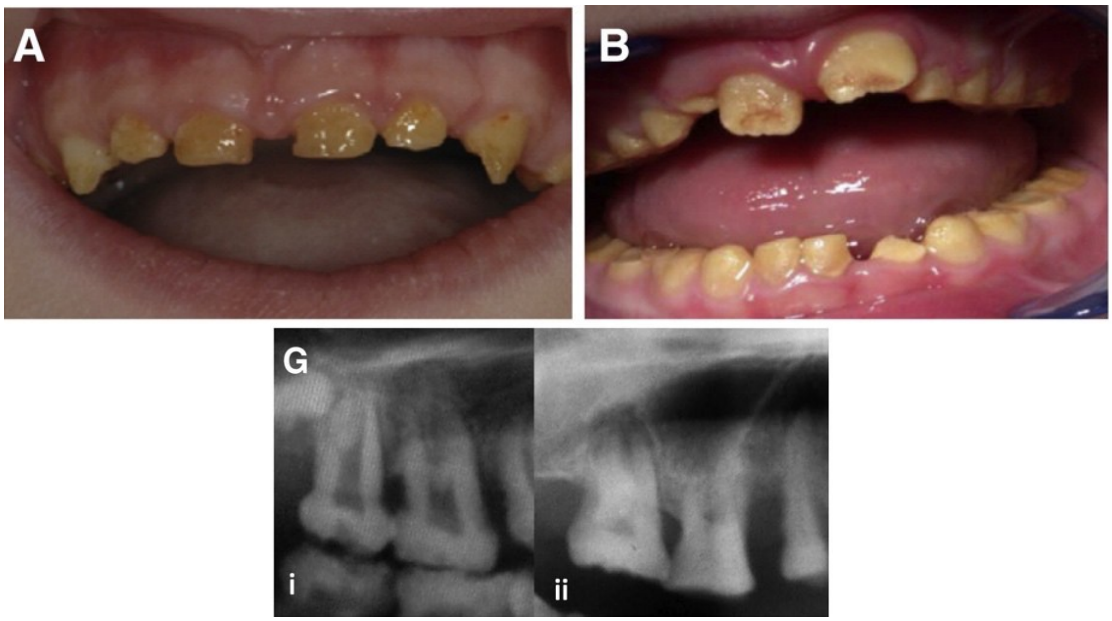


Figure 1-12 Clinical features of Jalili syndrome (Parry et al., 2009). At A & B, primary and permanent dentition respectively. At G, Dental radiograph, there is no predicted distinct contrast between the comparatively more radio dense enamel dentine, which restricts visibility into the ratios of enamel to dentine.

1.4.7. Diagnosis of AI

The AI diagnostic process includes the exclusion of other triggers, including such environmental factors and chronological disruptions. The determination of a probable pattern of inheritance along with the description of a phenotype that excludes chronological disturbances should be carries out. Radiographs could also be valuable for detecting enamel hypoplasia and hypomineralization (Crawford et al., 2007b).

It may be difficult to identify the enamel defect in AI teeth using the DDE index, as AI defects have various types; hypoplasia, hypocalcification and

hypomaturation. It has been shown that the Extended Enamel Defect Index (EDI) and Standardized image acquisition provide additional information regarding the phenotypic classification and enamel defect quantification in AI (Smith et al., 2009). The clinical diagnosis of AI can be assisted by asking the patient four questions as described by Crawford et al. as shown in Table 1-2. Questions to aid diagnosis of AI (Crawford et al., 2007a). Doing so, will help identify AI from other enamel defects, including fluorosis, which is known to be the most common differential diagnosis and can be difficult to differentiate from AI clinically (Patel et al., 2013).

1. Has anyone else in the family had anything like this?
2. Has there been anything in the patient's medical history which might have caused sufficient metabolic disturbance to affect enamel formation?
3. Are all the teeth affected in a similar manner?
4. Is there a chronological distribution to the appearance to the defect?

Table 1-4 Questions to aid diagnosis of AI (Crawford et al., 2007b)

1.4.7.1. Radiographic assessment of AI

Radiographic investigation of AI teeth can offer essential information about the enamel mineralization degree. Hypoplastic AI is characterized by a square crown, a thin radiopaque of enamel layer, thin or missing cusp and open contacts. In Hypomaturation type the enamel is of normal thickness but the density is similar to dentine depending on the severity of the case. The enamel density of hypocalcified AI is the lowest among all AI types (Wright et al., 1995). Also, other dental anomalies within AI include delay in eruption, crown resorption, taurodontism and pulp canal calcification were all have been observed frequently in hypoplastic AI (Collins et al., 1999).

1.4.7.2. Genetic Testing of AI

Knowing the pattern of inheritance for AI cases, as well as providing genetic guidance and counselling specific to each patient, is a crucial element of care and diagnosis (Aldred and Crawford, 1995). There has been a considerable gain in understanding of the genetics of AI, with the finding of mutations in multiple genes that were not initially identified in AI patients. Similar genes have been shown to be implicated in many types of AI, allowing more understanding of the link between phenotype and genotype connection (Hart and Hart, 2009).

The foundation of genetic started in 1990 with the project “The international Human Genome” and it was completed in 2003 which continues to be a remarkable scientific achievement. The purpose of this project was to compile the entire mapping of all human genes, which were estimated roughly to 20,500 genes (Health, 2017). The advancement of genetic knowledge and technology has progressed to the point that it now applied in both medical and dental fields.

The Online Mendelian Inheritance in Man database established in 1960 has been updated regularly, it contains information for over than 15 thousand genes as well as already identified Mendelian disorders. Clinicians and researchers are learning more about the aetiology and clinical implication of these genetic disorders. It can be applied on developing a prevention and therapeutic intervention strategies that target the aetiology of underlying disorder (Hart and Hart, 2009).

The progress of genetics in healthcare and medicine has also resulted in advancements in the dental field. The literature recognizes the difficulties associated with genetic testing in dentistry (Eng et al., 2012). Dentists must be trained, competent and confident in order to determine if a genetic testing is required. They should have the capability to ensure that the results are correctly evaluated and handled; genetic guidance is an important part of this process (Eng et al., 2012). In terms of legal issues, patient’s confidentiality and the possibility of discrimination are highly important points for dental team

when considering genetic analysis. Obtaining vast volume of new genetic information for patients and their effect may have on other family members that may or may not be related to their dental care must be handled with caution and with strong ethical principles and legal standards (Gettig and Hart, 2003). In dental field, genetic testing or counselling is not usually performed on a regular basis for dental conditions. In the literatures, chorionic villous sampling might be used to detect if a baby has a genetic or chromosomal condition like x-linked hypohidrotic ectodermal dysplasia, some AI forms and hypodontia (Wright et al., 2003, Aldred et al., 2003). DNA found in saliva can be studied to identify a variety of disorders including AI. If AI genetic testing and other disorders is to be adopted, it is critical to understand what problems dentists may have in implementing it effectively to improve clinical treatment and how to avoid them.

In the United Kingdom, with higher fees due to technological and clinical complexity, AI genetic testing could be done in three ways: Whole genome sequencing (WGS), Whole exome sequencing (WES) and targeted gene panel. The National Health Service (NHS) now offers genetic testing for AI patients (McDowall et al., 2018). The NHS has introduced a targeted 21 gene panel with mutations known to cause AI. This technique is likely to provide a negative outcome as not all genetic variants of AI have been discovered (Smith et al., 2017). It has been recognized that genetic information has the potential to improve AI treatment. Which would allow for a far more precise classification of AI and the application of this knowledge to inform patient comprehension and the geno-phenotype correlation (Aldred et al., 2003).

1.4.8. Differential diagnosis of AI

Chronological tooth formation disorders, localized tooth formation disorders and extrinsic tooth formation disorders should all be explored in the differential diagnosis.

Dental fluorosis is the most prevalent differential diagnosis. Fluorosis can range from mild white flecking of enamel to diffuse white discoloration with

irregular, disfiguring areas of staining and hypoplasia. Clinically, fluorosis can show areas of horizontal white banding correlating to the times of higher fluoride consumption, with the effect of premolars and 2nd permanent molars could be spared (chronological distribution). A history will often reveal excessive fluoride intake in some of the patient, whether because of a behavior like eating toothpaste in childhood or as a result from consumptionof a local water source (Chanmougananda et al., 2012).

Chronological enamel hypoplasia is a similar pattern of findings that can occur for a variety of reasons throughout tooth development. These reasons can range from long term gastrointestinal distress like celiac disease, vitamin D deficiency rickets, to anti leukemic treatment and can be diagnosed based on the history and chronological distribution of bands a cross the crowns of the teeth (Chanmougananda et al., 2012, Fulton et al., 2020).

1.4.8.1. Molar Incisor Hypomineralization (MIH)

The term MIH to characterize the clinical manifestations of hypomineralized enamel due to systemic causes. The terms used to describe this condition like hypomineralized permanent first molars (FPM), idiopathic hypomineralization, dysmineralized FPM and cheese molars. MIH caused by a disturbance in amelogenesis during the transition and maturation stages (William et al., 2006).

The definition of MIH according to (Weerheijm et al., 2001) is “a qualitative defect of systematic origin of the enamel, involving one or more first permanent molar, which is frequently associated with affected incisors”.

The prevalence varies depending on the country in which the survey was done. MIH was found to be prevalent in children from 2.8% in Hong Kong (CHO et al., 2008), 22% in Spain (Garcia-Margarit et al., 2014) to 40% in Denmark and Brazil (Wogelius et al., 2008, Soviero et al., 2009).

The association between deciduous molar hypomineralization (DMH), which generally refers to the primary 2nd molar and MIH was discovered in a study, suggesting that DMH might be used as an indicator for MIH. According to the study, DMH is prevalent among young children in the Netherlands at a rate of 9% (Elfrink et al., 2012).

The cause of MIH is still unclear. However, there were several factors thought to cause MIH including environmental and genetics factors, as well as prenatal, perinatal, and postnatal factors (Butera et al., 2021). The first four years of life are crucial for the development of MIH, rather than only the first year (Sönmez et al., 2013).

Hypomineralised enamel is characterized by an alteration in the translucency of the enamel (opacity), and the lesion in MIH is generally defined, as opposed to the diffuse opacities that are common in fluorosis (Weerheijm et al., 2001). Clinically, MIH affected enamel can range in colour from white to yellow/ brownish opacities. Even though MIH can affect several teeth, it is neither a chronological condition like tetracycline staining nor linear enamel hypoplasia. Equally, it does not affect the whole dentition as seen in amelogenesis imperfecta (Da COSTA-SILVA et al., 2011).

1.4.9. Management of AI

Treatment for amelogenesis imperfecta affected patients differs from one individual to another based on their clinical presentation and taking into consideration the severity of AI type, age, and patient's concerns. The primary aims of treatment to improve aesthetics of AI affected teeth, alleviate sensitivity, protect, and preserve the remaining tooth structure and improve masticatory function (Cogulu et al., 2009). A multidisciplinary strategy comprising a Paediatric dentist, Orthodontist and Prosthodontist is essential to attain those goals (Claman et al., 2003).

The care of an AI patient is a lifetime task. Therefore, a comprehensive management including immediate, intermediate, and long treatment plan should be available and guided by the paediatric dentist. The objective for young children with AI is to keep them engaged, motivated into adulthood, with appropriate oral hygiene habits and future treatment that is not hampered by previous dental treatment (Poulsen et al., 2008). In the current description of literature, there are many diverse AI management strategies. However, a weak evidence based for the management of AI present (Dashash et al., 2013).

When contemplating a restorative plan for AI patients, it is crucial to remember that the best patient care considers the stage of dental development (Chen et al., 2013). In the early phases of treatment approaches, prevention must be incorporated with emphasis on good oral hygiene and motivational instructions. Managing dentine hypersensitivity at this stage will aid in the maintenance of good oral hygiene with desensitizing agents, topical fluoride varnish and or tooth mousse CCP_ACP (casein phosphor-peptide amorphous calcium phosphate) that promote remineralization. A thorough dietary analysis and guidance are also necessary. It is critical to emphasize to patients with AI entails a greater caries risk, and poor diet can affect their teeth. Any carious teeth should be treated as soon as possible; nevertheless, teeth with poor long-term prognosis that are not restorable should be assessed and considered for extraction.

In primary teeth, anterior teeth can be restored with composite resin or polycarbonate crowns and preformed metal crowns for the deciduous molars (Crawford et al., 2007a). In the mixed and permanent dentition cast adhesive coping or prefabricated metal crowns can be used, and there is no consensus on which material is more superior (Zagdwon et al., 2003).

Long term treatment plan for management of patient with AI as well as addressing each problem as it arises, have been recommended from the American Academy of Paediatric Dentistry in 2013. Managing young patients in particular may require more behavioral and motivational approaches. Other potential treatment available including bleaching, micro-abrasion, resin

infiltration composite direct or indirect, prosthetic, and orthodontic work (Council, 2013).

The acceptance of direct composite restorations to restore both function and cosmetics with AI affected teeth has grown due to the continuous improvement of adhesive system. Direct composite restoration has several advantages over indirect restorations as it offers the benefit of not requiring complex laboratory procedures, its reversible and relatively consume less clinical time. Despite that when treating paediatric patient, indirect restorations are not recommended until the full development of clinical crown and gingival margins have matured. Whereas ceramic crowns, recession of direct composite restoration can be adjusted to the new gingival margin rather than replacing the whole coronal restoration (Sabatini and GUZMÁN-ARMSTRONG, 2009).

1.4.10. Challenges of AI management

There are several obstacles that AI patients have that must be properly handled as part of their entire rehabilitation. Successful treatment planning and management should take those aspects into their consideration. Some of the most prevalent problems that AI patients face, as well as their causes are summarized in Table 1.5

Restorative challenges	Causes
Psychosocial problems Low self-esteem Reclusive and withdrawn	Often due to being bullied at school as a child
Poor oral hygiene Chronic gingivitis	Patients avoid cleaning due to sensitivity Some avoid cleaning due to poor motivation as teeth are of a poor appearance
Sensitivity Difficult to etch or clean teeth without LA	Thin enamel Exposed dentine
Caries	Poor oral hygiene combined with thin enamel or hypomineralised enamel makes AI affected teeth more prone to rapid caries progression
Discoloration	Yellow dentine shining through thin enamel or may be complete lack of enamel Can be difficult to mask with conservative techniques
Loss of occlusal vertical dimension or alveolar space Loss of interocclusal space	Due to rapid tooth surface loss which may be compensated for by down growth of the maxillary complex Teeth trying to maintain opposing contacts Often require complex rehabilitation involving a reorganized approach and an increase in the occlusal vertical dimension
Reduced inter root space	Thin enamel or rapid loss of enamel post eruption results in teeth drifting closer together Risk of damage to adjacent teeth Difficult to prepare teeth for crowns and take impressions
Large pulp to crown ratio	Young teeth with large pulps. Lack of secondary dentine Increased risk of tooth losing vitality
Gingival maturation resulting in exposure of restoration margin	Occurs over a few months post full eruption of tooth If restoration placed too early, then margin maybecome visible after maturation. If lab made restoration, then it may need replacing
Decreased bond strength of resin to enamel	Higher protein content in AI affected enamel Results in abnormal etch pattern Etch pattern varies between phenotypes Different phenotypes can therefore give different bond strengths
Bonding to dentine	Due to rapid loss of enamel in some AI patients bonding to dentine is required

Table 1-5 Summary of challenges and its causes in AI management (Patel et al., 2013)

1.5. Dental adhesives

The manufacturing of resin based adhesive materials has facilitated countless improvements in the clinical care of dentistry. In 1948 the idea of dental adhesion was inaugurated in the practice of dentistry, when the first adhesive monomers were invented by the Swiss chemist Oskar Hagger (Hagger, 1948). In 1955, Buonocore issued an article “A simple method of increasing the adhesion of acrylic filling materials to enamel surfaces”. This paper considered as the base of adhesive in dentistry, specifically illustrated distinct methods of achieving bonding between the tooth and the filling material (Buonocore, 1955).

The use of resin materials to bond with tooth structure has altered theclinician’s capability and the concepts of tooth preparation, prosthodontic andorthodontic treatment. New adhesive materials increase the demand and the indications for the use of tooth-colored restorations.

1.5.1. Concept of adhesion

“Adhesion is a complex of physical and chemical mechanism that allows the attachment of one substrate to another” (Breschi et al., 2008a). An adhesive is a substance, often a viscous fluid, which binds together two compounds by solidifying and transmitting a pressure from one surface to the other. The measurement of the load bearing capacity of an adhesive joint is called the adhesive or adhesion strength (Akinmade and Nicholson, 1993).

Four mechanisms of adhesion have been identified which are (Allen, 1993):

1. Mechanical adhesion is adhesive interlocking with surface irregularities of the substratum.
2. Adsorption adhesion is a chemical interaction between both the bond and the adherent, through the primary force by ionic and covalent or secondary force by the hydrogen bond, dipole reaction or the Vander Waals valence force.

3. Diffusion adhesion is the interlocking of mobile molecules, including the adhesion of two polymers by polymer chain diffusion, ends at the interface.
4. Electrostatic adhesion is double electric layer across the metal interface with a polymer portion of the overall bonding process.

In dental practice, the bonding of resin-based materials to the enamel and dentine results from four mechanisms (Manuja et al., 2012):

a. Mechanical through resin diffusion and resin tag formation within the tooth structure.
b. Adsorption - chemical bonding of tooth structure to the inorganic component hydroxyapatite or the organic component which is primarily type I collagen.
c. Diffusion – deposits of substances on tooth surface to which resin monomers bind mechanically or chemically.
d. A combination of a, b, and c mechanisms.

For effective adhesion, there must be adequately close contact between the adhesive and enamel or dentine. The adhesive surface tension should be less than the substrate surface energy. Adhesive contact failures arise from three sites that are usually integrated when an actual failure happens: cohesive substrate failure, cohesive adhesive failure and failure of adhesive or substrate and the interface between them both (Ebnesajjad and Landrock, 2014).

1.5.2. Advantages of adhesion

Mechanical bonds are vastly inferior to adhesive bonds as they generate higher concentrations of stress and cannot completely exploit the adhesive properties. Nonetheless, adhesive bonds require substantially greater areas to bear the same load as mechanical bonds. Bonding agents are polymeric materials that can be applied without force, enabling the placement of fragile materials such as ceramics to be placed which cannot be attached with mechanical loading (Pocius, 2012). There are several other benefits of

adhesive bonds over mechanical, including fatigue resistant, better absorption of shock and vibration, possible to bond on different materials (Van Landuyt et al., 2007).

1.5.3. Prerequisites of good adhesion

i. One of the key elements for good adhesive bonds is for the surface to be clean and therefore in a high energy condition. Water films, organic particles and biofilms are often present and interfere with wetting and spreading during the clinical scenarios. They are not reversible by brushing the tooth with dentifrices alone. Teeth primed for dental restoration procedures contain low energy surfaces due to residues and smear deposits that are left on the surface (Baier, 1992).

ii. Surface roughness, wettability is improved through existence of micro-surface roughness for most restorative procedures. Wenzel equation described the relation between roughness and wettability. This equation suggests that wetting for contact angles below 90° is enhanced by surface roughness yet, declined for non-wetting surfaces with contact angles above than 90°. The effect of Wenzel has been approved for dental materials such as composites (Al-Omari et al., 2001, Wenzel, 1936).

iii. High contact angle and good wetting: Wetting, which is reliant on surface tension, also plays a significant role in adhesion. The surface wettability of a liquid is defined by a droplet's contact angle on the surface. Reduced contact angles (less than 90 degrees; wettable) induce beneficial wetting as the fluid can cover a wider surface area; greater angles (more than 90 degrees; unwettable) cause unfavorable wetting as the fluid can stay condensed on the surface Figure 1-12. The contact angle role theory (MALLOY et al.). A wettable surface is described as hydrophilic, and a non-wettable surface referred to as

hydrophobic. Due to the wetting and surface energies hypotheses, adhesion to enamel can be accomplished more readily than adherence to dentin. Enamel consists mainly of hydroxyapatite, which has a high energy surface; dentine also includes collagen and has low energy surface (Marshall et al., 2010).

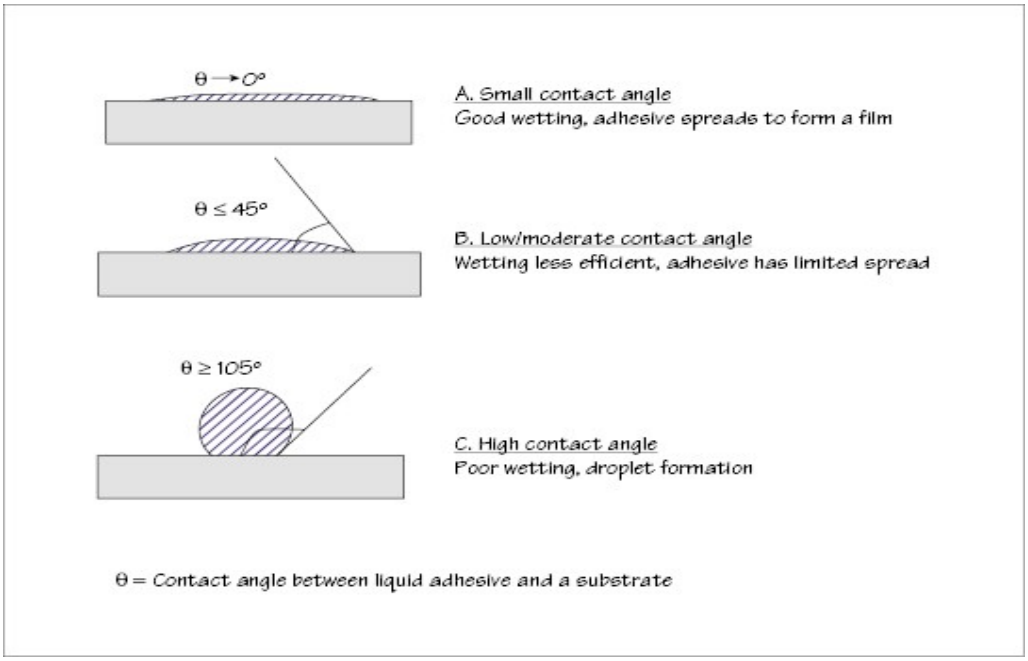


Figure 1-13 The contact angle role theory (MALLOY et al.)

iv. Adhesives with low viscosity and sufficient flow: although the surface tension is ideal for contact, the adhesive usually requires to be low enough in viscosity and capable of adequate flow to disperse and adjust within the time allocated for application to the specifics of the adhering surface. For most adhesives, the nature of viscosity of dental materials in response to shear stress is pseudoplastic (shear stress rises as viscosity declines). Many adhesive systems need effective penetration into the rugged characteristics of small surfaces. The propensity to flow into spaces can be calculated according to a penetration coefficient (Anusavice et al., 2012, Fan et al., 1975).

v. The adhesive separation phase: as the ability of hydrophilic and hydrophobic monomers to polymerize decreases, fluid movements from

dentine has increased, resulting in dilution and phase separation of adhesive solution. Water entanglement could also arise within the adhesive rich surface, negatively impacting adhesive polymerization and durability of the bond (Osorio et al., 2008).

vi. Solidification of adhesives: dental procedures involving adhesive use are compromised by limited clear exposure to curing light. It is apparent that sufficient degree of conversion is required for appropriate adherence. Measuring the relative conversion of available double bonds during polymerization reactions is the most common measuring procedure. This approach only considers the content of the adhesive and not the bonding interface (Yang et al., 2019).

1.5.4. Mechanism of adhesion

There are two stages to this process, firstly the bonding process to enamel and dentine is simply an interchange process comprising the substitution between the resin monomers and calcium phosphate minerals separated from dental hard tissue. These monomers are interlocked micromechanically in the porosities formed during setting. In the second stage the key mechanisms of micromechanical retention include diffusion and capillarity, this method is called microscopic hybridization (Van Meerbeek et al., 2003).

1.5.5. Enamel adhesion

After the implementation of the acid etch technique in dentistry by Buonocore, several researchers have tried to accomplish efficient and long-lasting adhesion methods between resins and the tooth structure.

Acid etching converts the smooth enamel into an irregular surface and raises free energy from its surface. When a liquid resin based material or adhesive mixture is spread to the irregular etched surface, the resin penetrates the surface's micro-porosities, facilitated by capillary action. Monomer in the resin material polymerize, and the material gets mechanically interlocked with surface of the enamel. The key mechanism of resin enamel adhesion is the

creation of resin micro-tags inside the enamel surface (Breschi et al., 2008b, Suppa et al., 2005).

Three different micromorphological patterns result from enamel etching Figure 1-13. The type I etching pattern is accomplished by partial dissolution of prism core without dissolving the prism peripheries. The pattern of type II etching in contrary to type I as dissolution of enamel in the peripheries while the cores remain intact. The etching in type III pattern is a more random and less distinct than the other types, in which it involves surrounding areas of enamel surface resemble to Type I and Type II, and other areas show prism morphology free patterns (Silverstone et al., 1975).

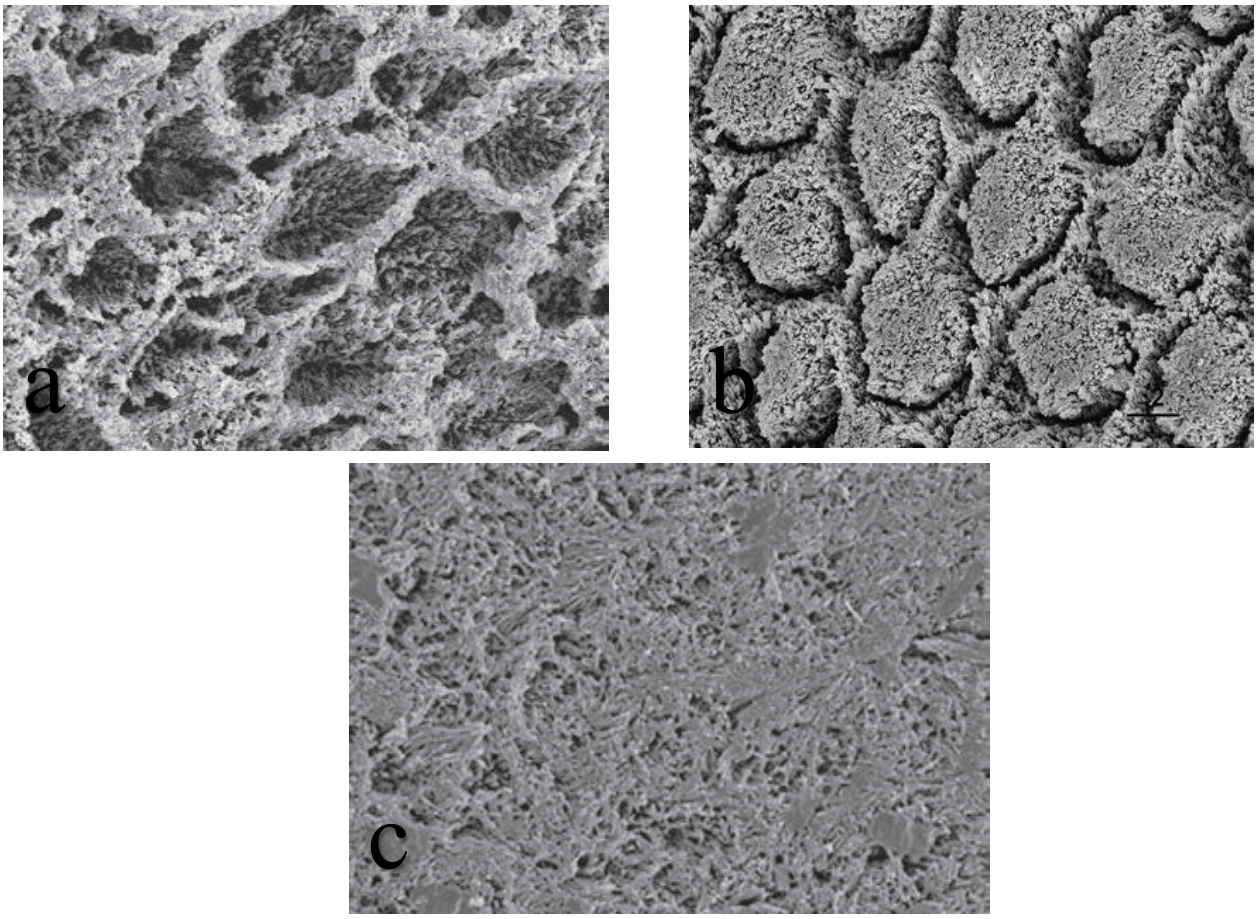


Figure 1-14 Scanning electron microscope: a, b, c showing *Type I, II, III etching pattern respectively, of etched enamel with 35% phosphoric acid for 15s (Silverstone et al., 1975).*

Etching tooth surface with an acidic medium is a standard clinical procedure that leads to the demineralization of the superficial enamel and dentine layers. To achieve this, several acids have been suggested, phosphoric (10 – 50%), fluoridated phosphoric, pyruvic, citric, maleic, oxalic, tannic, ethylenediaminetetraacetic, tricholracetic and polyacrylic acids (Bajraktarova-Valjakova et al., 2018). The acid solution pH and pKa are essential parameters that affects the aggressiveness and capacity of acids to demineralize tooth surface. A further significant factor is the physical state of the solution (gel or liquid). Gel type etching are simpler to handle on the enamel surface than liquid types and demonstrate broader and deeper penetration of the enamel (Baharav et al., 1988). A continuous brushing technique the better it defines the etched pattern during the application of etchant, thus enhancing the marginal adaptation of the resin restoration (Ben-Amar et al., 1988).

1.5.6. Dentin adhesion

Bonding to dentine has been a huge challenge since the acid etching technique was implemented almost five decades ago. Interaction of adhesive materials with dentine can be in various ways which are mechanically, chemically, or both (Yoshida and Inoue, 2012). The value of micromechanical bonding has been acknowledged, identical to what happens in the enamel bonding. Dentine adhesion mainly depends on the infiltration of adhesive monomers into the collagen fibers network left exposed by acid etching. Nevertheless, chemical bonding between polycarboxylic or phosphate monomers and hydroxyapatite was shown to be an integral part of the bonding process for adhesive materials which do not need etching such as glass ionomer cements and some phosphate based self-etch adhesives (Van Meerbeek et al., 2003).

1.5.6.1. Smear layer

Preparing a tooth surface spreads debris over the enamel and dentine surfaces, which will form the smear layer. the definition of smear layer is “any debris calcific in nature, produced by reduction or instrumentation of dentine,

enamel or cementum” (Gwinnett, 1993, Ishioka and Caputo, 1989). The smear layer is a 0.5 – 2 μm layer of debris with granular substructure that fully overlays the dentine, when examined under scanning electron microscopy (SEM) Figure 1-14 (Ishioka and Caputo, 1989). The morphological characteristics, content, and density of this layer are influenced strongly by the type of instrument, the technique of irrigation used and the tooth substrate location (Violich and Chandler, 2010). A blockage of dentinal tubules by a smear layer and can expand 1-10 μm into the tubules, creating a smear plug that are continuous with the smear layer and reduce dentine permeability about 86% (Violich and Chandler, 2010). The layer functions as a physical barrier to dentine permeability and should be eliminated or become permeable for monomers to interface with the dentin surface. The permeability ofdentinal tubules is substantially increased when the smear layer is removed. Under pulpal pressure, there is a mainly outward fluid flow of 20 – 70 cm/H2O (Pashley, 1991).

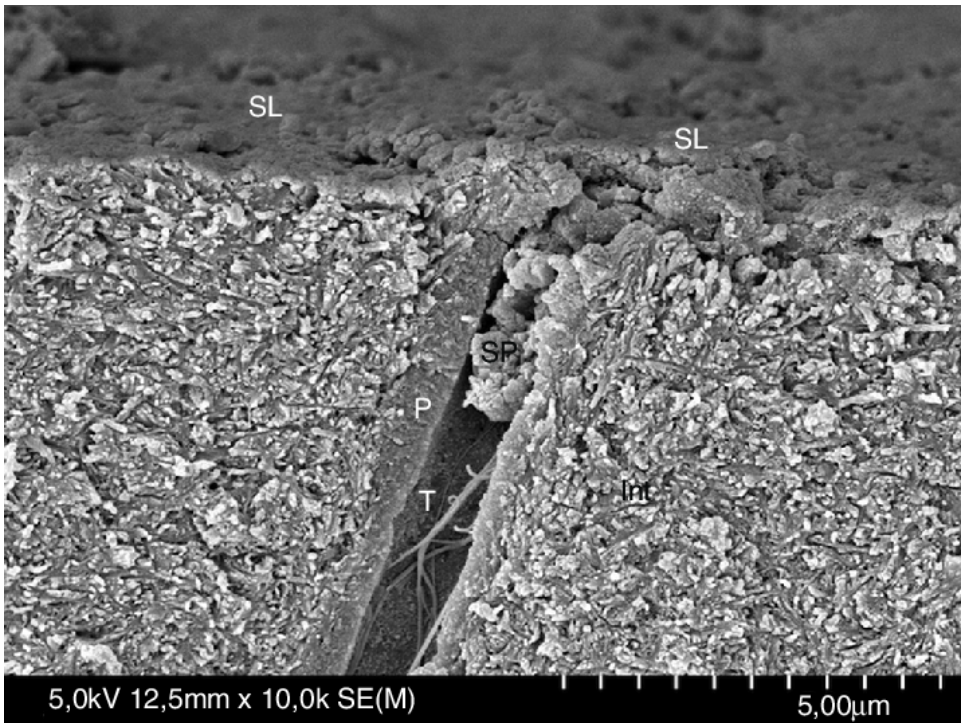


Figure 1-15 SEM micrograph of smear layer and a smear plug magnified $\times\ 10{,}000$*. SL = smear layer, SP = smear plug, T = dentinal tubules, P = peritubular dentine, Int = inter-tubular dentine (Sezinando, 2014)*

There are currently two different methods which can be used in resin bonding procedures: etch and rinse (E&R) and self-etch (SE) or etch and dry techniques. Despite of the strategy used, dentin bonding is based on the formation of a hybrid layer (HL), a framework consisting of demineralized collagen fibrils supported by the resin matrix (Narang et al., 2003). An acid etchant applied in the E&R technique strategy to dissolve smear layer and produce a surface layer of approximately 5-10 mm deep of demineralized dentine.

1.5.6.2. Dentine adhesion strategies

The combination usage of hydrophilic and hydrophobic monomer groups has been proposed to enhance adhesion due to the hydrophilic nature of the dentine matrix. The monomer's hydrophilic activity allows to permeate into the collagen matrix, resulting in the development of a hybridized collagen – resin layer. The hydrophobic features help bonding to the hydrophobic part of the resin matrix of restoration (Sezinando, 2014).

Dentine adhesion process include a conditioner or acid etchant is applied first, then a primer and ultimately a bonding agent or adhesive resin is used. As a result of these steps the bonding and sealing at the dentine restoration interface has improved dramatically. The penetration of primer into the open network of the collagen matrix exposed by dentine demineralization, and it's in situ polymerization is thought to cause hybridization (Perdigao et al., 2006).
During tooth preparation, the smear layer that develops on the tissue surface is an essential factor in the adhesion process. To address the smear layer's poor attachment strength, two techniques have been used: first removing the layer before bonding (etch and rinse approach) and secondly using bonding agents that infiltrate the layer to make it permeable to applied monomers (self-etching approach) Figure 1-15 (Tay and Pashley, 2001).

Total etching adhesives is a separate etch – rinse step is required when using "etch and rinse" adhesives. A range of etching agents have been used, for example maleic, citric, phosphoric, and nitric acids; with 30 – 37% of

phosphoric acid now being the most popular and preferred conditioning agent. Dentine etching exposes the collagen network fibrils that constitute the underlying substrate by removing the smear layer and hydroxyapatite mineral from the tissue surface. The smear layer and the top 1 – 6 µm of hydroxyapatite are dissolved by applying 37% of phosphoric acid to dentine for 15 seconds. The resultant layer is permeable, allowing adhesives to penetrate the fibrils spatial network (Pashley, 1991). The total etching method is regarded as the gold standard for the predicted tooth adhesion and is utilized in many bonding systems. The primer and adhesive resin are combined in a single application using simplified two etch and rinse adhesives approach (De Munck et al., 2005).

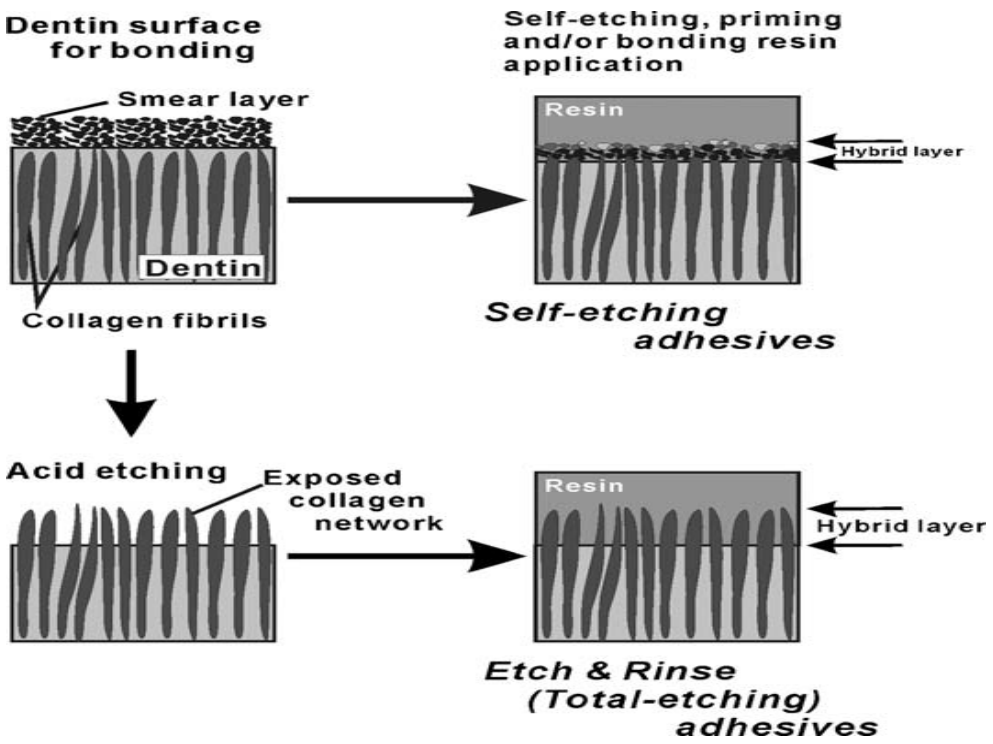


Figure 1-16 illustration of bond structure of a self-etching and etch - and – rinse adhesive system. Resin and collagen fibrils make up the structure of total etch adhesives 'hybrid layer'. A typical hybrid layer of self-etching adhesives results from a mixture of resin, collagen fibrils and inorganic materials. Thinner hybrid layer of self-etching adhesive than that of total etching system because of mild acidic monomer effects on dentine (Hashimoto et al., 2011).

Self – etching adhesives: The application of self-etching adhesives, which are based on rinse free acidic monomers, provide an alternative to the total etching technique. These monomers condition and prime dentine at the same time. This method has proved to be the most user friendly but technique sensitivity (Van Landuyt et al., 2007). Self-etching method eliminate the need for an additional washing step, which will reduce the application time, technique sensitivity and the possibility of administration mistakes. Self-etchingadhesives are applied in one or two steps, including the addition of one or more carboxylic or phosphoric acid groups to the monomers (Tay and Pashley,2001).

There are two types of self-etching system: mild and intense. A strong type of self-etching adhesive has a PH of 1 and create a bonding mechanism and interfacial ultra-morphology in dentine same as etch and rinse adhesives (Van Landuyt et al., 2007). The hybrid layer acquires a thickness of 3 – 4 μm with these adhesives and displays the features if a poorly organized collagen fibril network. Each fibril is divided by interfibrillar gaps at the surface of the hybrid layer, resulting in a “shag carpet” pattern. The other type of self-etching is the mild form it has PH of 2 which can degrade the surface of dentine to a depth of less than 1 μm, resulting in many hydroxyapatite crystals in the hybrid layer. The hybrid layer formed by these adhesives is significantly thinnerthan that created by strong form self-etching or etch and rinse adhesives, however the bond’s efficacy is unaffected. Functional monomers with specificcarboxyl for example 4-methacryloxyethyltrimellitic acid (4-MET) or phosphategroups like 2- methacryloxyethyl phenyl hydrogen phosphate (phenyl – P) or 10 – methacryloxydecyl dihydrogen phosphate (10 – MDP) may interact with the remaining hydroxyapatite (Sezinando, 2014).

1.5.7. Bonding to AI affected teeth

The restorative rehabilitation and management of patients with AI include the use of bonded restorations to enhance aesthetics (Souza et al., 2014). Clinically, however, bonding to AI affected teeth is challenging. The majority of

AI patient complaints of dislodge restorations (Chan et al., 2010). Bonding to AI affected teeth has been reported in very few studies.

The concept of bonding is based on micromechanical interlocking of adhesive resin with the enamel and dentine. As bonding to enamel depends on the micromechanical to the etched surface, whereas dentine depends on the hybridization to the exposed collagen mesh. An etchant typically with 30-40% phosphoric acid degrades the hydroxyapatite crystals and provides gaps for resin infiltration (Sholapurkar et al., 2008). The mineral composition of tooth structure is linked to their ability to interlock micromechanically with bonding adhesives. Enamel is predicted to have a stronger mechanical interlock with the adhesive resin than dentine substrate due to its higher mineral composition (Van Meerbeek et al., 2011). Enamel of normal tooth structure is a highly mineralized with a prismatic pattern of large crystals. While normal architecture of enamel affected by AI is lost and incomplete formation of enamel prisms with the rods obscured by amorphous material and poorly mineralized and therefore lower bond strength. More than 40% reduction in minerals affected by AI (El-Sayed et al., 2010).

From dental service and clinician viewpoint is essential to know what treatment per the type of AI patients need as well as the number, type and failure rate of restorations provided. For this reason, we decided to undertake a service evaluation of the burden of care of AI patients in the Paediatric Dentistry department at the Eastman Dental Hospital.

Following this our main objective was to explore the bonding strength to AI affected teeth and how it differs according to the type of AI. Originally, we intended to carry out a review of available evidence on bonding to teeth affected by AI and then planned to carry out laboratory investigations to explore gaps in the evidence. Because of COVID this was not possible. Therefore, in the final part of this investigation we looked at the use of AI classifications in the published literature.

2. The burden and impact of Amelogenesis Imperfecta care at the Eastman Dental Hospital

2.1. Study aims

2.1.1. Aim of study

- To assess the burden of care for child patients with Amelogenesis Imperfecta (AI) attending the Paediatric Dental Department at the Eastman Dental Hospital (EDH).

2.1.2. Objectives of the study

- To determine the type of treatment, duration, and number of appointments for patients receiving specialist care for AI
- To determine the care burden of AI patients and families in terms of travel and time commitments
- Compare different treatment modalities and burden of care by types of AI
- Investigate the restoration failure for each type of AI

2.2. Materials and methods

2.2.1. Study design

This is a service evaluation of AI patient records within the Eastman Dental Hospital.

2.2.2. Study population

This service evaluation used existing databases of patients with AI from the anomaliesdatabase, and asking colleagues in the Paediatric department, EDH.

2.2.3. Inclusion criteria

• Documented AI definitive or provisional diagnosis in the clinical notes.
• Patients with at least six months care in the service.

2.2.4. Exclusion criteria

• Unclear diagnosis
• Patient records were not available

2.2.5. Ethical Approval

The supervisors and clinical audit lead agreed that this project qualified as an audit, therefore the study was registered as a service evaluation and no ethical approval was needed. The service evaluation protocol was registered with the hospital governance team (Appendix 1: Service evaluation registration form).

2.3. Methodology

2.3.1. Identifying the study population

Patients with AI were identified from the anomalies database which was developed in the paediatric department in 2003, and it has been updated regularly under the supervision of a paediatric consultant. Additional patients were obtained by asking colleagues treating AI patients, to identify any patients that were not included in the anomalies database. A total of 84 patients were identified, and the dental notes of each patient were requested.

2.3.2. Data Protection

All collected data was stored in the postgraduate office with a safe entry door; electronic data was stored on the share drive in an EDH/EDI password protected device. Electronic data was transmitted via protected nhs.net email. The data obtained was anonymized through a unique ID number given to each patient, to ensure that when examining patient's notes could not be identified. Following submission of the study, patient identified information will be destroyed.

2.3.3. Data Collection

Information from both paper and electronic records are obtained retrospectively. All the data were collected from July - December 2019, using a datacollection form .

2.3.4. Pilot data collection forms

Four patient records were piloted, and minor modifications were made to the form including more details of operator level, episodes of care and detailed treatment under GA. A radiograph section was added for the number and type of x-rays taken. All records included in the study were analysed using the final version of the data extraction form (Appendix 2: Data Collection Form).

2.3.5. Data collection

For each patient, the paper data collection form was completed and transformed onto an Excel spreadsheet. The online route planner google maps for each patient, (https://www.google.co.uk/maps/) was used with patient postcodes, to measure the mileage to and from the hospital. Travel times to the Eastman Dental Hospital were also calculated, both by public transport and car. For each patient, treatment modalities were reported and broken down into primary, mixed, and permanent dentition.

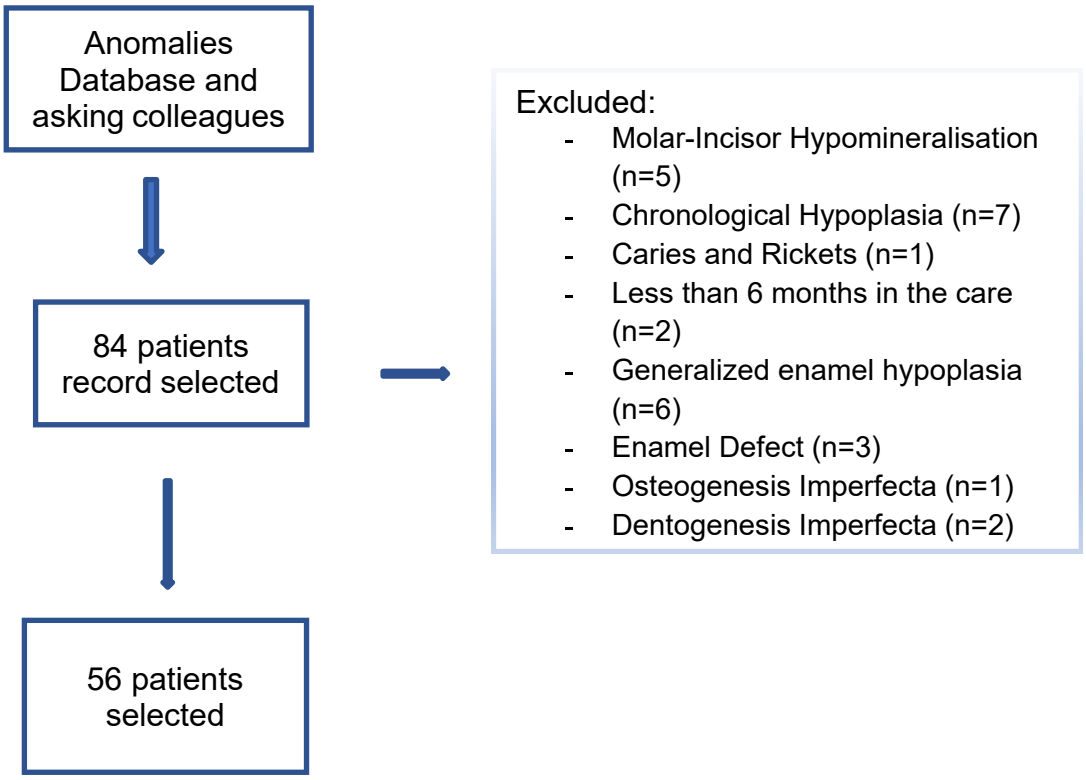
2.3.6. Statistical analyses

Data was collated on a spreadsheet and descriptive statistics such as Mean and standard deviation were produced using Microsoft Excel.

2.4. Results

Of the 84 patients identified, 28 were excluded as they did not match the inclusion criteria, resulting in 56 patients available for analysis. Data included demographics, treatment provided and an estimate of treatment burden.Figure 2-1 illustrates the flow chart used, and for the reasons for exclusion.

Figure 2-1 Process of patient record selection for study.



2.4.1. Gender

The male female demographics split of sample group was 23:33. There were more female AI patients accounts for 59% and 41% for the male within Eastman Dental Hospital.

2.4.2. American Society of Anesthesiologists (ASA) physical status classification

Most of the patients were grade 1 ASA (fit and healthy patients) with 87.5% and 12.5% with mild to moderate systemic disease (ASA 2). Figure 2-2 shows the medical status for dental care facility at paediatric department.

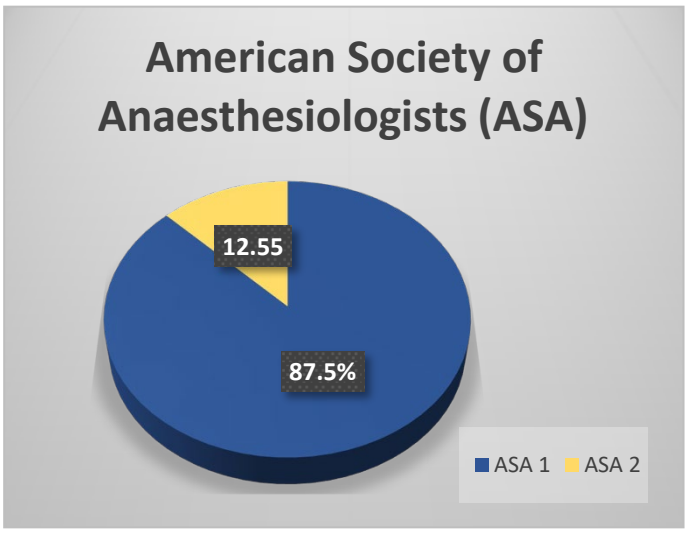


Figure 2-2 ASA classifications of patients

2.4.3. Referrals to Specialist Paediatric Dental Service

Most patients (n=49 87.5 %), were originally referred by their General Dental Practitioner (GDP), as shown in Table 2-1.

Table 2-1 Summary of referral source

Original Referrers	Number of patients	% of Patients
GDP	49	87.5 %
Orthodontist	2	3.5 %
CDS	3	5.3 %
Great Ormand Street Hospital (GOSH)	2	3.5 %

2.4.4. Types of AI as recorded by treating clinicians

The classification of AI patients was divided into two tables. Table 4 shows the classification according to the exact terminology of the treating clinicians was taken Figure 2-3.

Further examination was required in which there was uncertainty in the form of AI such as 6 unknown and 4 hypomineralized. After looking at the x-rays and clinical images, those four patients as well as the one who had not identified by form of AI were further examined. The other ambiguous forms were classified as shown in Figure 2-4.

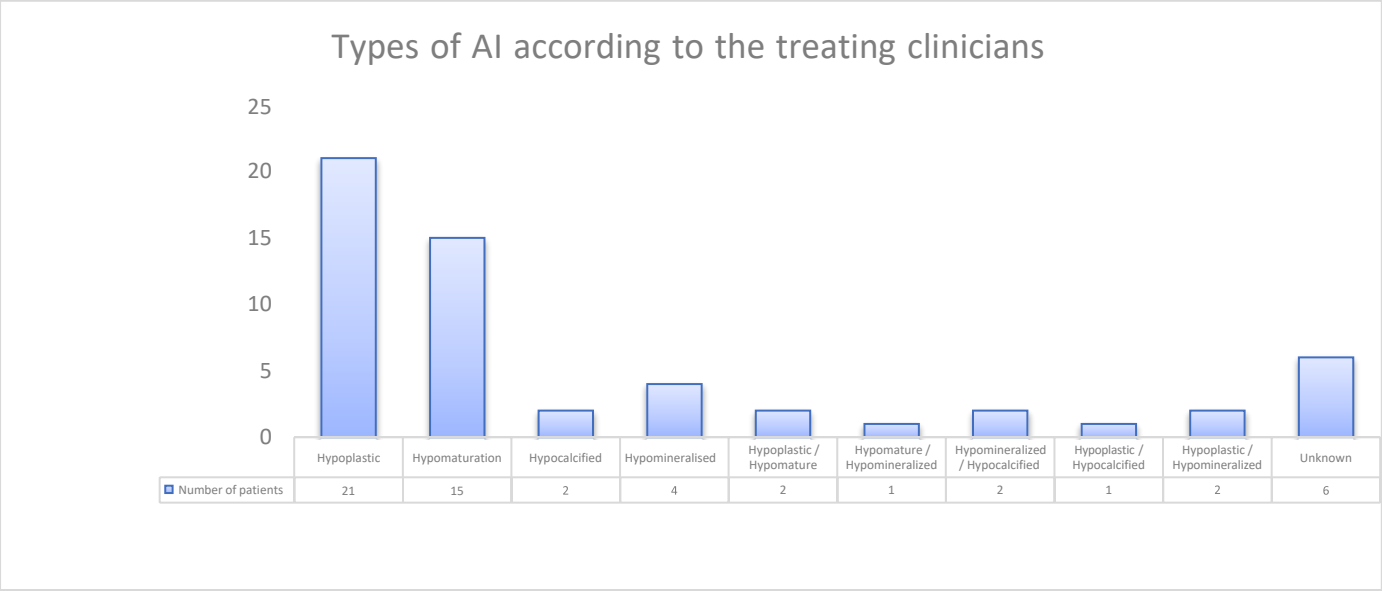


Figure 2-3 Types of AI according to the treating clinicians

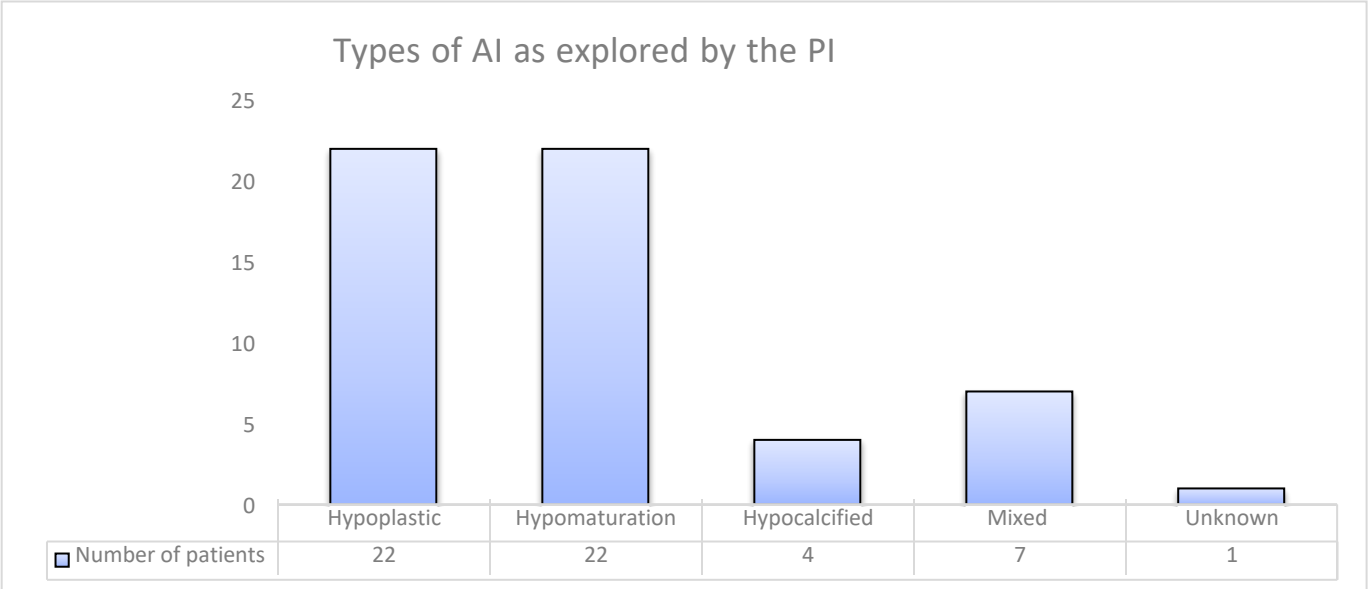


Figure 2-4 Types of AI as explored by the PI

2.4.5. Family History

There was no record in the notes of any discussion with the family about AI family history in 41% of patients n= 23. In 39% of the patients, n= 22, it was documented that there was no history of AI with the family. 20% of patients recorded with family history, n=11 as shown in Figure 2-5. Table 2-2 indicates the reported family history by each patient. Due to the small number of patients reported with family history it was difficult to extrapolate clear genetics conclusions.

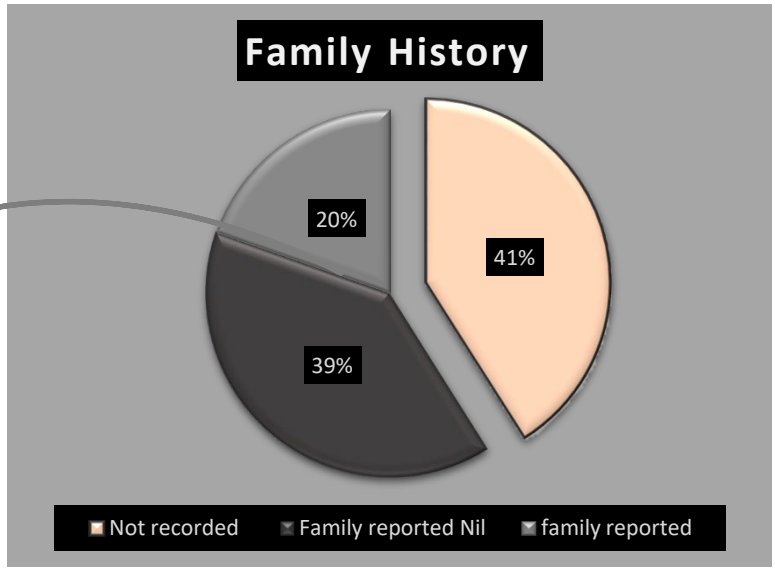


Figure 2-5 Report of family history with AI by percentage

Family History / patient reported	Number of family members recorded per patient
1	1 Brother
2	1 Brother, 1 Sister
3	1 Brother
4	1 Cousin
5	1 Uncle
6	1 Father, 1 Sister
7	1 Mother
8	1 Mother
9	1 Mother, 1 Brother
10	1 Father, 1 Brother
11	1 Sister

Table 2-2 Number of family members recorded per patient

2.4.6. Age

At the time of data collection, the average patient age was 12.8 years. There are eight patients at the age of 16 years old and above who are still in the service. The average age of first appointment for patient is 9.2 and the average age at discharge for patient is 14. As a result of histogram, the data were not normally distributed and do not appear to be symmetrical due to the narrow range of the sample as summarized in Table 2-3.

Table 2-3 Age of study sample in the department.

	Age (years)	Age at 1st appointment	Age at discharge
Average	12.8	9.2	14
Standard deviation (SD)	3.7	2.7	3

2.4.7. Dental development stage of patient while under specialist care

For patients to seek advanced dental care, the most common stages were in mixed dentition or from mixed to permanent dentition. As children were seen in mixed dentition about 82.14%, Permanent dentition 32.14% and primary dentition about 7.14%. Table 2-4 highlight the number of developmental stages in which patients provided specialist care.

Table 2-4 Developmental stages of AI patients in the study.

Developmental stage	Hypoplastic	Hypomature	Hypocalcified	Mixed	Unknown
Primary only			1		
Mixed only	13	16	1	5	1
Permanent only	2	4	1	1	
Primary – Mixed	1		1		
Primary – Permanent				1	
Mixed – Permanent	6	2			

2.4.8. Patients under the care and discharged

It has been found 31 out of 56 of the patients to be under the paediatric service at the time. Two patients were lost to follow up and have not been discharged and considered not a current as well account for 3.6% as shown in Table 2-5.

Table 2-5 Current and discharge patients

Patient management in service No of patients (% of patients)	Current	Discharge
Number	31 55.4%	23 41%

2.4.9. Reason for Discharge

Two patients were lost to follow up and had not been discharged and 18 patients discharged to their GDP. The other five patients who were discharged to specialist care locally. Table 2-6 illustrate reasons of discharge.

Table 2-6 *reason of discharge AI patients*

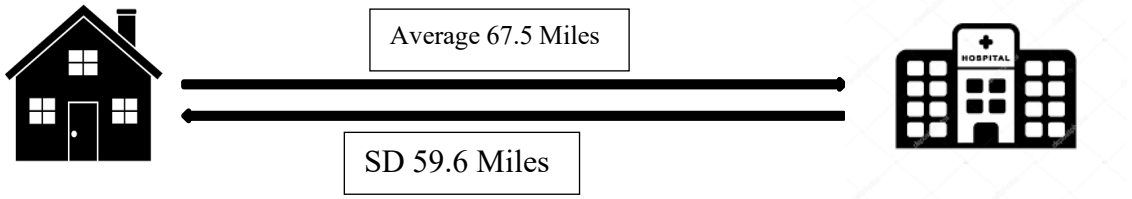
Discharge Reason	Number of patients
Discharged to GDP	18
Discharged to Restorative	2
Discharge to Orthodontist	3

2.4.10. Round trip mileage for appointments

For a single dental appointment, the average distance from home to hospital was 67.5 miles (108.6 km), with 46 miles (74 km) median based on the sample size. The 33.7 percent of patients were within the 15 miles of the hospital as described in Figure 2-6.

There was a significant variation in the average distances from home to hospital. The shortest distance was 4.38 miles (7 km) for Northwest London and the furthest was 143 miles (230 km) from Westham, Weymouth.

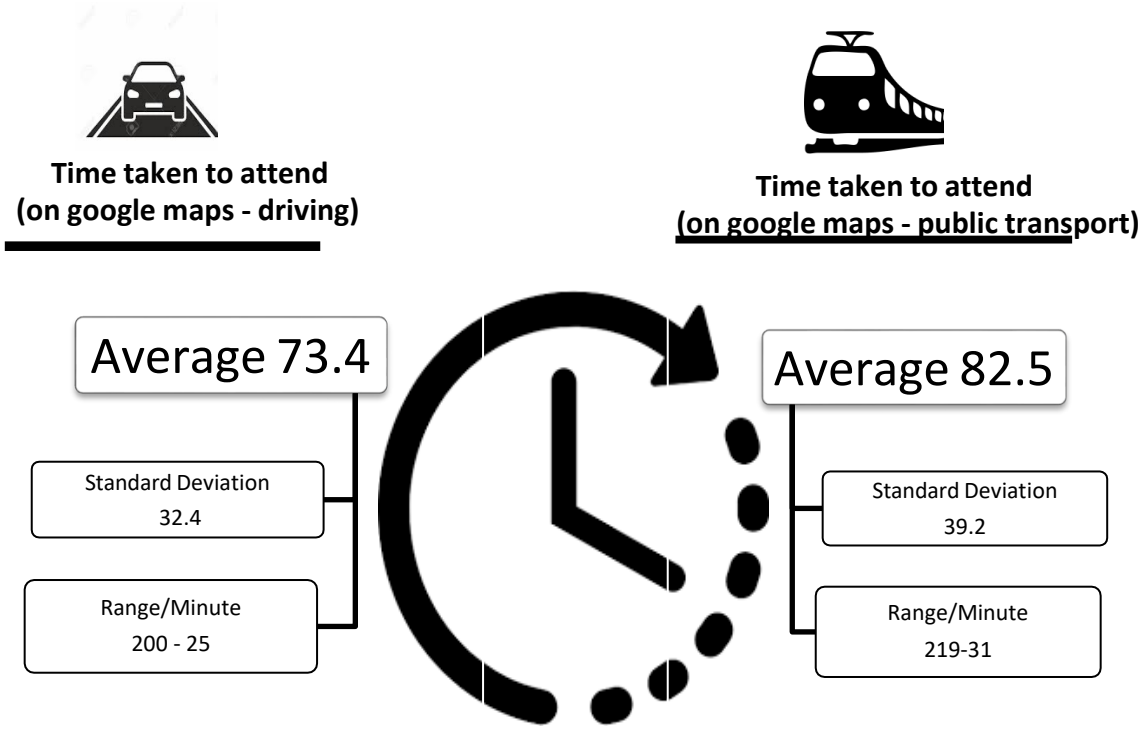
Figure 2-6 Round trip mileage



2.4.11. Time taken to attend appointment

The time taken by either driving or public transport to reach the hospital, google maps as below in Figure 2-7.

Figure 2-7 Time taken to attend one appointment



2.4.12. Duration of care

2.4.12.1. Duration of specialist care

Patients have had an average of 3.4 years of treatment with the specialist care with a range of 6 months to 12 years. The average duration of care for discharge patient in the paediatric department was 4.2 years and number of 23 patients had been discharged as shown in Table 2-7

Table 2-7 Duration of care for current patients

	Duration of care (Months)	Duration of care of discharge patients (years)
Average	3.4	4.2
Standard Deviation	3.1	3.6

2.4.12.2. Average number of appointments per year

The total number of appointments was 297 for all 56 patients on average of 5 appointments. The average numbers of appointments per year per AI type were analyzed as in the Table 2-8.

Table 2-8 number of appointments per year by AI type

Average Number of appointments /years	Mean	Standard Deviation
Hypoplastic	5.2	2.5
Hypomature	4.4	2.4
Hypocalcified	7.4	3.6
Mixed	**4**	1.4
Unknown	4	-
Total	5	2.5

2.4.12.3. Was not brought

The average rate of appointments patient was not brought was 0.7 as shown in Table 2-9 below.

Table 2-9 Patients was not brought to the appointments

	Was not brought
Average	0.7
Standard Deviation	1.1

2.4.12.4. Appointment cancellation

Number of cancellations appointments were looked at from the EPR and EPIC system. The average number of appointments got cancelled either by the patient himself or the hospital as shown in Table 2-10.

Table 2-10 Number of cancellations by the Hospital and Patient in the department

No. of cancellation	Hospital	Patients
Average	3.5	4.3
Standard Deviation	3.6	5

2.4.13. Clinicians involved in care

Patients have been seen by a variety of different clinicians including therapists, specialty registrars, specialty doctors, DCTs, Postgraduate students and consultants. Postgraduate students were the highest number of clinicians who are providing care for those patient accounts for 90 operators about 30% of total. In all cases, the lead clinician were consultants mostly as supervisor or

a treating clinician in some of the patients. The average number of operators per patient as shown in Table 2-11.

2.4.13.1. Number of operators

An average of 5.3 different clinician including therapist providing care per patient. The average number of operators in hypoplastic AI were 6.5 clinician per patient followed by mixed form of AI of an average 4.6 clinician Table 2-11.

Table 2-11 Number of operators per the type of AI

Average Number of Operators / types of AI	Number of operators	Mean	Standard Deviation
Hypoplastic	143	6.5	3.9
Hypomature	102	4.6	2.5
Hypocalcified	12	3	0.8
Mixed	38	5.4	4.6
Unknown	2	2	-
Total	297	5.3	3.5

2.4.14. Anaesthetic types

Five patients have had treatment under general anaesthesia about 9% with repeat of GA in 1.8% and 48% of patients has had at least one appointment of local anaesthetic. This makes 24 of patients with no experience with the L.A or G.A as described in Table 2-12.

Table 2-12 Anaesthetic types and number of patients per the type of AI treated in the department

Anaesthetic types / AI type	GA	GA & LA	LA	None
Hypoplastic	3	2	16	3
Hypomature	1	1	6	14
Hypocalcified	0	0	2	2
Mixed	1	1	3	4
Unknown	0	0	0	1
Percentages	9%	7%	48%	43%

2.4.14.1. General Anaesthetic (GA) episodes

Five of the patients have had dental treatment under a general anaesthetic count of 9% and one patient had two GA which makes 1.8%. All the patients have had the treatment during their mixed dentition developmental stage. There was no planned GA for any of the patients in the study as shown in Table 2-13.

Table 2-13 General anaesthetic episode per the developmental stage

GA episode per developmental stage	Number of GA
Primary dentition	
Completed	0
Planned	
Mixed dentition	
Completed	6
Planned	0
Permanent dentition	
Completed	0
Planned	

2.4.15. GA treatment

Total number of teeth treated under GA by developmental stage as shown in Table 2-14. All the treatment carried out under general anaesthetic was in a mixed dentition stage.

Table 2-15 shows the average number of teeth per patient with highest of composite restorations treatment under GA about 4 per patient and extraction on primary teeth on average 3.6 per patient treated under GA.

Table 2-14 Treatment under general anaesthesia per developmental stage

GA treatment by developmental stage	Number of teeth
Primary dentition	0
Extractions	
Preformed Metal Crown	
Pulpotomy and PMC	
Composite restorations	
Mixed dentition	18
Primary tooth extractions	9
Permanent tooth extractions	7
PMC (primary teeth)	2
Preformed metal crown (permanent teeth)	
Composite restorations	20
Gold Onlays	0
Fissure sealants	12
Exposure of teeth	8
Permanent dentition	0
Permanent tooth extractions	
Permanent tooth extractions	
PMC (primary teeth)	
Preformed metal crown (permanent teeth)	
Composite restorations	
Gold Onlays	
Fissure sealants	
Exposure of teeth	

Table 2-15 Average number of teeth per patient under GA by developmental stage

GA treatment by developmental stage	
<u>Primary dentition</u> **Extractions** **Preformed Metal Crown** **Pulpotomy and PMC** **Composite restorations**	0
<u>Mixed dentition</u> **Primary tooth extractions**	3.6
Permanent tooth extractions	1.8
PMC (primary teeth)	1.4
Preformed metal crown (permanent teeth)	0.4
Composite restorations	4
Gold Onlays	0
Fissure sealants	2.4
Exposure of teeth	1.6
<u>Permanent dentition</u> **Permanent tooth extractions**	0

2.4.16. Treatment types

2.4.16.1. Average number of teeth treated per patient

The whole treatment was combined to evaluate the average number of teeth per patient per AI type that needs intervention. The Table 2-16 below shows the averages and percentages of different treatment per AI type in details.

Table 2-16 Number of teeth treated per AI type

Average number of teeth per patient (n=56)	Hypoplastic n=22, 39.2%	Hypomature n=22, 39.2%	Hypocalcified n=4, 7.14%	Mixed n=7, 12.5%	Unknown n=1 ,1.7	Total 56	Average
Extractions	35/n=10 77.7%	3/n=2 6.6%	0	7/n=1 15.5%	0	45	0.8
Composite restorations	167/n=18 66.8%	39/n=10 15.6%	4/n=2 1.6%	40/n=4 16%	0	250	4.4
Fissure Sealant	85/n=13 45.7%	77/n=12 41.4%	4/n=1 2.15%	20/n=4 10.7%	0	186	3.3
PMC	81/ n=15 67%	11/ n=3 9%	16/ n=2 13.2%	13/ n=2 10.7%	0	121	2.1
Onlays	0	4/n=1 50%	0	4/ n=1 50%	0	8	0.14

Microabrasion	2/ n=1 16.6%	8/ n=5 66.6%	0	0	2/ n=1 16.6%	12	0.21
Bleaching	3/ n=3 18.75%	9/ n=8 56.25%	1/ n=1 6.25%	3/ n=2 18.75%	0	16	0.3
Failed composite restorations	13/ n=167 8%	2/ n=39 5%	1/ n=4 25%	9/ n=40 23%	0	25	0.4
Resin Infiltration	0	0	0	0	0	0	0
X-rays	111/ n=21 55.5%	57/ n=19 28.5%	13/ n=4 6.5%	18/ n=7 9%	1/ n=1 0.5%	200	2.5
Indirect coronal restorations: (PMC+ Onlays+ Indirect composite)	97/ n=17 67%	15/ n=4 10%	16/ n=2 11%	17/ n=3 12%	0	145	2.6
Other	37/ n=20 49.3%	14/ n=9 18.6%	13 / n=3 17.3%	11/ n=5 14.3%	0	75	1.3

2.4.16.2. Preformed Metal crowns

The average number of patients had pre-formed metal crown by Hall technique was 1.76 in comparison to conventional preformed metal crown was 0.23 in both primary and permanent dentition. Number of PMC in primary dentition 77 and 35 in permanent dentition. Conventional technique for preformed metal crown in both dentition account of 13 teeth. Table 2-17 shows number of preformed metal crowns and per AI type and developmental stage.

Table 2-17 Number of Preformed Metal crowns per AI type and developmental stage

No. Preformed Metal Crowns / AI type	primary	Mixed	Permanent	Total
Hypoplastic	8	55	11	74
Hypomature	0	7	4	11
Hypocalcified	16	0	0	16
Mixed	0	9	2	11
Unknown	0	0	0	0
total	24	71	17	112

2.4.16.3. Vital Bleaching

The average of all vital bleaching was 0.3 episode per patient. Hypomature AI cases had vital bleaching about n=9, 56.25%. Table 2-18 illustrate the episode and duration of bleaching and Table 2-19 shows bleaching episode per AI and developmental stage.

Table 2-18 Episode and duration of Vital Bleaching

Bleaching episode	No. of Bleaching	Duration of bleaching in months
Mixed Dentition	7	12.5
Permanent Dentition	9	17.5

Table 2-19 Number of bleaching episodes per AI type and developmental stage

Bleaching episode / Per AI type	primary	Mixed	Permanent	Total
Hypoplastic	0	1	2	3
Hypomature	0	5	4	9
Hypocalcified	0	1	0	1
Mixed	0	0	3	3
Unknown	0	0	0	0
Average	0	0.4	0.6	0.3
Total	0	7	9	16

2.4.16.4. Microabrasion

The average of all vital microabrasion was 0.2. Majority of cases were in Hypomature AI had microabrasion about 66.6%. Table 2-20 illustrate the episode of microabrasion per developmental stage and type of AI.

Table 2-20 Number of Microabrasion per AI type

Microabrasion Per AI type	primary	Mixed	Permanent	Total
Hypoplastic	0	0	2	2
Hypomature	0	3	5	8
Hypocalcified	0	0	0	0
Mixed	0	0	0	0
Unknown	0	2	0	2
Average	0	0.09	0.125	0.214
Total	0	5	7	12

2.4.16.5. Direct Composite Restorations

The total average of composite restorations was 4.1 where the primary dentition has average of 0.02 only. Table 2-21 , Table 2-22 shows number and averages of composite restorations per developmental stage and AI type.

Table 2-21 Number of composite restorations per developmental stage

Developmental stage	No. of composite restorations	Anterior composite restoration	Posterior composite restoration
Primary Dentition	4	4	0
Mixed Dentition	131	86	45
Permanent Dentition	95	50	45
Average	4.1	2.5	1.6
Total	230	140	90

Table 2-22 Number of composite restorations per AI type

Direct Composite Restorations Per AI type	primary	Mixed	Permanent	Total
Hypoplastic	4	94	60	158
Hypomature	0	12	26	38
Hypocalcified	0	6	0	6
Mixed	0	19	9	28

2.4.16.6. Number and reasons of failed restorations

The average number of failed restorations less than one restoration failed per patient seen in all the developmental stage. The most common reasons for restorations failure were debonding, dental caries or secondary caries, discoloration of composite and due to sensitivity post operational. Majority of failed composite restorations occurred in hypocalcified (25%, n=4) and mixed type (23%, n=40) with debonding being the most common reason.Table 2-23 shows number of debonding of composite restoration per AI type and Table 2-24 illustrate the reasons of reasons of failure per developmental stage.

Table 2-23 Number of failed composite restoration due to debonding per AI type (n= indicates the overall composite restorations including the composite restoration provided under general anaesthesia)

Failed Composite Restorations per AI type	primary	Mixed	Permanent	Percentages
Hypoplastic	1	7	5	13/ n=167 8%
Hypomature	0	0	2	2/ n=39 5%
Hypocalcified	0	1	0	1/ n=4 25%
Mixed	0	9	0	9/ n=40 23%
Unknown	0	0	0	0
total	1	17	7	25

Table 2-24 Reasons of failed restorations

Reason of failed restorations	Primary Dentition	Mixed Dentition	Permanent Dentition	Total
Fracture / Fallen restorations	1	17	7	25
Discolorations	0	8	12	20
Defected / 2nd caries	0	2	5	7
Sensitivity post. Op restoration	0	0	1	1
Average	0.018	0.48	0.446	0.94

2.4.17. Radiographs

The average number of x-rays per patient was 3.5 and orthopantomography x-ray was the commonest tool to aid diagnosis was used in 36.5% followed by Bitewings x-ray in 32%. Table 2-25, Table 2-26 illustrate the number, type of x-ray and average per AI type and developmental stage.

Table 2-25 Number of x-rays per developmental stage and type of AI

Number of x-rays per AI type	Primary	Mixed	Permanent	Total	Average / patient
Hypoplastic	2	72	37	111	5
Hypomature	0	39	18	57	2.6
Hypocalcified	2	8	3	13	3.25
Mixed	1	14	3	18	2.5
Unknown	0	1	0	1	1
total				200	3.5

Table 2-26 Types of x-rays per developmental stage

Type of x-ray	Primary Dentition	Mixed Dentition	Permanent Dentition
Bimolars	3	9	0
Periapical	0	19	17
Bitewings	2	41	21
OPG	0	53	20
USO	0	12	3
Total	5	134	61

2.4.18. Other Treatments

The other treatments as described in Table 2-27 shows Fuji as interim restoration with average of 2 per patient. Hypoplastic AI had 59% followed by Hypocalcified AI of about 18.1%.

Table 2-27 Number and percentages of other treatments per AI type treated in the department

Other Treatments	Hypoplastic	Hypomature	Hypocalcified	Mixed	Unknown	Total	Average
GIC	26/ n=10 59%	5/ n=4 11.3%	8/ n=2 18.1%	5/ n=2 11.3%		44	2
Scaling and polishing	2/ n=2 16.6%	6/ n=2 50%	2/ n=1 16.6%	2/ n=2 16.6%		12	0.2
RCT	1	1				2	0.03
Gingivoplasty	1					1	0.018
Soft tissue excision		1				1	0.018
Essex	2/ n=2					2	0.035
Mouthguard	1 33.3%		2/ n=1 66.6%			3	
RPD	1					1	0.018
URA	1					1	0.018
Twin block	1					1	0.018
Tooth Mousse			1			1	0.018
Duraphat	1	1		4/ n=1		6	0.1
Total	37	14	13	11		75	1.34

2.4.19. Inhalation Sedation

Thirteen patients were found to have treatment under inhalation sedation (HIS) accounts for 23.2% with average of 2.5 of all patients has had IHS. Table 2-28 shows the number of inhalation sedation appointments by developmental stage and per AI type.

Table 2-28 Number of inhalation sedation appointments per AI type

Inhalation sedation per AI type	primary	Mixed	Permanent	Total	Average
Hypoplastic	0	28	5	33	1.4
Hypomature	0	3	2	5	0.2
Hypocalcified	0	6	0	6	0.3
Mixed	0	12	2	14	0.6
Unknown	0	0	0	0	0
total	0	49	9	58	2.5

2.4.20. Local Anaesthetic (LA) appointments

Local anaesthetic was administered at least one for 27 patients equals to 48.2 % of all patients. The Table 2-29 shows number of appointments patient had L.A per developmental stage and averages per AI type.

Table 2-29 Number of local anaesthetic appointments per AI type in the department

L.A appointments per AI type	primary	Mixed	Permanent	Total	Average
Hypoplastic	0	41	21	62	2.8
Hypomature	0	7	8	15	0.7
Hypocalcified	0	4	0	4	1
Mixed	0	15	5	20	2.8
Unknown	0	0	0	0	0
total	0	67	34	101	1.8

2.4.21. Summary of key findings

- There is inconsistency of reporting classification of AI as four patients did not have a correct diagnosis, and six have not been classified or inheritance determination documented, nor was it noted if it was as part of syndrome if genetic testing or family pedigree have been carried out.

- Within the limitation of a small sample group, more failed composite restorations occurred in the hypocalcified (25%, n=4)and mixed types (23%, n=40) with debonding being the most common reason (47% n=25).

- Discharge pathway was unclear with lost to follow up patients.

- The average length of treatment was 3.4 years of treatment with the specialist care with a range of 6 months to 12 years and the average duration of care for discharge patient was 4.2 years

- The average number of appointments cancelled by parents was 4.3. with low average rate of was not brought 0.7. Families, however, demonstrated dedication towards their appointments. Although the average number of appointments per year was 5 and for a single dental appointment, the average distance from home to hospital was 67.5 miles with rounded trip on google maps and the time taken by either driving or public transport to reach hospital were 73.4 and 82.5 minutes respectively.

- Patients and their families had a significant treatment burden, with majority of patients receiving treatment under general or local anaesthesia. This include many dental extractions and restorative rehabilitation were documented. Treatment load was further increased by periodontal and hygienist treatment, orthodontic treatment, and indirect coronal restorations with lab work.

2.5. Discussion

2.5.1. Methodology

All the patients were grouped into age and developmental stage as primary, mixed, and permanent dentition as per the developmental stage and per the type of AI to avoid inaccuracy if there are any disturbances in dental development and eruption to allow detailed analysis of treatment and pharmacological behavior management was carried out according to the stage and form. All the radiographic x-rays of all patients were reviewed and analyzed. Also, for certain cases the clinical intra-oral images were examined if the form of AI was not clear or marked as hypomineralized. The classification of AI patients was divided into two tables, one as the exact terminology of the treating clinicians was taken and the second table, further studied in which there was ambiguity in the type of AI (6 unknown, 4 hypomineralized) and the other uncertain types were allocated to hypoplastic and hypomature, hypocalcified as subgroup of hypomineralized and third type is a mixed between hypomineralization and hypoplasia.

The severity of clinical presentation of each type of AI, post eruptive break down, rapid attrition, heavy calculus deposits and gingival hypoplasia could hinder the accurate identification of the type of amelogenesis imperfecta.

Digital photographs and affordable genetic testing in the future could help to improve the correct AI diagnosis.

2.5.1.1. Diagnosis of AI

In this report we refer to the classification of AI by Witkop (Witkop Jr, 1988). As he classified amelogenesis imperfecta into four main categories: hypoplastic, hypomature, hypocalcified and mixed.

Hypoplastic and hypomature were the main types of AI in the sample but the diagnosis was less evident for other patients. None of the subtypes of

hypoplasia has been identified in hypoplastic form. Four patients were diagnosed with hypomineralization, which is not the main type of the classification. Those four patients as well as the one's which had not classified by type of AI were investigated further by looking at the x-rays and clinical photographs.

There was no record of any family discussion about an AI family history in 41 % of patient in the study. It was also noticed in 39.3% that there had been no previous AI background with the family. Understanding that amelogenesis imperfecta is an inherited condition is essential to recognizing the heritage path which is an important part of the cycle of AI diagnosis.

2.5.1.2. Referral and discharged pathway

Most patients were referred during their mixed dentition stage and at first consultation the average age of the patients was 9.2 years. Four patients were under the age of 5 years old seen in the sample and in their primary dentition. Around 87 % patients were referred by their GDP. It is very important those patients got referred at early age for early acclimatization, prevention and to protect wear of tooth structure.

Eight of the patients lost to follow up, 6 of them got discharge to their GDP and two patient they were not discharge neither they are in the service. The number of discharge patients in this study was 23.

2.5.2. Appointments, duration of care, time and mileage travelled to attend appointment

The distance travelled for hospital care varied considerably between individuals. This study focused on distance and time travel to attend one appointment, not only measuring the distance alone. As it has the possibility to underestimate the effect of distance and travel time at health results, at which patients may make several journeys to attend one visit for a dental treatment over the course of the year.

Distance and travel time may be a major obstacle to access to healthcare, failing to attend or cancelling appointments, but may also be included as a key obstacle to health wellbeing, functional disability, travel costs and career or family responsibilities. Those aspects must be taken into consideration when deciding on where to establish specialist care service or improve patient access to an existing service and eventually enhance the health outcomes.
By considering the distance and travel time, the children absence from school will be reduced, the parent or guardian to provide a legitimate excuse for absence to the attendance department at school will be minimized too.

2.5.3. Clinicians involved with care

Continuity of treatment is a defining and primary objective of a paediatric dentist and is associated with the standard of patient care received over the period in the service of care. The consistency of paediatric dentist care can helps gain the trust and confidence and an efficient promoter of the patient's health services through early identification of any problems.

Continuity of healthcare is embedded in a long-term patient - dentist relationship in which the clinician understands the background of the patient from practice and can easily incorporate new knowledge and decisions from the perspective of patient's health totality viewpoint without a thorough investigations or re-examine of records (Harnagea et al., 2017). Given that the majority of the clinicians delivering treatment were postgraduate students, it is critical to strike balance between providing high quality care and meeting training requirements, which include the time, technique, and experience with challenging cases. Therefore, the high number of clinicians providing care is not recommended and needs to be re-evaluated. This can be accomplished by treating children and adolescents with AI in a dental anomalies clinic led by consultant in Paediatric and Restorative dentistry with a focus on treatment planning and availability of local specialist care can reduce the burden as shared care. In addition, it will facilitate for a clear pathway for the transition to adult specialist care if needed in future.

2.5.4. Treatment

Treatment was documented in detail for each patient by the developmental stage and the type of AI. Not to forget that before starting definitive treatment plan, prevention was included in the early stages of all treatment plan, with a specific emphasis on providing good oral hygiene guidance and encouragement for patient. Also treating hypersensitivity with either desensitizing agents, topical fluoride and tooth mousse products encourage remineralization or with interim restorations.

Conservation of the tooth structure is essential in patients with AI, and minimally invasive treatment options were considered where possible as young patients have large pulps and incomplete root formation. Therefore, the use of microabrasion will remove some enamel about 25 – 200 micrometer of the superficial stains and enhance discoloration (Sundfeld et al., 2014). Bleaching and microabrasion were most commonly performed in hypomature group (56%, n= 8 and 67%, n=5 respectively) with an average 0.3 and 0.2. The hypomature AI teeth present with mottled opaque white to yellow- brown discoloration and this treatment approach is successful in managing such cases (Wright, 2002).

More failed composite restorations occurred in hypocalcified (25%, n=4) and mixed type (23%, n=40) with debonding being the most common reason, and this is because of lower mineral content and lower hardness values of teeth affected by hypocalcified AI (Hyun et al., 2009). This is analogous to results in other literature where the longevity of composite restorations is affected as decreases in bonding strength in the hypocalcified and hypomature type of AI (FARIA-e-SILVA et al., 2011). However, the use of rubber dam or anterior cellulose crowns was not recorded in this study, which could have influenced the success or failure rates of anterior composite resin restorations.

The data is likely to underrepresent the amount of treatment required as most of patients were still under ongoing care in the department. No doubt more

care will be required over time. Firm conclusions can hardly be drawn from this data since the severity of AI is unknown in each person in this sample.

2.5.5. General anaesthetic

Treatment under general anaesthesia is assessed on a case-by-case basis taken into consideration the age of child, underlying medical condition, level of anxiety, sensitivity of the teeth, condition of oral hygiene, loss of dental hard tissue, eruption difficulties of the teeth, school absence and school bullying, aesthetic concern with anterior teeth, treatment burden and financial concerns for families in other places. These reasons might be legitimate grounds for planning a GA decision. This could be helpful in reducing the number of the appointments in the surgery and effectively tackling the psychological and social effects linked with AI.

In this study 9% percent of the patient had at least one GA for dental treatment with 1.8% repeated GA. On average extractions of primary teeth were most common 3.6, 1.8 extraction of permanent teeth, composite 4, PMC 1.4 in primary and surgical exposure of 1.6.

Children with AI require lifelong dental care. Clinician must first address the aims and limits of treatment, set reasonable standard to avoid any dissatisfaction with sensible treatment plan under GA is a vital part of delivering service to patients with AI.

2.5.6. Inhalation sedation and local anaesthesia

The technique of inhalation sedation was used in 23% of the patients with an average of at least one appointment for each patient. It was more commonly used in the mixed dentition stage with 87.5%. As inhalation sedation used in reducing the anxiety and making patients to feel more relaxed also it is helpful in reducing the pain and discomfort while administering local anaesthesia.

Local anaesthesia was administered at least once for more than 55% in 31 of the patients with an average of 1.8 appointment. Acclimatization with topical and behavior management in previous visit anticipate what is coming in the next appointment is very important.

2.6. Conclusion

This service evaluation provides data on the burden of care for children with AI. The high number of appointments, treatment needs, and miles travelled illustrate the scope of complications that can occur and stress the need for comprehensivemanagement of this condition. The findings identified some areas within the treatment path that could be strengthened. It is crucial that early identificationof the condition can aid a specialist in acclimatization and prevention at an early stage during their primary dentition as the study has shown referrals werenot being made and the primary teeth either extracted or full coronal coverageused when treatment is subsequently given. It is important to concentrate on classification of AI while diagnosis when seen by a specialized paediatric dentist. Emphasizing on investigation of family history and to be recorded in the clinical notes. As this is life-long condition, this group of patients who will still require further follow up and treatment in adulthood. It important to have aspecific discharge protocol.

Amelogenesis Imperfecta patients experience poor aesthetic, teeth sensitivity, impaired chewing function which needs regular early age dental care. This care will continue life-long and effect of the general well-being of individual himself and his family as well. Most types of AI require intensive dental care, which can take time and place a major burden on services, patient and families as the evidence has shown. In this audit treatment burden investigated in 56 of the patients. treatment plan carried out either under local anaesthesia, inhalation sedation or general anaesthesia differs from patient to another. Also, multi-disciplinary approach in some of the cases involving orthodontic, restorative and lab work will raise the demand to services.

These observations will form the foundation of the advancement of care approaches and seek to enhance the management of AI with emphasis on patient's quality of care by understanding of the burden and impact of care and by the need for developing a clearer pathway of specialist services.

At present, no quality of care set for AI patient management while multi-disciplinary approach can be beneficial. Considerable time and effect expenditure have been demonstrated. Therefore, this field needs further progress with a focus on effective care for AI patients in the paediatric department.

As this service evaluation shows more failed composite restorations occurred in hypocalcified 25% and mixed type 23% with debonding being the most common reason it was important to look at the current evidence of bonding strength to AI affected teeth.

This service evaluation has been published at the European Archives of Paediatric DentistryJournal on the 19 June 2021 (Appendix 3: Published paper at European Archives of Paediatric Dentistry).

3. Bonding Strength to Teeth with Amelogenesis Imperfecta: A Systematic literature review

3.1. Study Aim

To review the available scientific evidence on the adhesive interface between teeth affected with AI and restorative materials in order to recognize the techniques which can enhance their reliability and durability in the different types of AI.

3.2. Material and methods

3.2.1. Inclusion and Exclusion Criteria

Studies eligible for inclusion had to be laboratory or clinical research related to bonding to AI affected teeth. There were no limitations on language or time.

Inclusion criteria were as follows:

- Original articles in humans
- adhesive materials bonded to AI affected teeth
- bonding system to AI teeth
- restorative materials on AI teeth

The exclusion criteria were as follows: studies not related to bonding of adhesive materials to AI affected teeth, single case reports, studies looking at the treatment technique and all vivo studies excluded.

3.2.1. Search strategy

A systematic literature search was conducted using search terms in both electronic search through Medline (Ovid)/ PubMed, the Cochrane library, Google Scholar, hand search journals and websites.

The search method was constructed from keywords related to Amelogenesis Imperfecta along with bonding adhesives keywords. This method was structured as follows: hypoplastic or hypocalcified or hypomature or

hypomineralisation or hypomineralization or amelogenesis imperfecta or AI and bond strength or adhesive or bonding or resin or composite or restorations or retention or survival or durability or longevity or sealant or infiltration.

The searched articles were between 1967 and 2020.The reference list of the originally collected studies were scanned by hand for possible relevancy of papers. The search was carried out between August 2019 and February 2020 Figure 3-1.

3.2.2. Data Extraction and analysis

The data were extracted independently by two authors (HA, PA). For lab-based studies, the number of participants (teeth) with AI / control, storage media, bonding protocols and materials used, the test carried out, and the outcomes were recorded. For clinical trials, the study design, sample size and age range of participants, methodology, follow up and the results were obtained. The classification of AI used in this review was according to the mode of inheritance, clinical and radiographic phenotype, molecular defect, and biochemical result if known (Aldred et al., 2003). Any potential conflict was resolved by discussion between the authors.

3.2.3. Quality assessment

For the non-RCT studies the Newcastle Ottawa Scale (NOS) was used to assess the methodological quality. Based on three predefined domains which each contains 8 elements, the NOS measures the quality of evidence from the score of zero to nine, including: participant selection, comparability, and outcomes. A study with a grade of 0-3 very high risk of bias (poor quality), 4-6 has high risk (fair quality) and 7-9 is high quality.

.

For randomized control trials, we used the Cochrane risk-of-bias tool for randomized trials (RoB2) (Sterne et al., 2019).

shown in

3.3. Results

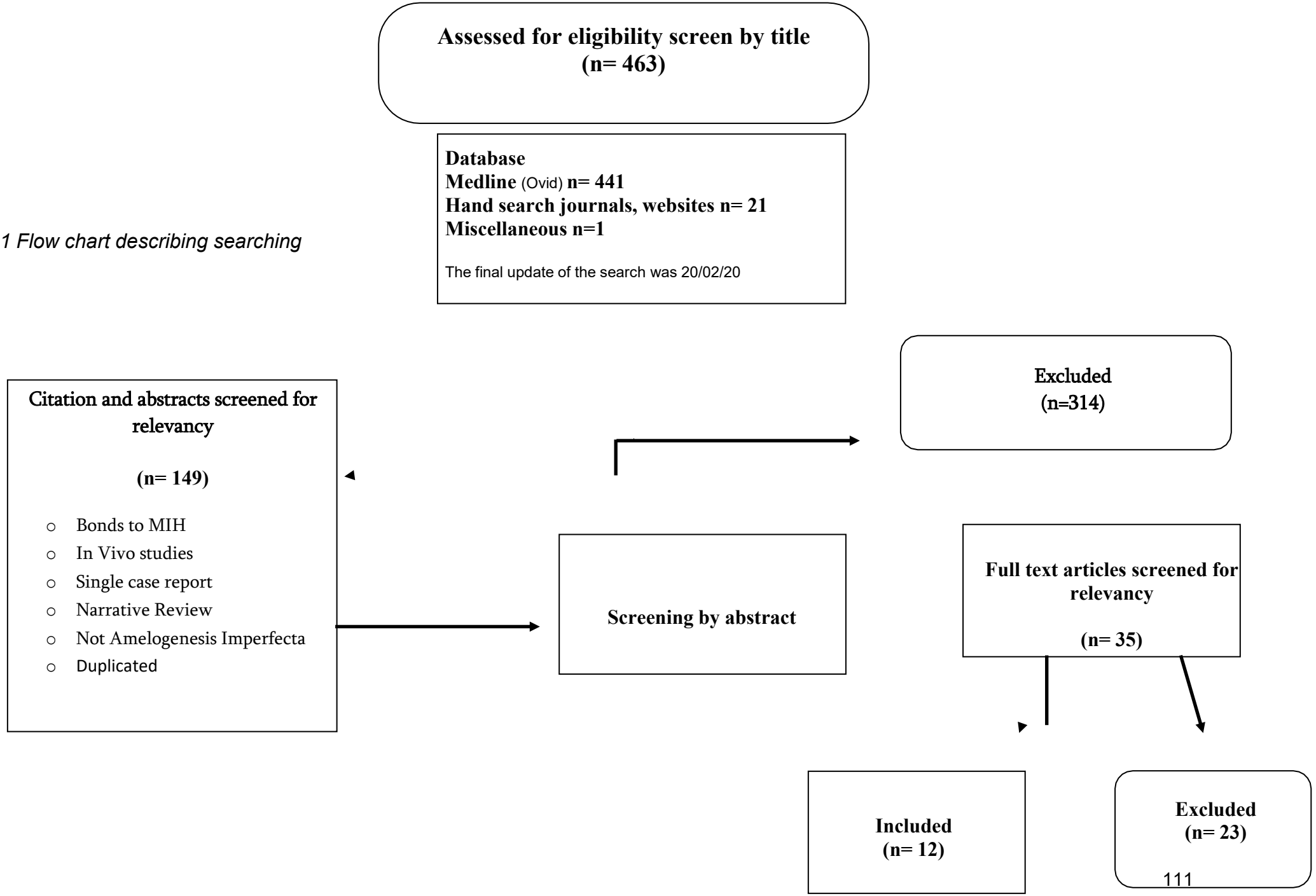
3.3.1. Study Selection

The article selection process summarized in Figure 3-1. The initial search found 463 eligible papers, and titles and abstracts were assessed by two authors (HA, PA). The full text of articles was retrieved and screened according to inclusion criteria resulting in 12 studies: 8 laboratory and 4 clinical studies.

Results of the quality assessment are as described in Table 3-1 and shown in Figure 3-2 with studies being of fair to high quality. Articles selected are shown in Table 3-3 and Table 3-4.

.

Figure 3-1 Flow chart describing searching strategy



Study ID	Selection				Comparability*	Outcome		Total
	Representativeness of exposed cohort (⋆)	Selection non-exposed cohort (⋆)	Ascertainment of exposure (⋆)	Outcome of interest was not present at the start of the study	(⋆⋆)	Assessment of outcome (⋆)	Adequacy of follow up (⋆)	
(Ahmed et al., 2019)	⋆	⋆	⋆	-	- ⋆	⋆	⋆	* ⋆ ⋆ ⋆ ⋆ ⋆ (6)
(Bayrak et al., 2019)	⋆	⋆	⋆	-	* ⋆	⋆	⋆	* ⋆ ⋆ ⋆ ⋆ ⋆ ⋆ (7)
(Epasinghe and Yiu, 2018)	⋆	⋆	⋆	-	* ⋆	⋆	⋆	* ⋆ ⋆ ⋆ ⋆ ⋆ ⋆ (7)
(Chougule et al., 2018)	⋆	⋆	⋆	-	* ⋆	⋆	⋆	* ⋆ ⋆ ⋆ ⋆ ⋆ ⋆ (7)
(Yaman et al., 2014)	⋆	⋆	⋆	-	- ⋆	⋆	⋆	* ⋆ ⋆ ⋆ ⋆ ⋆ (6)
(Lundgren and Dahllöf, 2014)	⋆	⋆	⋆	-	* ⋆	⋆	⋆	* ⋆ ⋆ ⋆ ⋆ ⋆ ⋆ (7)
(FARIA-e-SILVA et al., 2011)	⋆	⋆	⋆	-	* ⋆	⋆	-	* ⋆ ⋆ ⋆ ⋆ ⋆ (6)
(Markovic et al., 2010)	⋆	⋆	⋆	-	- ⋆	⋆	⋆	* ⋆ ⋆ ⋆ ⋆ ⋆ (6)
(Sönmez et al., 2009)	⋆	⋆	⋆	-	* ⋆	⋆	⋆	* ⋆ ⋆ ⋆ ⋆ ⋆ ⋆ (7)
(Şaroğlu et al., 2006)	⋆	⋆	⋆	-	* ⋆	⋆	-	* ⋆ ⋆ ⋆ ⋆ ⋆ (6)
(Seow and Amaratunge, 1998)	⋆	⋆	⋆	-	* ⋆	⋆	⋆	* ⋆ ⋆ ⋆ ⋆ ⋆ ⋆ (7)

Table 3-1 Quality assessment *of studies using Newcastle-Ottawa scale for assessing studies in the systematic review, blue colour to indicate clinical studies*

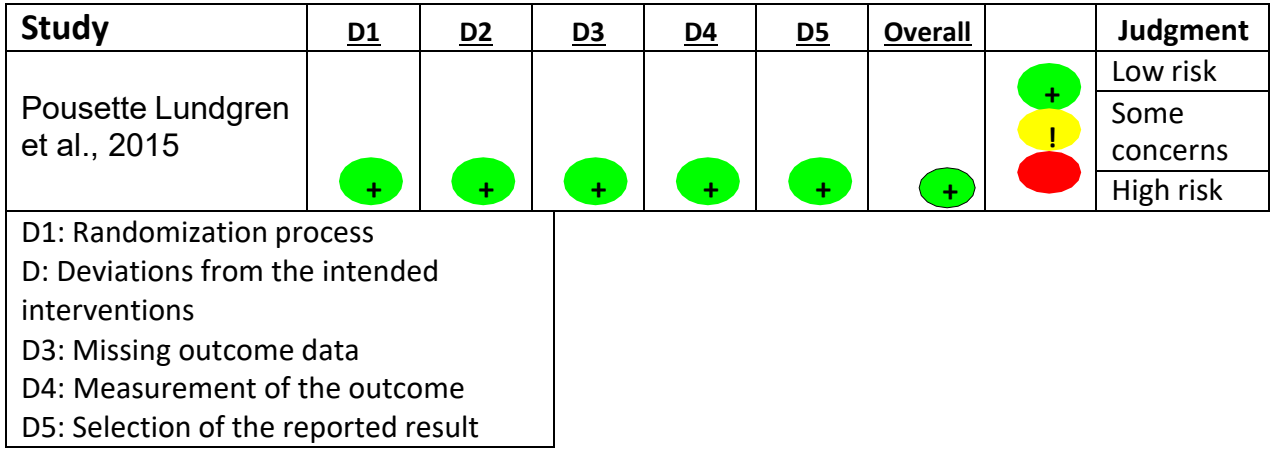


Figure 3-2 Quality assessment adopted from Cochrane Collaboration risk-of-bias tool for randomized trials (RoB2) for randomized controlled trials

3.3.2. Numbers and types of samples included

The laboratory studies included looking at different types of AI, hypocalcified AI was the most common type (5 studies), and one study in each of the following types: hypomature AI, hypoplastic AI and finally a hypoplastic and hypomineralized type of AI. The number of primary teeth included in the five laboratory studies investigated hypocalcified AI were 42 teeth and 18 permanent teeth. The study which examined the hypomature AI obtained 40 permanent teeth for both control and intervention groups. In the hypoplastic AI study 35 teeth were included as shown in Table 3-2.

Table 3-2 Number of samples in laboratory studies

Type of AI	Number of studies	Primary teeth (Sample both study and control)	Permanent teeth (Sample both study and control)	Total
Hypocalcified AI	5	42	18	60
Hypomature AI	1	-	40	40
Hypoplastic AI	2	3	32	35

Two clinical studies investigated hypoplastic and Hypomineralized/Hypomature types of AI. Also, a case series studied different types of AI as followed: hypoplastic, hypomature and hypocalcified. Furthermore, one clinical study looked at hypocalcified AI solely. The number of samples according to the types of AI in each clinical study included in the review are described in Table 3-3.

Table 3-3 Number of samples in clinical studies

Type of AI	Sample	Age (year)
15 – Hypoplastic **12 – Hypomineralized/Hypomature**	27 patients 227 permanent teeth	11 – 22
38 – Hypoplastic **44 – Hypomineralized/Hypomature**	82 patients	6 - 25
Hypocalcified	4 patients 32 permanent teeth	8 -11
8 – Hypoplastic **2 – Hypomature** **2 - Hypocalcified**	12 patients	4 - 17

3.3.3. Study characteristics

Eight laboratory in vitro experiments were included: all studies used (AI affected extracted teeth) and control group. some of the studies also indicates the severity of AI affected extracted teeth. Three of them assessed adhesion quality by microtensile bond strength tests, followed by failure analysis, two used micro-shear bond strength tests without failure mode analysis (FARIA-e-SILVA et al., 2011, Chougule et al., 2018, Yaman et al., 2014). Three studies tested effects of deproteinization on bond strength with 5% NaOCl. Two examined etching patterns using 34% and 37% phosphoric acid and another study looked at etching patterns of 5.25% sodium hypochlorite by using scanning electronic microscope (SEM) (Bayrak et al., 2019, Sönmez et al., 2009, Şaroğlu et al., 2006). Among two of the previous studies evaluated microhardness differences for AI affected teeth (FARIA-e-SILVA et al., 2011) as summarized in Table 3-3.

Four clinical studies included: one randomized control trial during 24 months of follow up with a randomized split mouth design and a patient - blind data acquisition protocol, assessed the quality and longevity of Procera and IPS crowns, with 27 enrolled patients aged between 11 to 22 years of age (Pousette Lundgren et al., 2015). A cross sectional, retrospective study assessed the longevity of dental restoration in 82 patients, 6 to 25 years with mean age 14.5

years (Lundgren and Dahllöf, 2014). Another case series study with a non-randomized convenience sample of 12 patients with follow up postoperatively varied for 2 to 11 years (Markovic et al., 2010). The adhesive bond strength described according to the type of AI as follows:

1. Hypoplastic AI: Yaman et al investigated the microtensile bond strength of self-etch (SE) and etch-and-rinse (ER) adhesive systems to hypoplastic enamel of AI teeth. The adhesive's bond strength to the enamel affected by AI was significantly lower than that of normal enamel, the average μTBS results for research groups shown in Table 3-4. But it also shows better adhesive bond strength in comparison to the Hypocalcified AI (HCAI) were 14,2 MPa with the etch-rinse (ER) Adoper Single Bond 2 adhesive system in the study by Faria-E-Silva et al.

Table 3-4 Microtensile bond strength of adhesive systems to the enamel affected by HPAI and control groups.

Groups	Bond strength (MPa)
Group 1 (ER-control)	31.59
Group 2 (ER-HPAI)	19.63
Group 3 (SE-control)	29.24
Group 4 (SE-HPAI)	18.21

2. Hypomature AI: Chougule et al measured the shear bond strength (SBS) on orthodontic brackets by a conventional bonding procedure to hypomature AI teeth which has shown the lowest SBS at 5.48 MPa versus control group which was at 11.5 MPa.

3. Hypocalcified AI:

    a. Enamel and dentin shear bond strength were investigated by Şaroğlu et al in his study. The enamel shear bond strength of hypocalcified

AI teeth (13.92 MPa) was significantly lower than sound primary teeth (27.77 MPa). In addition, bond strength to dentin of HCAI teeth showed lower values (10.08 MPa) in comparison to normal teeth dentin (18.52 MPa).

b. Faria-E-Silva et al investigated the hardness and micro-shear bond strength for both the enamel and dentin of permanent hypocalcified AI teeth. The hardness of sound enamel (360.4 KHN, kgf/mm$^{2)}$ was greater than the hardness of hypocalcified enamel of AI teeth (53.3 KHN, kgf/mm$^{2)}$, while the hardness of dentin did not vary between sound or hypocalcified AI teeth. The dentin bond strength for both sound and AI affected teeth (24.6 MPa, 30.3 MPa respectively) was substantially higher than the enamel bond strength for the control group 24.0 MPa and intervention HCAI group 14.2 MPa.

c. In his study, dentine of Hypocalcified AI teeth were tested to investigate the effects of additional etching on microtensile bond strength. He found that that bond strength to dentine of HCAI affected teeth 19.27 MPa was substantially lower than that of sound dentine which was at 26.26 MPa. Additional phosphoric acid etching greatly decreased the bond strength of adhesive to sound dentine at 29 .44 MPa and did not enhance the bond strength to hypocalcified AI (24.62 MPa) (Epasinghe and Yiu, 2018).

In the clinical studies included in this review:

- Hypocalcified AI: One randomized control trial included in the study, compared two different types of crowns Procera and IPS e-max and found no significant difference in quality between two crowns with success rate of 97% after 2 years and significant decrease in post treatment tooth sensitivity (Pousette Lundgren et al., 2015).

- different types of AI (Hypoplastic & Hypomature): In another cross sectional, retrospective study by Lundgren et al found

that longevity of composite resin and glass ionomer restorations less than control group or the prosthetic crown treatment. As well as shorter longevity for mixed amelogenesis imperfecta compared to hypoplastic AI. After 5 years, the survival rate of composite resin restorations in AI affected teeth was 50%, which was substantially less than the 80% survival rate in the control group.

- Hypoplastic, Hypomature and Hypocalcified AI: non-randomized convivence sample in a case series with different restorative treatment modalities provided for a follow up period varied from 2 to 11 years.

- Hypocalcified AI: a study investigated the clinical success of deproteinization.

Via these clinical based and laboratory trials, various bonding protocols have been applied to enhance adhesion to AI affected teeth. Four experiments evaluated different adhesives forms (etch and rinse adhesives, self-etch adhesives, or universal adhesives) (Epasinghe and Yiu, 2018, Sönmez et al., 2009, Şaroğlu et al., 2006, Bayrak et al., 2019). Four studies applied deproteinization method sodium hypochlorite (NaOCI application).

All the studies included evaluating bonding to AI affected teeth with normal enamel showed substantially lower strength in bonding to AI affected teeth.

Table 3-5 The laboratory studies included: features of the teeth, materials, bonding technique, tests, and findings:

Author, year	Number of AI-affected teeth	Number of Sound teeth	Type & Severity of AI	Tooth storage media	Materials used	Bonding protocol	Performed tests	Results
(Chougule et al., 2018)	30	10	Hypomature AI - severity: not described	24-h storage in distilled water at 37ºC:	-5% NaOCl - 2% NaF - 37% phosphoric acid - Transbond XT primer (3M Unitek) - Transbond XT adhesive (3M Unitek) - Universal testing machine (UTM)	**For all groups:** no grounded of enamel, phosphoric acid etching (15s), washed and dried until frosty appearance. **Group I (control):** dried, primer on bracket, adhesive (thin coat) centre of crown, air thinning, composite, light cure (40s) **Group II:** same as control group but	μSBS test (UTM)	**μSBS:** **Gr 1 (control):** 11.505 MPa **Gr 2 (conventional bonding procedure):** 5.48 MPa **Gr 3 (NaOCl conditioning):** 6.659 MPa **Gr 4 (NaF conditioning):** 7.651MPa μSBS: very substantial difference between the strength of all four groups were observed. In AI cases, brackets bonded by conventional technique displayed

						bracket bonded on hypomature enamel of AI teeth **Group III:** rinse (15s), dry, 5% NaOCI (1 min), etching (15s), rinse & drying, primer and adhesive (thin coat), light cure **Group IV:** 2% NaF (4 min), 2* rinse (5 min), etching, rinse & dried, primer and adhesive, light cure		lower SBS than NaOCI and NaF.

(Epasinghe and Yiu, 2018)	4	4	Hypocalcified AI All enamel had chipped exposing dentine with yellowish-brown discoloration	5% chloramine T at 4º C (Maximum 6 months)	-Clearfil SE bond (Kuraray, okayama, Japan) -Caulk tooth conditioner gel (Dentsply De Trey, York, PA, USA; 34% phosphoric acid -Microhybrid composite (Filtek Z250; 3M ESPE,	Normal teeth were grounded with 180-grit silicon carbide paper under running water (30s). The AI teeth prepared without instrumentation and the bonded surface was therefore clear of smear layer. Two groups according to the presence and absence of the etching step: Group 1: adhesive, light cure, composite placement Group 2: etching (15s), rinsing (15s), drying, adhesive, composite placement	After 24-h storage in distilled water at 37ºC: µTBS test + failure analysis (UTM, SEM)	µTBS of Clearfil SE bond to normal and AI affected teeth: **Clearfil SE Bond only:** * Sound dentine 36.16 MPa * AI affected dentine 19.27 MPa 2. (SE with etching): * Sound dentine 29.44 MPa * AI affected dentine 24.62 MPa **µTBS:** significant difference between sound and AI affected dentine. Additional etching: Adverse effect on adhesive to sound dentine No improvement on adhesive to AI affected dentine **Failure Analysis:** Both AI affected teeth exhibited similar pattern of failure

(Yaman et al., 2014)	18	14	Hypoplastic AI -	0.9% sodium chloride in water at 4º C for one week	- Adoper single Bond 2 3M ESPE, USA - Clearfil SE bond (Kuraray, Kurashiki, Japan) -Filtek Supreme XT 3M ESPE, Germany	-For all groups: bonding mesial and distal of enamel surface, grounded enamel (600-grits), washing (15 s). Group I & II: ER adhesive (control) & ER adhesive (HPAI affected enamel), etching (15s), rinsing (15s), adhesive (2 coats), air thinning (5s), light cured (10s), composite placement, light cured /layer (20s). Group III & IV: SE adhesive (control) & SE adhesive (HPAI affected	µTBS test + failure analysis (UTM, SEM)	µTBS: **Gr 1 (ER- control):** 31.59 MPa **Gr 2 (ER- HPAI):** 19.63MPa **Gr 3 (SE - control):** 29.24MPa **Gr 4 (SE- HPAI):** 18.21 MPa **µTBS:** No significant difference between SE and ER adhesives.

						enamel), SE primer (20s), air thinning, adhesive, air drying, light cure (20s), composite placement, light cured/ layer (20s)		
(FARIA-e-SILVA et al., 2011)	5	5	Hypocalcified AI - severity not described	0.05% thymol saline solution (maximum 3 months)	-5% NaOCl - Single Bond 2 (3M ESPE, USA) -Filtek™ Z350 restorative (3M ESPE)	-For all groups: Enamel surface was grounded 1200 grit for enamel test -Half the number hemi-sectioned teeth received conventional bonding procedure - the other corresponding half of the same tooth was immersed in 5% NaOCl (Washed and air - dried)	Microshear bond strength test + Knoop Hardness number for both enamel and dentin (MTM, KHN)	µSBS: **Gr 1 - Enamel (ER-control):** 24.0 MPa **Gr 2 – Enamel (ER-HCAI):** 14.2 MPa **Gr 3 - Dentin (ER - control):** 30.3 MPa **Gr 4 – Dentin (ER-HCAI):** 24.6 MPa Hardness (KHN, kgf/mm2): **Gr 1 - Enamel (control):** 360.4 **Gr 2 – Enamel (HCAI):** 53.3

						All samples: ER adhesive applied according to manufacturer's instructions, air thinning (5s), light cured (10s), composite placement, light cured (20s) For all groups Enamel surface was wert grounded again for 600 and 1200 grit for Dentin test		**Gr 3 - Dentin (control):** 51.1 **Gr 4 – Dentin (HCAI):** 57.1 **µSBS:** significant difference between the dentin bond strength and enamel bond strength in both sound and AI affected teeth -Exposure to NaOCl did not affect or enhance the bond strength for control and study groups -Positive linear behavior between enamel hardness and bond strength
(Şaroğlu et al., 2006)	7	7	Hypocalcified AI Advanced destruction of all the teeth with generalized yellow brown discoloration of enamel	storage in deionized water until used	20% phosphoric acid (Heraus Kulzer, Germany) 5% NaOCl Gluma One Bond (Heraus Kulzer, Germany)	Enamel surfaces of all samples were moist grounded (200 – 400 and 600 grit) **Group 1 (control):** - enamel and dentine	After 24-h storage in deionized water at 37ºC: µSBS test (UTM)	µSBS: **(Control and deproteinized enamel surfaces):** Gr 1 (control + sound): 27.77 MPa Gr 2 (control + HCAI): 13.92 MPa Gr 3 (NaOCl + sound): 23.74 MPa

					- Charisman microfilled composite (Heraus Kulzer, Germany	surfaces of HCAl and sound primary teeth were etched (20s), washing (5s), drying (1 - 2s) **Group 2 (study):** -Application of 5% NaOCl (60s) after etching, washing, drying, adhesive (2 coats), composite placement, light cured (60s)		Gr 4 (NaOCl + HCAl): 27.36 MPa (**Control and deproteinized dentin surfaces):** Gr 1 (control + sound): 18.52 MPa Gr 2 (control + HCAl): 10.08 MPa Gr 3 (NaOCl + sound): 19.91 MPa Gr 4 (NaOCl + HCAl): 9.13 MPa **µSBS:** significant enhancement in enamel bond strength in treatment group compared to conventional procedure Application of NaOCl did not affect dentin bonding in control and treatment groups
(Bayrak et al., 2019)	9	9	Hypocalcified AI with physiological root resorptions	0.1% thymol at 25º C	- 37% phosphoric acid etchant (Bisco,	all samples were moist grounded (320,	After 24-h storage in deionized water at	**µTBS:** Group 1: (control + sound): 19.44 MPa

					Schaumburg, USA) - 5% NaOCl (Werax, Spotdent, Turkey) - 0.12% aqueous ClO2 (Solumium Dental, Hungary) - Single Bond 2 (3M ESPE, USA) -Filtek™ Z250 restorative (3M ESPE) -Light cure (Elipar Free light II, 3M ESPE)	400 and 600 grit) **Group 1 (control):** etching (30s), rinsing, drying **Group 2 (NaOCl):** etching (30s), rinsing & drying, 5% NaOCl (60s), rinsing & drying **Group 3 (ClO2):** etching (30s), rinsing, drying, 0.12% ClO2 (60s), rinsing and drying **For all groups:** adhesive (20s), air thinning (5s), light cure (10s), composite placement	37ºC: μTBS test + failure mode (UTM)	(control + HCAI): 12.20 MPa Group 2: (NaOCl + sound) 20.47 MPa (NaOCl + HCAI) 13.12 MPa Group 3: (ClO2 + sound) 24.4 MPa (ClO2 + HCAI) 15.51 MPa **μTBS:** No substantial difference between control values and the NaOCl groups was observed. Nevertheless, the ClO2 results was significantly higher than the control and NaOCl groups. Failure analysis: Mainly adhesive failure was observed No mixed failure identified cohesive in dentin 6% (only for HCAI enamel) Cohesive failure in sound teeth 3%

(Seow and Amaratunge, 1998)	5	2	Hypoplastic and Hypomineralized AI -	The patients had kept their teeth dry until the time of study	37% phosphoric acid 0.5% NaOCl 50 mm of silver	Teeth were immersed in ultrasonic bath of 0.5% NaOCl (30 mins), drying, etching (60s), washing (60s), drying, teeth coated (50 mm silver)	Etching patterns (SEM)	In sound teeth group type 1 and 2 patterns were observed **Etching pattern of AI teeth:** Pitted HPAI: type I pattern X-linked (female): type II pattern X-linked (male): type III pattern Hypomineralized: equal distribution of type I, II, III patterns Smooth hypoplastic: no significant change
(Ahmed et al., 2019)	10 (study group HCAI pre-treated with NaOCl)	10 (control group HCAI etched with phosphoric acid)	Hypocalcified AI -	In distilled water at room temperature	37% phosphoric acid 5% NaOCl	Group 1 (control): etching (15s), washing, and drying (10s) Group 2 (study): 5.25% NaOCl (20s), rinsing (20s), drying (10s), etching (15s)	Samples fixed in 25% glutaraldehyde in phosphate buffer, washing and drying for analysis of etching patterns (SEM)	**Etching patterns:** Group 1: showed type I, II and predominance of type III (65.63%) etching patterns Group 2: display surface roughness with predominance of type 1 & 2 etching patterns 5.25% NaOCl prior for etching enhances the etching patterns

Authors, year Study design	Study design	Number of patients (Age range)	Number of AI teeth	Control group	Type & Severity of AI	Materials used	Bonding protocol	Follow up	Results
(Pousette Lundgren et al., 2015)	Prospective, randomized, single-blind clinical trial (a split-mouth experimental design)	27 (11 – 22 yr.)	119 Procera crowns	108 IPS emax crowns AI affected teeth	Hypoplastic and Mixed AI Severity – not described	- Procera crowns - zirconia inner coping - IPS emax crowns	**Group 1:** 119 Porcera crowns **Group 2:** 108 IPS Emax press crowns (without zirconia inner coping)	- Permanent restoration monitored at 1, 12, and 24 months (100% overall recall rate) - Each visit: quality of restorations, comparing anatomic form, marginal integrity, surface, and colour (according to 1977 California Dental Association Guidelines), caries, gingival bleeding, trauma history, endodontic problems - X-rays follow up: apical radiograph at 2 years	-No significant difference between Porcera and IPS e.max press crowns. -In 97 % of crowns in both crown groups had excellent or acceptable quality after 2 years. -Significant reduction in tooth sensitivity -3% of crowns had endodontic complications
(Lundgren and Dahllöf, 2014)	retrospective cross-sectional study	82 (6 – 25 year)	326 composite resin restoration	63 composite resin restoration	15 – Hypoplastic 12 - Hypomature - 6 mild - 30 moderate - 46 severe	Clinical cross sectional Retrospective study of dental records	- Clinical examination: family history, caries, gingival bleeding index, number of restorations, type of restorations, quality of restorations, comparing anatomic form, marginal integrity, surface and colour, bitewings x-rays if available radiograph more than 2 years. - Data from dental records 10 years and more: calculation of longevity of the restorations in months, date of restoration at the start, date of replacement or	-	- longevity of dental restorations was significantly lower in AI patients. - 24.7% of the AI group require replacement during the observation time - Mix AI have shorter restorations longevity than Hypoplastic AI - Composite restorations lower

Table 3-6 The clinical studies included: study design, teeth characteristics, materials, bonding protocol, follow-up, and results

							extraction as end point, indication and cause of failure for restoration		survival than Porcelain crowns
(Sönmez et al., 2009)	Case control study	4 (8 -11 year)	14	18	Hypocalcified AI	- 20% Phosphoric acid (Heraeus Kulzer) - Gluma one bond (Heraeus Kulzer) - strip crowns (Swedent, Akarp) - Charisma composite (Heraeus Kulzer) - light cure (Polofil Lux unit, Voco)	**Group 1** (control): 20s etching, rinsing 5s, drying 1-2s, adhesive (2 coats), light cure 20s, composite placement **Group 2** (treatment): etching 20s, 5% NaOCl 60s, rinsing, adhesive, composite placement	Clinical success: US Public Health Service (USPHS) modified Ryge criteria up to 36 months	- Deproteinization had no significant effect on the success of the adhesive restorations
(Markovic et al., 2010)	Case series (non-randomized sample)	12 (4 – 17 year)	-	-	8 -Hypoplastic, 2 Hypomature, 2-Hypocalcified - Severity not described	- full dental treatment - photographs	Case series of 12 patients with full preventive and treatment plan	Treatment follow up between 2- 11 years with a recall every 3 months	- Complex dental treatment - No measurement of failed restorative treatment

Table 3-7 The clinical studies included: study design, teeth characteristics, materials, bonding protocol, follow-up, and results, continued

3.3.4. Types of bonding

3.3.4.1. Different adhesives

Various types of adhesives used in the laboratory studies included in the review. Those are described according to the AI type:

Epasinghe and Yiu et al, Yaman et al and FARIA-e-SILVA et al evaluated the use of a self-etch adhesives (SE) (Clearfil TM SE Bond) and a etch and rinse (ER) (Adoper Single Bond 2 3M ESPE).

- Hypoplastic AI: Yaman et al was the only study which compared the use of self-etch (SE) with a etch and rinse adhesives, found nosignificant differences (Clearfil TM SE Bond 19.63 MPa and Adoper Single Bond 2 3M ESPE 18.21 MPa).
- Hypomaturation AI: In his research Chougule et al has used 37% orthophosphoric acid for 15s and a thin coat of Transbond XT primer and adhesive (3M Unitek) in all the groups.
- Hypocalcified AI:
    1) Epasinghe et al tested the self-etch adhesive, the Clearfil TM SE bond to sound dentine and dentine of HCAI in one group and an additional etching step with 34% phosphoric acid for the intervention group with the same Clearfil TM SE adhesive.
    2) In his study, assessed the two steps, etch and rinse adhesive with self-etch Adoper Single Bond 2 (3M ESPE) was applied to all samples to examine the microshear bond strength (FARIA-e-SILVA et al., 2011).
    3) Another study conducted by Bayark et al, assessed microtensile bond strength where all the samples were bonded with self-etch Single Bond adhesive (3M ESPE).
    4) Primary teeth affected with Hypocalcified AI were examined for shear bond strength. Teeth were etched only with 20% of phosphoric acid (Heraus Kulzer, Germany) and two layers of etch and rinse adhesive of Gluma One Bond (Heraus Kulzer, Germany) were applied to samples (Şaroğlu et al., 2006).

In the clinical studies, one study carried out by Sönmez et al. on clinical success of deproteinization on hypocalcified AI affected teeth. Similar to the bonding protocol done by Şaroğlu et al the teeth were also etched only with a percentage of 20 of phosphoric acid (Heraus Kulzer, Germany) and two layers of etch and rinse adhesive of Gluma One Bond (Heraus Kulzer, Germany) were applied to samples. (Sönmez et al., 2009)

Epasphinge et al evaluated bond strength of self-etch adhesives to dentine with Clearfil TM SE Bond on the presence and absence of the etching step, found bond strength to AI affected teeth significantly lower than the normal (Clearfil TM SE Bond 19.27 MPa to AI affected teeth and 36.16 MPa to sound dentine). Extra phosphoric acid etchant substantially reduced the bond strength of Clearfil TM SE Bond to sound teeth and further etching step did not improved the bond strength of Clearfil TM SE Bond to AI affected teeth.

FARIA-e-SILVA et al assessed the two steps, etch and rinse adhesive with Adoper Single Bond 2 (3M ESPE) between AI affected enamel and dentine, bond strength of dentine (24.6 MPa) higher than the bond to enamel (14.2 MPa).

Yaman et al reported that the most common mode of failure was adhesive failure at the resin / enamel interface of approximately 35.7% of the ER-HPAI and 28.4% of the SE-HPAI and mixed failures with partially cohesive failure in the dentine and resin adhesive. Also, Epasphinge et al found in his research a mixed mode of resin adhesive and dentine failures. In both studies, no association between mode of failure and the type of adhesive was reported.

3.3.5. Deproteinization

Enamel deproteinization with sodium hypochlorite was first proposed in 1994 as a case study by Venezie et al. to enhance bonding of orthodontic brackets to hypocalcified AI affected teeth. Sodium hypochlorite is non-specific

proteolytic agent which is efficient in eliminating organic compounds without damaging tooth structure (Venezie et al., 1994, Mohammadi, 2008). The following studies looked at the enamel and dentine deproteinization according to the type of amelogenesis imperfect affected teeth. In another laboratory study, Ahmed et al showed significant improvement of etching patterns needed for good resin bonding, after using 5.25% NaOCl for 60s prior to acid etching (37% phosphoric acid gel).

Sonmez et al evaluated the effect of clinical deproteinization on Hypocalcified AI permanent teeth with application of 5 percent sodium hypochlorite after a minute of acid conditioning, and after 36 months of follow up, had no significant impact on the effectiveness of adhesive restorations.
Similarly, Faria E Silva et al reported that 5% sodium Hypochlorite prior for adhesive procedure on permanent HCAI teeth did not enhance bond strength of enamel and dentine for control and treatment sample groups.

- Hypomaturation AI:
  A part of his study of shear bond strength, Chougule et al measured the SBS of bonded bracket after deproteinized teeth surfaces with 5% NaOCl. Teeth were acid etched with 37% orthophosphoric acid prior and after the application of sodium hypochlorite. The mean shear bond strength value of hypomature AI teeth shown an increase SBS from (5.48 MPa of conventional bonding group) to (6.65 MPa of NaOCl group).

- Hypocalcified AI:
  a. Şaroğlu et al showed that application of 5% NaOCl for1 minute after the application of acid conditioning in flat surface of 3mm in diameter was prepared on primary teeth, enamel shear bondstrength was significantly enhanced in HCAI teeth about 27.36 MPaversus 13.92 MPa in the control group of HCAI. Furthermore,deproteinization had no significant effect on shear bond strength ofdentine in both the HCAI and the control group of primary teeth.
  b. In another study, Bayark et al compare dentine deproteinization effects with sodium hypochlorite and chlorine dioxide, showed that

deproteinization with sodium hypochlorite (NaOCl) did not significantly affect micro-tensile bond strength of dentine in both the HCPAI and sound primary teeth compared to deproteinization with chlorine dioxide (ClO2) has significantly improved bond strength of both HCPAI and Sound teeth (NaOCl 13.12NaOCl 13.12 MPa vs ClO2 15.51 MPa in comparison to the tensile bond strength of control group of hypocalcified primary teeth 12.2 MPa.).

c. Deproteinization with 5% sodium hypochlorite prior to etching the surface on permanent hypocalcified AI, did not show any influence on enhancement of bond strength to enamel or dentin of sound or AI affected teeth (FARIA-e-SILVA et al., 2011).

d. In another laboratory study, Ahmed et al showed examined primary molars affected with hypocalcified AI to investigate the etching patterns after deproteinization. Teeth were pre-treated using 5.25% NaOCl for 60s prior to acid etching (37% phosphoric acid gel) and his findings concluded that the etching patterns which is needed for good resin bonding were significantly improved.

e. Sonmez et al evaluated the effect of clinical deproteinization on Hypocalcified AI permanent teeth with application of 5% percent sodium hypochlorite after a minute of acid conditioning, had no significant impact on the effectiveness of adhesive restorations.

3.3.6. Etching patterns

Three studies have evaluated etching patterns of AI affected teeth under scanning electron microscopy (SEM). Seow et al examined five types of AI under SEM pitted and smooth hypoplastic, male and female x-linked form and hypomineralized in primary and permanent AI affected teeth with of 37% phosphoric acid application for one minute.

In sound teeth, type 2 and 3 etching patterns were observed. The three types of etching patterns were found to be distributed similarly in the hypomineralized form of AI.

- Hypoplastic AI:
    - Yaman et al investigated variations in morphology of enamel surfaces treated with phosphoric acid etchant or self-etching primer. Application of 35% orthophosphoric acid for 30 seconds to sound and HPAI affected teeth, resulting in a suitable etching pattern compared to the SE adhesive system, which produced shallow groves that demarcates incremental enamel growth and very small diameter like pits of prism core. No typical etching pattern was visible on HPAI enamel. The etched enamel surface often appeared to be coated with a fibrous layer. In contrast, 20 seconds of etched enamel surface with SE primer revealed a less unique pattern which created a very minor etching effect on HPAI, with most of the surface remaining unetched.

- Hypoplastic / Hypomineralized AI:

    In examination of five types of AI affected teeth under SEM, pitted and smooth hypoplastic, male and female x-linked form and hypomineralized in primary and permanent AI affected teeth with of 37% phosphoric acid application for one minute. In case of AI affected teeth, pitted hypoplastic form showed predominance of type 1 with preferential removal of the prism cores. In comparison, the main etching pattern in the x-linked (female) was type 2 in which the peripheries of the prims were dissolved. The etched enamel in the x-linked (male) variant displayed a type 3 pattern in which the prism dissolution pattern was irregular and did not appear to be connected to the prism structure. In sound teeth, type 2 and 3 etching patterns were observed. The three types of etching patterns were found to be distributed similarly in the hypomineralized form of AI affected teeth in the study conducted by Seow et al.

- Hypocalcified AI:

  Ahmed et al findings were consistent with Seow and Amaratungo, comparing the etching patterns of hypocalcified primary AI molars into two groups: control samples were etched with 37% phosphoric acid for 15 seconds and the study samples were pre-treated with 5.25% of sodium hypochlorite (NaOCI) for 60 seconds prior to acid etching. Under Scanning Electron microscopy (SEM) the two groups were analyzed independently. The etched hypocalcified enamel surfaces (control group) showed three distinct etching patterns, mainly type III etching (65.63%) followed by types I and II. On the contrary, the pre-treated hypocalcified surfaces (study group) showed significant surface roughness with type I and type II predominance (82.5%) etching patterns which is required for good adhesion.

3.3.7. Mode of failure

Analysis of fracture patterns and microscopic observation using scanning electron microscope (SEM) on fractured beams according to the type of AI affected teeth:

- Hypoplastic AI:

  The mode of failure in the hypoplastic enamel mostly was due to adhesive failure between the interface of the resin and the enamel approximately 35.7% of the ER-HPAI and 28.4% of the SE-HPAI. Also mixed with partially cohesive failure exist between the dentin and resin (Yaman et al., 2014).

- Hypocalcified AI:
  a. The failure mode was primarily found to be adhesive for both hypocalcified AI and sound primary teeth (Bayrak et al., 2019).
  b. The mode of failure of all the samples was categorized as cohesive failure within the resin adhesive, adhesive failure

between the resin / dentin, dentin cohesive failure and a mixed failure (Epasinghe and Yiu, 2018).

3.3.8. Fluoride

One study has evaluated the effects of 2% of sodium fluoride treatment in hypomaturation AI affected teeth. Application of fluoride to the surface of enamel of affected teeth can recover the mineral lost during the anomaly formation by facilitating remineralization and decreasing the solubility of tooth enamel while which will be providing etching patterns appropriate for composite placement. Chougule et al observed significantly greater shear bond strength when bonding orthodontic brackets to AI affected teeth after the application of 2% sodium fluoride (NaF) for four minutes prior to acid etching of tooth surface (Chougule et al., 2018). Where the SBS of hypomature AI teeth treated with sodium fluoride demonstrated a higher value of bond strength at 7.65 MPa in comparison to the conventional bonding group and sodium hypochlorite group at 5.48 MPa, 6.65 MPa respectively.

3.3.9. Resin Infiltration and sealant

None of the laboratory studies investigated Icon infiltration and its effects on the bond strength and failure pattern when NaOCl was applied after or before etching and prior for Icon application. Also, sealant bonded to AI affected teeth in comparison to those bonded to sound teeth and the quality of NaOCl when added to AI affected teeth before or after etching and their retention rate. Chay et al. found that when using Icon, the bond strength results were highly variable, with substantial standard deviations, presumably indicating inconsistent penetration in hypomineralized enamel such as Molar Incisor Hypomineralization cases. He also reported that etching with NaOCl before Icon penetration increased bond strength much more than Icon without NaOCl (Chay et al., 2014). Studies investigating both techniques in Molar - Incisor Hypomineralization (MIH) were not included in this review as part of the inclusion/ exclusion criteria.

3.4. Discussion

3.4.1. The disparity between studies

There were differences in tooth storage media used. This included deionized water and thymol for two studies each, sodium chloride, chloramine, distilled water and even teeth kept dry with patients. All the researchers had no reason for their decision. These solutions may alter or interfere with the enamel microstructure, for instance deionized water as aqueous storage media can cause tooth surface demineralization which has been recorded (Armstrong et al., 2017). The storage temperature and the time of immersion was not always specified which may influence the microstructure of the enamel.

Some of the studies, examined primary AI affected teeth and other have used permanent AI affected teeth. Primary molar teeth appear to be less affected (Gjørup et al., 2009), therefore it may not be possible to extrapolate results from primary teeth to permanent and vice versa. A study was conducted to investigate an AI family member of five generations.

According to the studies the enamel surface preparation was also variable. The enamel surface was ground from 180 to 1200 grits in the lab. This can include the end, or the full length of the enamel prism rods: these two parameters might affect bond strength qualities.

The magnitude of the severity of the Amelogenesis Imperfecta may also affect the strength of the bond or the durability of restorative treatment. Most of the studies did not report the severity. This could be because the severity of the AI condition is not well defined and specified. Lundgren and Dahllöf, 2014, described it as mild cases where changes in enamel mineralization affected less than 1/3 of the tooth surfaces and the teeth had normal sensitivity. In moderate cases where enamel mineralization about 1/3 to 2/3 of tooth surfaces and teeth had mild

sensitivity. In the severe form of AI cases teeth more than 2/3 of the tooth surfaces were affected and exhibited high sensitivity. It was characterized by some authors according to the discolorations as yellow to brown with or without enamel breakdown and cases manifesting physiological root resorption.

Regarding materials used and application were not always the same from one study to the other. Self-etch adhesives can exhibit different pH values and therefore have different effects on enamel. Similarly, according to the studies the concentration of NaOCl and the point of its application differed.

According to the laboratory reports, the shearing methods were not the same: 3 studies carried out micro-shear bond strength tests and other three studies evaluated microtensile bond strength tests. Therefore, a meta-analysis of the bond strength values was not feasible.

3.4.2. Bonding to AI affected teeth vs sound enamel

All the studies clearly highlighted lower bond strength values to AI affected teeth. This can be due to differences in morphology and micro morphology compared to normal enamel, irregular etching patterns, decreased micro-tags in prism rod, less dense or incompletely formed enamel prism that retains moisture, and increased protein content, as well as decreased mineral content.

The linear association between enamel hardness and bond strength was observed, and the strength of the dentin bond was higher than that of the enamel bond. However, the strength of the bond to normal dentin was significantly greater than that of normal enamel (FARIA-e-SILVA et al., 2011). Dentin calcium levels in teeth affected by AI are higher than normal dentin levels. In addition, AI affected dentin is distinguished by peritubular dentin thickening and partial dentin tubules obliteration. The boundaries between inter-tubular and peritubular dentin cannot be

easily defined. This morphological pattern, therefore, corresponds to sclerotic dentin, which is likely to be more resistant to demineralization by acid etching causing AI affected dentin with lower bond strength (Bayrak et al., 2019, Sánchez-Quevedo et al., 2004).

3.4.3. Is there an adhesive of preference?

Yaman et al., 2014, the self-etch adhesive system exhibited lower bond strength levels to sound healthy enamel than etch and rinse adhesive, however, no statistical significant difference was detected between the bond strength measures obtained from the two adhesive systems in hypocalcified AI.

In his study, he also suggested that due to low inter-crystal porosity, self-etch adhesive can form insufficient micro tags and that phosphoric acid etching in ER adhesive can increase the mineral loss than the primer in the SE adhesive which could not demineralize the hypomineralized layer, which explains the relatively higher ER bond strength values by more micro retentive surface.

Nevertheless, acidic monomers are integrated with a priming agent in the primer of SE adhesive systems, allowing these monomers to penetrate to the same depth at which demineralization exists. As microporosities produced by the adhesive and provide micro mechanical interlocking with enamel, this is considered as an advantage.

In contrast with Epasinghe et al, has shown that additional etching with phosphoric acid did not enhance the bonding of a self-etch adhesive to AI affected dentine.

Interestingly, none of the clinical trials have investigated the variations in the success rate of a self-etch adhesive composite relative to composite bonded with an etch and rinse adhesive.

The result of this review indicates that when the option is between SE and ER adhesives, the clinical performance of composite resin restorations bonded to HCAI affected enamel may not be affected by the bonding agent type. However, among the different types of AI, enamel structure of AI affected teeth varies. Since the bonding ability of current adhesive resin materials can be influenced by these micro morphological differences, generalizations of findings are difficult. More studies are needed, before clinical advice can be given to evaluate and determine the best bonding protocols for different types of AI affected teeth.

3.4.4. Is deproteinization effective for bond strength enhancement?

The high protein content of AI affected enamel have been attributed to the increased failure rates in resin bonding and reduced the micromechanical adhesion. Therefore, in order to achieve optimal adhesion, it would be necessary to withdraw the excess content of protein organic matter and acquired pellicle prior to acid etching, the application of NaOCl was suggested as a possible strategy (Şaroğlu et al., 2006, Harleen et al., 2011, Espinosa et al., 2008). In endodontic, oxidizing NaOCl is already used to dissolve organic material (Moorer and Wesselink, 1982).

A substantial increase in bond strength to AI affected enamel was reported by Şaroğlu et al., however deproteinization did not influence dentine shear bond strength in both AI affected teeth and normal, when NaOCl was applied after etching for 60 seconds in primary teeth. Ahmed et al., also reported significant increase in etching patterns of type I and II in pretreated enamel surfaces prior to acid etching in primary molars which is essential for optimal resin bonding. Whereas

clinical, Sönmez et al and Faria – e – Silva et al found no increase in bond strength. However, Bayark et al., found no difference in bond strength with sodium hypochlorite (NaOCI) and significantly higher bond strength when the enamel surface was pretreated with chlorine dioxide (CIO2).

The time of deproteinization, before or after etching, might be very crucial. Şaroğlu et al suggested that enamel was rectified by etching and thereby facilitate protein degradation by NaOCI, allowing the adhesive to penetrate easily to enamel crystals, thus increasing the strength of the bond. Where he reported enhanced enamel bond strength but did not affect shear bond strength of dentin. On the other hand, Bayark et al and clinically Sönmez et al showed no difference in the enamel bond strength when using NaOCI after etching. Furthermore, Bayark et al observed significant increase in bond strength when using chlorine dioxide after etching. In addition, Faria – e – Silva et al reported when NaOCI was used, microhardness and shear bond strength to enamel affected by amelogenesis imperfecta did not increase, when using NaOCI before etching.

A few studies that have explored the effects of sodium fluoride application for 4 minutes before etching of enamel which thought it could replace the loss of minerals in enamel while creating suitable etching patterns appropriate for the placement of composite. This will allow good infiltration of bonding agents into the etched, fluoride-treated surface of demineralized enamel that might contribute to an increase in shear bond strength (Schmidlin et al., 2004, Chougule et al., 2018).

3.4.5. Quality assessment

Among the 12 studies included, 7 showed a low risk of bias and other 5 studies exhibited a medium risk (fair quality). The sample size calculation has been assessed in most laboratory experiments (n=7). The

primary goal of power analysis is to assist researchers in determining the smallest sample size necessary to detect the impact of a particular test at the acceptable level of significance, and as such it is critical (Cohen, 1992). In terms of blinding the test operator, only two studies were found adequate with blinding of the outcome assessment, and this could be due to the inability to blind the type of adhesive for instance.

3.4.6. Future research explore

Future research could target the bond strength of alternative materials to AI affected teeth in addition to composite, for example, glass ionomer cement (GIC) or compomers. Other experiments should investigate the use of ethanol wet bonding in AI affected teeth to reduce the water content. In general, the different types of AI need to be investigated. Further investigation of fluoride also merits attention. Finally, further clinical trials should be conducted to verify the findings of laboratory research.

3.5. Conclusion

This review highlights the necessity to enhance bonding to AI affected teeth. There are currently very limited resources of what can be done to solve and enhance adhesion to amelogenesis imperfecta affected teeth. Within the limitation of this review, bond strength to AI affected teeth did not vary significantly when using self-etch (SE) in comparison to etch and rinse (ER) adhesives.

Deproteinization with sodium hypochlorite does not enhance bond strength and efficacy of chlorine dioxide requires more studies. Use of sodium fluoride prior to etching could enhance bond strength of adhesive. Clinical and laboratory studies should be carried out on the effectiveness of ICON infiltration to enhance bond strength to AI affected teeth. Given the limited number of studies involved, and the variations in the type and severity of AI and the adhesives used, these findings should be considered with caution. In order to achieve improved bonding with AI affected teeth, more research is needed.

One of the problems was the different classifications used by each study in this review, therefore it was important to study the standardization and different classifications quoted in the literature in the recent years. The focus on the last part of this project was a systematic review of classification systems used in AI.

Finally clinical recommendation within the scope of this review and with very limited evidence of what can be accomplished to solve the difficulty of adhesion to AI affected teeth. Clinical deproteinization with either the 5% of sodium hypochlorite for primary teeth prior to etching step or the use of chloride dioxide for both primary and permanent teeth. Also, pretreatment of enamel surface with 2% of sodium fluoride for 4 minutes can be used solely or in combination with the previous step to provide suitable etching patterns so, that the adhesive agents can infiltrate into the expose collagen mesh and in addition it will replace the lost minerals which will improve bond strength. Those

techniques to be used under full isolation such as rubber dam for successful long-term adhesion and contaminate free surfaces regardless the type of adhesion system whether using self-etch or etch and rinse.

4. Diagnostic classification of Amelogenesis Imperfecta: A review

Introduction

There have been many classifications used since 1945 for AI. In 1971, Witkop proposed a clinical classification primarily based on three clinical presentations (phenotype) as Hypoplastic, Hypocalcified and Hypomaturation. Within these three classifications there were subgroups among these to represent the different modes of inheritance for example Autosomal dominant pitted hypoplastic and x-linked recessive hypomaturation (Aldred et al., 2003).

As these clinical conditions with genetic causes can have a wide range of manifestations, a new classification was presented by Witkop in 1988, with more subgroups equal to fifteen with the main three clinical phenotypes still exists, another fourth clinical presentation has been added hypomaturation – hypoplastic with taurodontism (Witkop Jr, 1988).

As the understanding of genetics has increased, a new alternativeclassification of AI was proposed that does not use the phenotype as the primary feature. The classification was based on the following (when known) molecular defect, biochemical result, also mode of inheritance and finally on the phenotype. The awareness that phenotype may vary considerably, as wellas growing genomic knowledge currently accessible, led to this categorization(Aldred et al., 2003).

Investigating the molecular genetics, will aid for more precise diagnosis which might also be extended and applied to other dental anomalies (Aldred et al., 2003). The four main areas of this classification:

1. Mode of Inheritance: autosomal dominant, autosomal recessive, x-linked, isolated case.
2. Molecular basis: chromosomal, localization, locus, mutation (when known).
3. Biochemical outcome: putative result of mutation (when known).
   Phenotype: description of clinical and / or radiographic findings and other relevant if any. Those are hypoplastic, hypocalcified, hypomaturation, hypomaturation-hypoplastic with taurdontism.

When carrying out the review for the previous section of this thesis the authors noticed a wide variety of classification systems were used and it did not appear that they were always applied correctly. When the COVID pandemic occurred, meaning that we could not carry out the lab experiments initially proposed in this study, we decided to review the use of AI classification systems instead.

4.1. Study Aim

A classification system of Amelogenesis Imperfecta (AI) was first proposed by Weinmann et al, in 1945, and with better understanding of genetics of AI with different genotype / phenotype presentations, various classification have been developed, with the latest by Aldred et al in 2003. The purpose of this review is to analyze AI studies to provide evidence for how many different classifications are quoted in the literature, the consistency and standardization of clinicians and researchers reporting on AI classifications.

4.2. Material and Methods

4.2.1. Inclusion and Exclusion Criteria

All studies (research papers and reviews) with any proposed classification system on the diagnosis of AI in the primary and permanent dentition including case reports and narrative reviews. There was no attempt to specify the strategy in relation to study design or language published in the period 2015 onwards up to date. 2015 was used as a cut-off point to limit studies for pragmatic purposes and because we were interested in more recent use of the full range of systems available. Exclusion criteria as follows: any studies on animals, studies on genetic and inheritance patterns, psychological impact of AI, syndromes and medicine, other aspects of dental anomalies.

4.2.1. Data collection and analysis

The data extracted by (HA, SP, PA) included study design, the diagnostic classification used, was it correctly used by comparing to the original classification, description of each type of AI, detected by physical examination (who examined). The data were summarized in Table 4-2. Any potential conflict was resolved by discussion between the authors.

4.2.1. Search strategy

A comprehensive search was done using search keywords in both electronic, hand search journals and websites including Medline (Ovid) PubMed, the Cochrane library and Goggle scholar. Keywords linked to Amelogenesis Imperfecta were used to create the search method. The following is a breakdown of the method: hypoplastic, hypomature, hypocalcified, hypomineralized, hypomineralised, enamel defect. The searched articles were ranged in date from 2015 to the present. The reference list of the studies that were originally collected was reviewed by hand for publications that could be relevant. The search was conducted between January to March 2021.

4.3.Results

4.3.1. Study selection

The initial search yielded 230 studies, and following screening of the title, abstract and full text a total of 33 studies were eventually included. Figure 4-1 summarizes the search process.

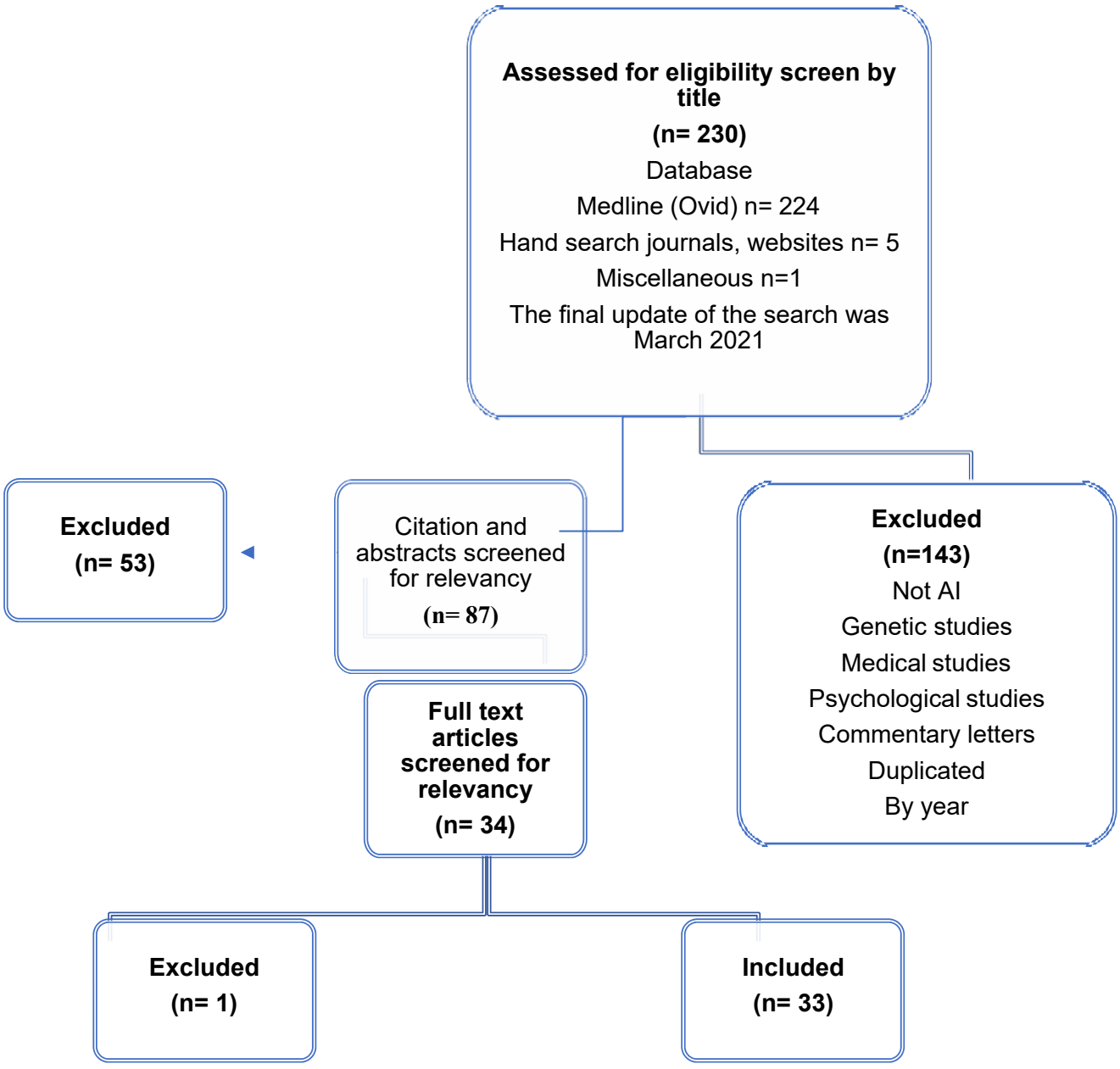


Figure 4-1 Flow chart describing the search strategy

Table 4-1 results of included studies

Author / year	Type of study	Diagnostic classification	Was it correctly used?	Description of AI types	Diagnosis: who examined?)
(Gabardo et al., 2020)	Case control study	Aldred 2003	Phenotype only	Yes	Not specified
(Quandalle et al., 2020)	Case – control study	Witkop classification 1988	Yes	Yes	Not specified
(Sabandal et al., 2020)	Case report	Witkop classification 1988	Yes	Yes	Central Interdisciplinary Ambulance in the School of Dentistry, University of Munster but not specified
(Adorno-Farias et al., 2019)	Retrospective study	Witkop classification 1988	Yes	Yes	Not specified
(Ahmed et al., 2019)	Laboratory study	Witkop and Sauk 1976	Yes	Yes	Department of Paediatric Dentistry
(Kammoun et al., 2019)	Cross sectional study	Aldred classification 2003	No description if known either of mode of inheritance, molecular basis or biochemical	AI of hypoplastic type was diagnosed when dental development anomaly was showing a thin or an absence of enamel	Prosthetics Department but not specified
(Kirzioglu et al., 2019)	Case control study	Not described	-	-	Department of Paediatric Dentistry
(Mori et al., 2019)	Case report	Not described	-	-	Not specified
(Moussally et al., 2019)	Case report	Not described	-	No	Not specified

(Ortiz et al., 2019)	Clinical report	Aldred 2003	Yes, with phenotype only no description if mode of inheritance, molecular or biochemical were known	No - hypomature AI include abnormally rough and pitted tooth surfaces and discoloration of teeth with hypersensitivity	Not specified
(Epasinghe and Yiu, 2018)	Laboratory study	Not described	-	Yes	Not specified
(Kammoun et al., 2018)	Laboratory study	Aldred 2003	Yes, by phenotype only	Yes	Not specified
(Klink et al., 2018)	Case series	Not described	-	-	Department of Prosthodontics but not specified
(Lundgren et al., 2018)	Randomized control trial	Sundell and Koch	Yes	Yes	Department of Paediatric Dentistry
(Singh et al., 2018)	Case series	Witkop and Sauk 1976	Yes, with phenotype but no description regarding mode of inheritance if known	Yes	Not specified
(Strauch and Hahnel, 2018)	A review	Aldred 2003	Yes	Yes	-
(Toupenay et al., 2018)	Review of case reports	Witkop classification 1988	Yes	Yes	Not specified
(Cagetti et al., 2017)	Report of two cases	Aldred 2003	Yes, based on phenotype only	Yes	Not specified
(Güth et al., 2017)	Case report	Not described	-	No	Department of Prosthodontics
(Leevailoj et al., 2017)	Case study	Not described	-	Definitions of basic types of AI	Department of Oral Medicine
(Belcheva et al., 2016)	Laboratory study	Witkop classification 1988	Phenotype only	Not described	Not specified

(Bogosavljević et al., 2016)	Case report	Witkop classification 1988	Phenotype only	Yes	Departments of Paediatric Dentistry
(Dursun et al., 2016)	Case report	Not described	-	Yes	Not specified
(Moreira et al., 2016)	Case report	Aldred classification 2003	Yes	Yes	Not specified
(Pousette Lundgren et al., 2016)	An interview study	Not described	-	Yes	Not specified
(Rogers et al., 2016)	Case report	Not described	-	-	paediatric dental clinic
(Sabandal and Schäfer, 2016)	A review	Witkop classification 1988	Yes	Yes	-
(Shibata et al., 2016)	Case report	Aldred classification 2003	yes	yes	Not specified
(Zimmermann et al., 2016)	Case report	Not described	-	The diagnosis was a severe form of amelogenesis imperfecta type II	Restorative and periodontology department but not specified
(Bhatia et al., 2015)	Report of three cases	Aldred 2003	Yes, with mode of inheritance	Yes	Department of Paediatric Dentistry
(Gerdolle et al., 2015)	Case report	Witkop classification 1988	Yes	Yes	Not specified
(Marquezin et al., 2015)	Case report	Witkop classification 1988	Yes	Yes	Department of Paediatric Dentistry
(Nigam et al., 2015)	Case report	Not described	Phenotype only	Yes	Department of Oral Pathology,

4.3.2. Diagnostic classification of Amelogenesis Imperfecta

In this review, four main classifications were identified and quoted in the studies. The majority did not use or mention any diagnostic classification (n=12, 36%).

The remainder used or cited both Witkop 1988 and Aldred 2003 classifications (n=9, 27%) each, Witkop and Saulk 1976 (n=2, 6%), Sundell and Koch 1985 (n=1, 3%) (Figure 4-2).

Most of the studies which used the diagnostic classification of AI have used it correctly when compared to the original classification but of these, 9 of them only focused on the phenotype with no description of genetic inheritance.

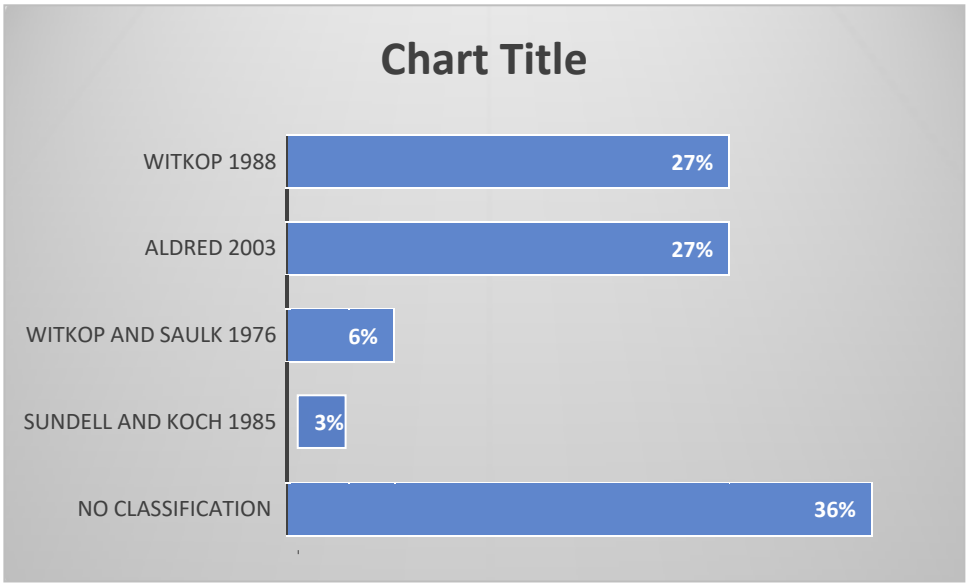


Figure 4-2 Distribution of diagnostic classification systems of revised studies

4.4.Discussion

One of the most notable and relevant concerns related to AI are the different diagnostic classifications, making comparisons between studies difficult. Because of the discrepancy of classifications, any effort to evaluate delivery of care by AI type would have been futile. It also emphasizes how challenging it to distinguish AI types based on clinical phenotypes alone. Nonetheless, accurate diagnosis is crucial in deciding treatment and prognosis. The growing field of AI genetic testing will help with classification or in the treatment and service development (McDowall et al., 2018).

In this study, n=12, 36% of the papers did not use or cite a classification and 43% only described the phenotype with no information regarding the basic genetic information if known, which might be due to the different existing classifications being difficult to grasp. This coincided with Aldred et al. findings of the muddled and inconsistent usage of Arabic and Roman numbers or their absence in some of the classifications.

Where there is known to be significant variations in the clinical appearance, the phenotype alone does not seem to support allocation to diagnostic classification as this study showed n=9 out of 21 studies who used diagnostic classification reported phenotype only. Therefore, a clear guidance for the basic genetic information for clinicians to allow them for a better description of classification is crucial.

As this review showed clinical phenotype has been used to classify AI. This strategy is hampered by three major problems. Some AI types do not fit within this category. Enamel changes after eruption might be visible, making precise AI classification challenging. Some types of AI are linked to other medical conditions, even for single gene variations, they can manifest in a wide range of clinical features (Ratbi et al., 2015). The diagnosis of AI predates the renal calcification by several years in condition such as nephrocalcinosis and AI, allowing patients to be identified by dentists considerably sooner than currently

(de la Dure-Molla et al., 2014). It has been realized that genetic knowledge has the potential to improve management of AI. There are additional benefits to accurate diagnosis, making clinical decision, determine prognosis, genetic counselling, and the expansion of evidence-based research (McDowall et al., 2018). Precise diagnosis permits prompt genetic counselling, and preventative measures can be done to prevent subsequent dental problems for the individual or even future siblings (Shivhare et al., 2016). The classification proposed by (Aldred et al., 2003) was a more comprehensive as it did not depend on the phenotype as the primary discriminator. A clinical AI index for clinicians to standardize AI classification and scoring for AI severity is needed.

4.5.Conclusion

This review showed the variations and inconsistency of classification used for studies published from 2015. It has highlighted that about 36% of studies did not used any of the existing classifications. The classification proposed by Witkop 1988, and Aldred 2003 were the most commonly used (27% respectively).

Studies have described classification according to its phenotype only with no other information on the underlying genetic if it was known or not. As a result, detailed guidance for clinicians on fundamental genetic information is critical for a clearer explanation of classification hence it is a hereditary disorder.

5. Future work

5.1. Future work

Clinicians have experienced challenges bonding to restorative material, because of the bonding strength and etching limitations to the enamel. Detailed laboratory study regarding the properties of bonding materials between the enamel and restoration may aid the dentists in selection the optimum material to manage these teeth. Also, to compare both the primary and permanent teeth together with more emphasis to study the permanent teeth affected with AI because the clinical description of primary teeth could be similar to that of permanent teeth, however, these teeth appear to be less affected. Also, the ideal storage media and temperature for the samples and to include the full length of the enamel prism rods when the enamel surface is grounded as it might affect the bond strength qualities. By conducting microtensile bond strength testing using universal testing machine and micro-shear bond strength using a mechanical testing machine.

Future research could target the bond strength of alternative materials to AI affected teeth in addition to composite, for example, glass ionomer cement (GIC) or compomers. Other experiments should investigate the use of ethanol wet bonding in AI affected teeth to reduce the water content examined under scanning electron microscopy and microtensile bond strength test. Finally, further clinical trials should be conducted to verify the findings of laboratory research.

Clinical and laboratory studies should be carried out on the effectiveness of ICON infiltration to enhance bond strength to AI affected teeth by application of ICON before bonding process and then evaluated for shear bond strength with a universal testing machine. Also, the effects of deproteinization of both sodium hypochlorite and chlorine dioxide comparing before and after etching step with microtensile and shear bond strength tests.

Further studies to test the effectiveness of sodium fluoride to enhance bonding strength by application of fluoride before acid etching with different bonding systems, sodium hypochlorite in one group and chlorine dioxide in the other

group for comparison. In addition, test the effects of fluoride application before etching and sodium hypochlorite after etching on the same sample. A shear bond strength to be measured using universal testing machine.

Detailed simplified guidance for clinicians on fundamental genetic information is critical for a clearer explanation of classification. A clinical AI index for clinicians to allow easier application in pragmatic approach and standardize of the classification which can include clear definition and scoring for AI severity as well is needed. The severity of AI may have an impact on the bonding strength or the long-term effectiveness of the treatment, therefore, a clear definition and scoring for AI severity is crucial.

In summary, further clinical and laboratory studies are needed to improve the bond strength of restorative materials, as well as quality of care and treatment provided to this group of patients.

6. Conclusion

6.1. Conclusion

Patients with AI may have increased sensitivity, poor oral hygiene, and rapid tooth loss, along with along with abnormalities in enamel thickness, colour and form, all of which can affect their appearance, masticatory ability, and oral health quality of life.

AI is a rare condition, so clinical trials are difficult to conduct due to the small number of patients and so to understand and help this group of patients we planned a pragmatic approach which involved a literature review and retrospective study of burden of care, Originally it was also planned to undertake laboratory studies to investigate the bonding of different AI types, however, due to the COVID19 pandemic, it was not possible to conduct the lab tests to complete this project.

The service evaluation provides data on the burden of care for children with AI. Most patients are treated under general or local anesthetics, imposing a considerable treatment burden on patients and their families. The long duration of treatment it involves, the high number of appointments needed every year and the long-distance travelled by the families to attend a single appointment in this study illustrates the wide range of problems that might arise, as well as the complex management that is necessary for this condition.

This study has shown the need for early referrals of patients with a suspected diagnosis of AI in the primary dentition, with late presentation, extensive treatments are needed frequently so teeth are extracted, or full crown coverage provided. Therefore, it would be advantageous if primary care providers were more aware of referral options. For correct diagnosis and comparison of AI, all patients should be investigated about their family pedigree and documented. A defined discharge pathway is also required to be improved to facilitate the treatment needed during adulthood.

AI will continue to affect the aesthetics, functional and life quality issues for these individuals, demanding further interventions. There is no defined

standard of care set for AI patients management even though a multi-disciplinary approach can be beneficial. Considerable time and effect expenditure have been demonstrated. The impact of care provides a base for the foundation of the advancement of care approaches and seek to enhance the management of AI.

In the systematic review showed the bonding strength and hardness to AI affected teeth were less than the normal teeth. The enamel of AI affected teeth had alteration in micromorphology disordered prisms, coated with an amorphous structureless layer of organic protein which influences etching to demineralize the enamel in AI and affects the values of bonding strength of adhesive systems. Different adhesive systems when comparing self-etch (SE) to etch and rinse (ER) did not differ substantially in bond strength to AI affected teeth.

Deproteinization with 5% sodium hypochlorite did not enhance bond strength but the use of 2% sodium fluoride before etching for 4 minutes could enhance the bond strength of the adhesive. The efficacy of chlorine dioxide requires more studies which could be an alternative option. This review showed the need for ongoing investigations of the effects of AI on teeth may aid in the development of longer lasting bonded restorations.

The review of classifications of AI showed the variations and inconsistency of classifications used for studies published from 2015. It has highlighted that about 36% of studies have not used any of the existing classification. Studies have shown classifications based on phenotype, with no information on whether the underlying genetic was known. More awareness is needed for the dentists to allow better description of both the mode of inheritance and the clinical phenotype of AI, as well as a standard form when investigating patients suspected of AI diagnosis.

7. References

ADORNO-FARIAS, D., ORTEGA-PINTO, A., GAJARDO, P., SALAZAR, A., MORALES-BOZO, I., WERLINGER, F., ROJAS-FLORES, S., MOLINA-BERRÍOS, A., ECHEVERRÍA-LÓPEZ, S. & JARA-SANDOVAL, J. 2019. Diversity of clinical, radiographic and genealogical findings in 41 families with amelogenesis imperfecta. *Journal of Applied Oral Science,* 27.

AHMED, A. M., NAGY, D. & ELKATEB, M. A. 2019. Etching Patterns of Sodium Hypochlorite Pretreated Hypocalcified Amelogenesis Imperfecta Primary Molars: SEM Study. *Journal of Clinical Pediatric Dentistry,* 43, 257-262.

AKINMADE, A. & NICHOLSON, J. 1993. Glass-ionomer cements as adhesives. *Journal of Materials Science: Materials in Medicine,* 4, 95-101.

AL-OMARI, W., MITCHELL, C. & CUNNINGHAM, J. 2001. Surface roughness and wettability of enamel and dentine surfaces prepared with different dental burs. *Journal of Oral Rehabilitation,* 28, 645-650.

ALDRED, M. & CRAWFORD, P. 1995. Amelogenesis imperfecta—towards a new classification. Wiley Online Library.

ALDRED, M., SAVARIRAYAN, R. & CRAWFORD, P. 2003. Amelogenesis imperfecta: a classification and catalogue for the 21st century. *Oral diseases,* 9, 19-23.

ALLEN, K. 1993. Current theories of adhesion and their relevance to adhesive technology. *Le Journal de Physique IV,* 3, C7-1511-C7-1516.

AMBERGER, J. S., BOCCHINI, C. A., SCHIETTECATTE, F., SCOTT, A. F. & HAMOSH, A. 2015. OMIM. org: Online Mendelian Inheritance in Man (OMIM®), an online catalog of human genes and genetic disorders. *Nucleic Acids Research,* 43, D789-D798.

ANITHA, M., SATHVIKALAKSHMI, B., KHAN, A. F. & SUDHARSHAN, R. 2018. Amelogenesis Imperfecta–3

Cases. *Journal of Advances in Medical and Pharmaceutical Sciences*, 1-8.
ANUSAVICE, K. J., SHEN, C. & RAWLS, H. R. 2012. *Phillips' science of dental materials*, Elsevier Health Sciences.
ARMSTRONG, S., BRESCHI, L., ÖZCAN, M., PFEFFERKORN, F., FERRARI, M. & VAN MEERBEEK, B. 2017. Academy of Dental Materials guidance on in vitro testing of dental composite bonding effectiveness to dentin/enamel using micro-tensile bond strength (μTBS) approach. *Dental Materials,* 33, 133-143.
AYERS, K., DRUMMOND, B., HARDING, W., SALIS, S. & LISTON, P. 2004. Amelogenesis imperfecta-multidisciplinary management from eruption to adulthood. Review and case report. *NZ Dent J,* 100, 101-4.
BAHARAV, H., CARDASH, H., PILO, R. & HELFT, M. 1988. The efficacy of liquid and gel acid etchants. *Journal of Prosthetic Dentistry,* 60, 545-547.
BAIER, R. 1992. Principles of adhesion. *Operative Dentistry*, 1-9.
BAILLEUL-FORESTIER, I., MOLLA, M., VERLOES, A. & BERDAL, A. 2008. The genetic basis of inherited anomalies of the teeth: Part 1: Clinical and molecular aspects of non-syndromic dental disorders. *European Journal of Medical Genetics,* 51, 273-291.
BAJRAKTAROVA-VALJAKOVA, E., GROZDANOV, A., GUGUVCEVSKI, L., KORUNOSKA-STEVKOVSKA, V., KAPUSEVSKA, B., GIGOVSKI, N., MIJOSKA, A. & BAJRAKTAROVA-MISEVSKA, C. 2018. Acid etching as surface treatment method for luting of glass-ceramic restorations, part 1: acids, application protocol and etching effectiveness. *Open access Macedonian journal of medical sciences,* 6, 568.
BALIAN, A., MALERBA, A. & STROHMENGER, L. 2017. Amelogenesis Imperfecta in the Deciduous Dentition: Effectiveness of a Preventive Approach. *JOURNAL OF DENTAL HEALTH, ORAL DISORDERS & THERAPY,* 7, 1-2.

BAYRAK, S., TULOGLU, N. & TUNC, E. S. 2019. Effects of Deproteinization on Bond Strength of Composite to Primary Teeth Affected by Amelogenesis. *Pediatric Dentistry,* 41, 304-308.

BELCHEVA, A., PHILIPOV, I. & TOMOV, G. 2016. Scanning electron microscopy of enamel and dentin of teeth with hypocalcified amelogenesis imperfecta. *Folia Med (Plovdiv),* 58, 54-59.

BEN-AMAR, A., BAHARAV, H., LIBERMAN, R. & NORDENBERG, D. 1988. Continuous brushing acid-etch technique and microleakage of class V composite restorations. *Journal of Prosthetic Dentistry,* 59, 573-576.

BENAZZI, S., NGUYEN, H. N., KULLMER, O. & HUBLIN, J. J. 2015. Exploring the biomechanics of taurodontism. *Journal of Anatomy,* 226, 180-188.

BENIASH, E., STIFLER, C. A., SUN, C.-Y., JUNG, G. S., QIN, Z., BUEHLER, M. J. & GILBERT, P. U. 2019. The hidden structure of human enamel. *Nature communications,* 10, 1-13.

BERKMAN, M. & SINGER, A. 1971. Demonstration of the lyon hypothesis in X-linked dominant hypoplastic amelogenesis imperfecta. *Birth Defects Original Article Series,* 7, 204-209.

BERKOVITZ, B., HOLLAND, G., MOXHAM, B., HOLLAND, G. & MOXHAM, B. 2009. Oral anatomy, Histology and Embryology.: Mosby. *4th International edition,* 4.

BERKOVITZ, B. K., HOLLAND, G. R. & MOXHAM, B. J. 2017. *Oral Anatomy, Histology and Embryology E-Book*, Elsevier Health Sciences.

BHATIA, S. K., HUNTER, M. L. & ASHLEY, P. F. 2015. Amelogenesis imperfecta with coronal resorption: report of three cases. *Dental update,* 42, 945-950.

BOGOSAVLJEVIĆ, A., MIŠINA, V., JORDAČEVIĆ, J., ABAZOVIĆ, M., DUKIĆ, S., RISTIĆ, L. & DAKOVIĆ, D. 2016. Treatment of teeth in the esthetic zone in a patient with amelogenesis

imperfecta using composite veneers and the clear matrix technique: A case report. *Vojnosanitetski pregled,* 73, 288-292.

BRESCHI, L., MAZZONI, A. & FERRARI, M. 2008a. Adhesion to intra-radicular dentin. *Ferrari M with Breschi L, Grandini S. Fiber posts and endodontically treated teeth: a compendium of scientific and clinical perspectives. Wendywood: Modern Dentistry Media*, 15-37.

BRESCHI, L., MAZZONI, A., RUGGERI, A., CADENARO, M., DI LENARDA, R. & DORIGO, E. D. S. 2008b. Dental adhesion review: aging and stability of the bonded interface. *Dental Materials,* 24, 90-101.

BUONOCORE, M. G. 1955. A simple method of increasing the adhesion of acrylic filling materials to enamel surfaces. *Journal of Dental Research,* 34, 849-853.

BUTERA, A., MAIORANI, C., MORANDINI, A., SIMONINI, M., MORITTU, S., BARBIERI, S., BRUNI, A., SINESI, A., RICCI, M. & TROMBINI, J. 2021. Assessment of Genetical, Pre, Peri and Post Natal Risk Factors of Deciduous Molar Hypomineralization (DMH), Hypomineralized Second Primary Molar (HSPM) and Molar Incisor Hypomineralization (MIH): A Narrative Review. *Children,* 8, 432.

CAGETTI, M. G., CATTANEO, S., HU, Y. Q. & CAMPUS, G. 2017. Amelogenesis imperfecta: A non-invasive approach to improve esthetics in young patients. Report of two cases. *Journal of Clinical Pediatric Dentistry,* 41, 332-335.

CANGER, E. M., CELENK, P., YENISEY, M. & ODYAKMAZ, S. Z. 2010. Amelogenesis imperfecta, hypoplastic type associated with some dental abnormalities: a case report. *Brazilian Dental Journal,* 21, 170-174.

CHAN, H.-C., MAI, L., OIKONOMOPOULOU, A., CHAN, H., RICHARDSON, A., WANG, S.-K., SIMMER, J. & HU, J.-C. 2010. Altered enamelin phosphorylation site causes

amelogenesis imperfecta. *Journal of Dental Research,* 89, 695-699.

CHAN, H. C., ESTRELLA, N. M., MILKOVICH, R. N., KIM, J. W., SIMMER, J. P. & HU, J. C. C. 2011. Target gene analyses of 39 amelogenesis imperfecta kindreds. *European journal oforal sciences,* 119, 311-323.

CHAY, P. L., MANTON, D. J. & PALAMARA, J. E. 2014. The effect of resin infiltration and oxidative pre-treatment on microshear bond strength of resin composite to hypomineralised enamel. International journal of paediatric dentistry, 24, 252-267.

CHANMOUGANANDA, S. C., ASHOKAN, K. A., ASHOKAN, S. C., BOJAN, A. B. & GANESH, R. M. 2012. Literature review of amelogenesis imperfecta with case report. *Journal of Indian Academy of Oral Medicine and Radiology,* 24, 83.

CHEN, C.-F., HU, J. C. C., ESTRELLA, M. R. P., PETERS, M. C. & BRESCIANI, E. 2013. Assessment of restorative treatment of patients with amelogenesis imperfecta. *Pediatric Dentistry,* 35, 337-342.

CHO, S. Y., KI, Y. & CHU, V. 2008. Molar incisor hypomineralization in Hong Kong Chinese children. *International Journal of Paediatric Dentistry,* 18, 348-352.

CHOUGULE, M. S., KURIL, R. B., DALAI, J. B. & MAURYA, S. B. 2018. An in vitro comparison of shear bond strength using different bonding techniques in amelogenesis imperfecta cases. *International Journal of Orthodontic Rehabilitation,* 9, 64.

CLAMAN, L., ALFARO, M. A. & MERCADO, A. 2003. An interdisciplinary approach for improved esthetic results in the anterior maxilla. *The Journal of prosthetic dentistry,* 89, 1-5.

CLARKSON, J. 1989. Review of terminology, classifications, and indices of developmental defects of enamel. *Advances in Dental Research,* 3, 104-109.

CLARKSON, J. & O'MULLANE, D. 1989. A modified DDE Index for use in epidemiological studies of enamel defects. *Journal of Dental Research,* 68, 445-450.

COHEN, J. 1992. Statistical power analysis. *Current Directions in Psychological Science,* 1, 98-101.
COLLINS, M. A., MAURIELLO, S. M., TYNDALL, D. A. & WRIGHT, J. T. 1999. Dental anomalies associated with amelogenesis imperfecta: a radiographic assessment. *Oral Surgery, Oral Medicine, Oral Pathology, Oral Radiology, and Endodontology,* 88, 358-364.
CORREA-FARIA, P., MARTINS-JUNIOR, P. A., VIEIRA-ANDRADE, R. G., MARQUES, L. S. & RAMOS-JORGE, M. L. 2013. Perinatal factors associated with developmental defects of enamel in primary teeth: a case-control study. *Brazilian oral research,* 27, 363-368.
COUNCIL, O. 2013. Guideline on dental management of heritable dental developmental anomalies. *Pediatric Dentistry,* 35, 179-184.
CRAWFORD, P. J., ALDRED, M. & BLOCH-ZUPAN, A. 2007a. Amelogenesis imperfecta. *Orphanet Journal of Rare Diseases,* 2, 1-11.
CRAWFORD, P. J., ALDRED, M. & BLOCH-ZUPAN, A. 2007b. Amelogenesis imperfecta. *Orphanet Journal of Rare Diseases,* 2, 17.
DA COSTA-SILVA, C. M., AMBROSANO, G. M., JEREMIAS, F., DE SOUZA, J. F. & MIALHE, F. L. 2011. Increase in severity of molar–incisor hypomineralization and its relationship with the colour of enamel opacity: a prospective cohort study. *International Journal of Paediatric Dentistry,* 21, 333-341.
DAMSKY, C. H. & WERB, Z. 1992. Signal transduction by integrin receptors for extracellular matrix: cooperative processing of extracellular information. *Current Opinion in Cell Biology,* 4, 772-781.
DASHASH, M., YEUNG, C. A., JAMOUS, I. & BLINKHORN, A. 2013. Interventions for the restorative care of amelogenesis imperfecta in children and adolescents. *Cochrane Database of Systematic Reviews*.

DE LA DURE-MOLLA, M., QUENTRIC, M., YAMAGUTI, P. M., ACEVEDO, A.-C., MIGHELL, A. J., VIKKULA, M., HUCKERT, M., BERDAL, A. & BLOCH-ZUPAN, A. 2014. Pathognomonic oral profile of Enamel Renal Syndrome (ERS) caused by recessive FAM20A mutations. *Orphanet Journal of Rare Diseases,* 9, 1-13.

DE MUNCK, J. D., VAN LANDUYT, K., PEUMANS, M., POITEVIN, A., LAMBRECHTS, P., BRAEM, M. & VAN MEERBEEK, B. 2005. A critical review of the durability of adhesion to tooth tissue: methods and results. *Journal of Dental Research,* 84, 118-132.

DE SOUZA-E-SILVA, C. M., PARISOTTO, T. M., STEINER-OLIVEIRA, C., GAVIAO, M. B. D. & NOBRE-DOS-SANTOS, M. 2010. Oral rehabilitation of primary dentition affected by amelogenesis imperfecta: a case report. *Journal of Contemporary Dental Practice,* 11, 71-77.

DINESHSHANKAR, J., SIVAKUMAR, M., BALASUBRAMANIUM, A. M., KESAVAN, G., KARTHIKEYAN, M. & PRASAD, V. S. 2014. Taurodontism. *Journal of Pharmacy & Bioallied Sciences,* 6, S13.

DUAN, X., YANG, S., ZHANG, H., WU, J., ZHANG, Y., JI, D., TIE, L. & BOERKOEL, C. 2019. A Novel AMELX Mutation, Its Phenotypic Features, and Skewed X Inactivation. *Journal of Dental Research*, 0022034519854973.

DURSUN, E., SAVARD, E., VARGAS, C., LOISON-ROBERT, L., CHERIFI, H., BDEOUI, F. & LANDRU, M. 2016. Management of amelogenesis imperfecta: a 15-year case history of two siblings. *Operative dentistry,* 41, 567-577.

EBNESAJJAD, S. & LANDROCK, A. H. 2014. *Adhesives technology handbook*, William Andrew.

EL-SAYED, W., SHORE, R., PARRY, D., INGLEHEARN, C. & MIGHELL, A. 2010. Ultrastructural analyses of deciduous teeth affected by hypocalcified amelogenesis imperfecta from a family with a novel Y458X FAM83H nonsense mutation. *Cells Tissues Organs,* 191, 235-239.

ELFRINK, M., TEN CATE, J., JADDOE, V., HOFMAN, A., MOLL, H. & VEERKAMP, J. 2012. Deciduous molar hypomineralization and molar incisor hypomineralization. *Journal of Dental Research,* 91, 551-555.

ENG, G., CHEN, A., VESS, T. & GINSBURG, G. 2012. Genome technologies and personalized dental medicine. *Oral Diseases,* 18, 223-235.

EPASINGHE, D. J. & YIU, C. K. Y. 2018. Effect of etching on bonding of a self-etch adhesive to dentine affected by amelogenesis imperfecta. *Journal of Investigative and Clinical Dentistry,* 9, e12276.

ERICKSON, R. L., BARKMEIER, W. W. & LATTA, M. A. 2009. The role of etching in bonding to enamel: a comparison of self-etching and etch-and-rinse adhesive systems. *Dental Materials,* 25, 1459-1467.

ESPINOSA, R., VALENCIA, R., URIBE, M., CEJA, I. & SAADIA, M. 2008. Enamel deproteinization and its effect on acid etching: an in vitro study. *Journal of Clinical Pediatric Dentistry,* 33, 13-19.

FAGRELL, T. G., DIETZ, W., JÄLEVIK, B. & NORÉN, J. G. 2010. Chemical, mechanical and morphological properties of hypomineralized enamel of permanent first molars. *Acta Odontologica Scandinavica,* 68, 215-222.

FAN, P., SELUK, L. & O'BRIEN, W. 1975. Penetrativity of sealants: I. *Journal of Dental Research,* 54, 262-264.

FARAH, R., DRUMMOND, B., SWAIN, M. & WILLIAMS, S. 2008. Relationship between laser fluorescence and enamel hypomineralisation. *Journal of Dentistry,* 36, 915-921.

FARIA-E-SILVA, A. L., DE MORAES, R. R., DE SOUSA MENEZES, M., CAPANEMA, R. R., DE MOURA, A. S. & MARTELLI-JÚNIOR, H. 2011. Hardness and microshear bond strength to enamel and dentin of permanent teeth with hypocalcified amelogenesis imperfecta. *International Journal of Paediatric Dentistry,* 21, 314-320.

FINCHAM, A., LUO, W., MORADIAN-OLDAK, J., PAINE, M., SNEAD, M. & ZEICHNER-DAVID, M. 2000. Enamel biomineralization: the assembly and disassembly of the protein extracellular organic matrix. *Development, function and evolution of teeth*, 37-61.

FULTON, A., AMLANI, M. & PAREKH, S. 2020. Oral manifestations of vitamin D deficiency in children. *British Dental Journal,* 228, 515-518.

GABARDO, G., MACIEL, J. V. B., FRANCO, A., DE LIMA, A. A. S., DA COSTA, T. R. F. & FERNANDES, Â. 2020. Radiographic analysis of dental maturation in children with amelogenesis imperfecta: A case-control study. *Special Care in Dentistry,* 40, 267-272.

GARCIA-MARGARIT, M., CATALÁ-PIZARRO, M., MONTIEL-COMPANY, J. M. & ALMERICH-SILLA, J. M. 2014. Epidemiologic study of molar-incisor hypomineralization in 8-year-old S panish children. *International Journal of Paediatric Dentistry,* 24, 14-22.

GAROT, E., COUTURE-VESCHAMBRE, C., MANTON, D., BEAUVAL, C. & ROUAS, P. 2017. Analytical evidence of enamel hypomineralisation on permanent and primary molars amongst past populations. *Scientific Reports,* 7, 1-10.

GERDOLLE, D., MORTIER, E., RICHARD, A. & VAILATI, F. 2015. Full-mouth adhesive rehabilitation in a case of amelogenesis imperfecta: a 5-year follow-up case report. *Int J Esthet Dent,* 10, 12-31.

GETTIG, E. & HART, T. C. 2003. Genetics in dental practice: social and ethical issues surrounding genetic testing. *Journal of Dental Education,* 67, 549-562.

GJØRUP, H., HAUBEK, D., HINTZE, H., HAUKALI, G., LØVSCHALL, H., HERTZ, J. M. & POULSEN, S. 2009. Hypocalcified type of amelogenesis imperfecta in a large family: clinical, radiographic, and histological findings, associated dento-

facial anomalies, and resulting treatment load. *Acta Odontologica Scandinavica,* 67, 240-247.
GORDON, L. M., COHEN, M. J., MACRENARIS, K. W., PASTERIS, J. D., SEDA, T. & JOESTER, D. 2015. Dental materials. Amorphous intergranular phases control the properties of rodent tooth enamel. *Science (New York, N.Y.),* 347, 746.
GÜTH, J.-F., KAULING, A. E. C., SCHWEIGER, J., KÜHNISCH, J. & STIMMELMAYR, M. 2017. Virtual simulation of periodontal surgery including presurgical CAD/CAM fabrication of tooth-colored removable splints on the basis of CBCT data: a case report. *Int J Periodontics Restorative Dent,* 37, e310-e320.
GUTIERREZ-SALAZAR, M. & REYES-GASGA, J. 2001. Enamel hardness and caries susceptibility in human teeth. *Revista Latinoamericana de Metalurgia y Materiales,* 21, 36-40.
GUTIÉRREZ-SALAZAR, M. D. P. & REYES-GASGA, J. 2003. Microhardness and chemical composition of human tooth. *Materials Research,* 6, 367-373.
GWINNETT, A. J. 1993. Quantitative contribution of resin infiltration/hybridization to dentin bonding. *American Journal of Dentistry,* 6, 7-9.
HABELITZ, S., MARSHALL, S., MARSHALL JR, G. & BALOOCH, M. 2001. Mechanical properties of human dental enamel on the nanometre scale. *Archives of Oral Biology,* 46, 173-183.
HAGGER, O. 1948. Neue Katalysatoren zur Polymerisation der Äthene bei Raumtemperatur. *Helvetica Chimica Acta,* 31, 1624-1630.
HARLEEN, N., RAMAKRISHNA, Y. & MUNSHI, A. 2011. Enamel deproteinization before acid etching and its effect on the shear bond strength–An in vitro study. *Journal of Clinical Pediatric Dentistry,* 36, 19-24.
HART, T. & HART, P. 2009. Genetic studies of craniofacial anomalies: clinical implications and applications. *Orthodontics & Craniofacial Research,* 12, 212-220.

HASHEM, A., KELLY, A., O'CONNELL, B. & O'SULLIVAN, M. 2013. Impact of moderate and severe hypodontia and amelogenesis imperfecta on quality of life and self-esteem of adult patients. *Journal of Dentistry,* 41, 689-694.

HARNAGEA, H., COUTURIER, Y., SHRIVASTAVA, R., GIRARD, F., LAMOTHE, L., BEDOS, C. P. & EMAMI, E. 2017. Barriers and facilitators in the integration of oral health into primary care: a scoping review. BMJ open, 7, e016078.

HASHIMOTO, M., NAGANO, F., ENDO, K. & OHNO, H. 2011. A review: Biodegradation of resin–dentin bonds. *JapaneseDental Science Review,* 47, 5-12.

HEALTH, N. I. O. 2017. National Human Genome ResearchInstitute (NHGRI). *RNA synthesis and processing*.

HU, J. C. & SIMMER, J. 2007. Developmental biology and genetics of dental malformations. *Orthodontics & Craniofacial Research,* 10, 45-52.

HU, J. C.-C., CHUN, Y.-H. P., AL HAZZAZZI, T. & SIMMER, J. P. 2007. Enamel formation and amelogenesis imperfecta. *Cells Tissues Organs,* 186, 78-85.

HYUN, H. K., LEE, S. K., LEE, K. E., KANG, H. Y., KIM, E. J., CHOUNG, P. H. & KIM, J. W. 2009. Identification of a novelFAM83H mutation and microhardness of an affected molar in autosomal dominant hypocalcified amelogenesis imperfecta. *International Endodontic Journal,* 42, 1039-1043.

IIJIMA, M., FAN, D., BROMLEY, K. M., SUN, Z. & MORADIAN-OLDAK, J. 2010. Tooth enamel proteins enamelin and amelogenin cooperate to regulate the growth morphology of octacalcium phosphate crystals. *Crystalgrowth & design,* 10, 4815-4822.

ISHIOKA, S. & CAPUTO, A. 1989. Interaction between the dentinal smear layer and composite bond strength. *TheJournal of prosthetic dentistry,* 61, 180-185.

JÄLEVIK, B., NORÉN, J. G., KLINGBERG, G. & BARREGÅRD, L. 2001. Etiologic factors influencing the prevalence of demarcated opacities in permanent first molars in a group of Swedish children. *European Journal of Oral Sciences,* 109, 230-234.

JALILI, I. & SMITH, N. 1988. A progressive cone-rod dystrophy and amelogenesis imperfecta: a new syndrome. *Journal of Medical Genetics,* 25, 738-740.

JOINER, A. 2004. Tooth colour: a review of the literature. *Journal of Dentistry,* 32, 3-12.

JOINER, A. & LUO, W. 2017. Tooth colour and whiteness: A review. *Journal of Dentistry,* 67, S3-S10.

KAMMOUN, R., BEHETS, C., MANSOUR, L. & GHOUL-MAZGAR, S. 2018. Mineral features of connective dental hard tissues in hypoplastic amelogenesis imperfecta. *Oral diseases,* 24, 384-392.

KAMMOUN, R., ZMANTAR, T., LABIDI, A., ABBES, I., MANSOUR, L. & GHOUL-MAZGAR, S. 2019. Dental caries and hypoplastic amelogenesis imperfecta: clinical, structural, biochemical and molecular approaches. *Microbial pathogenesis,* 135, 103615.

KIM, J., PARAVINA, R. & CHEN, J.-W. 2007. In vivo evaluation of color of primary teeth. *Pediatric Dentistry,* 29, 383-386.

KIM, J., SIMMER, J., HART, T., HART, P., RAMASWAMI, M., BARTLETT, J. & HU, J. C. 2005. MMP-20 mutation in autosomal recessive pigmented hypomaturation amelogenesis imperfecta. *Journal of Medical Genetics,* 42, 271-275.

KIM, J.-W., LEE, S.-K., LEE, Z. H., PARK, J.-C., LEE, K.-E., LEE, M.-H., PARK, J.-T., SEO, B.-M., HU, J. C.-C. & SIMMER, J. P. 2008. FAM83H mutations in families with autosomal-dominant hypocalcified amelogenesis imperfecta. *The American Journal of Human Genetics,* 82, 489-494.

KIRZIOGLU, Z., CEYHAN, D. & BAYRAKTAR, C. 2019. Dental age estimation by different methods in patients with amelogenesis imperfecta. *Forensic science international,* 298, 341-344.

KLINK, A., GROTEN, M. & HUETTIG, F. 2018. Complete rehabilitation of compromised full dentitions with adhesively bonded all-ceramic single-tooth restorations: Long-term outcome in patients with and without amelogenesis imperfecta. *Journal of dentistry,* 70, 51-58.

KUNIN, A. A., EVDOKIMOVA, A. Y. & MOISEEVA, N. S. 2015. Age-related differences of tooth enamel morphochemistry in health and dental caries. *EPMA Journal,* 6, 3.

LACRUZ, R. S., HABELITZ, S., WRIGHT, J. T. & PAINE, M. L. 2017. Dental enamel formation and implications for oral health and disease. *Physiological Reviews,* 97, 939-993.

LEEVAILOJ, C., LAWANRATTANAKUL, S. & MAHATUMARAT, K. 2017. Amelogenesis Imperfecta: case study. *Operative dentistry,* 42, 457-469.

LINDUNGER, A. & SMEDBERG, J.-I. 2005. A retrospective study of the prosthodontic management of patients with amelogenesis imperfecta. *International Journal of Prosthodontics,* 18.

LUNDGREN, G. P. & DAHLLÖF, G. 2014. Outcome of restorative treatment in young patients with amelogenesis imperfecta. a cross-sectional, retrospective study. *Journal of Dentistry,* 42, 1382-1389.

LUNDGREN, G. P., VESTLUND, G.-I. M. & DAHLLÖF, G. 2018. Crown therapy in young individuals with amelogenesis imperfecta: Long term follow-up of a randomized controlled trial. *Journal of dentistry,* 76, 102-108.

LUNDGREN, G. P., WICKSTRÖM, A., HASSELBLAD, T. & DAHLLÖF, G. 2016. Amelogenesis imperfecta and early restorative crown therapy: an interview study with adolescents and young adults on their experiences. *PloS One,* 11.

MAHONEY, E. K., ROHANIZADEH, R., ISMAIL, F., KILPATRICK, N. & SWAIN, M. 2004. Mechanical properties and microstructure of hypomineralised enamel of permanent teeth. *Biomaterials,* 25, 5091-5100.

MALLOY, K. M., ROSEN, D. & ROSEN, M. R. Pocket Dentistry.

MANUJA, N., NAGPAL, R. & PANDIT, I. K. 2012. Dental adhesion: mechanism, techniques and durability. *The Journal of clinical pediatric dentistry,* 36, 223.

MARGOLIS, H., BENIASH, E. & FOWLER, C. 2006. Role of macromolecular assembly of enamel matrix proteins in enamel formation. *Journal of Dental Research,* 85, 775-793.

MARKOVIC, D., PETROVIC, B. & PERIC, T. 2010. Clinical findings and oral rehabilitation of patients with amelogenesis imperfecta. *European Archives of Paediatric Dentistry,* 11, 201-208.

MARQUEZIN, M. C. S., ZANCOPÉ, B. R., PACHECO, L. F., GAVIÃO, M. B. D. & PASCON, F. M. 2015. Aesthetic and functional rehabilitation of the primary dentition affected by amelogenesis imperfecta. *Case reports in dentistry,* 2015.

MARSHALL, S. J., BAYNE, S. C., BAIER, R., TOMSIA, A. P. & MARSHALL, G. W. 2010. A review of adhesion science. *Dental Materials,* 26, e11-e16.

MCDOWALL, F., KENNY, K., MIGHELL, A. & BALMER, R. 2018. Genetic testing for amelogenesis imperfecta: knowledge and attitudes of paediatric dentists. *British Dental Journal,* 225, 335-339.

MICHAELIDES, M., HARDCASTLE, A. J., HUNT, D. M. & MOORE, A. T. 2006. Progressive cone and cone-rod dystrophies: phenotypes and underlying molecular genetic basis. *Survey of Ophthalmology,* 51, 232-258.

MOHAMMADI, Z. 2008. Sodium hypochlorite in endodontics: an update review. *International Dental Journal,* 58, 329-341.

MOORER, W. & WESSELINK, P. 1982. Factors promoting the tissue dissolving capability of sodium hypochlorite. *International Endodontic Journal,* 15, 187-96.

MORADIAN-OLDAK, J. 2012. Protein-mediated enamel mineralization. *Frontiers in bioscience: a journal and virtual library,* 17, 1996.

MOREIRA, R. F., FIGUEIREDO, R. G., OLIVEIRA, H. E., FONSECA, A. C. L. D. & MIRANDA, M. S. D. 2016. Immediate desensitization in teeth affected by amelogenesis imperfecta. *Brazilian dental journal,* 27, 359-362.

MORI, H., IZAWA, T., MORI, H., WATANABE, K., KANNO, T. & TANAKA, E. 2019. Skeletal open bite with amelogenesis imperfecta treated with compression osteogenesis: a case report. *Head & face medicine,* 15, 1-7.

MOUSSALLY, C., FRON-CHABOUIS, H., CHARRIÈRE, A., MALADRY, L. & DURSUN, E. 2019. Full-mouth Rehabilitation of Hypocalcified-type Amelogenesis Imperfecta With Chairside Computer-aided Design and Computer-aided Manufacturing: A Case Report. *Operative dentistry,* 44, E145-E158.

NANCI, A. 2008. Oral Mucosa, Ten Cate's Oral Histology. *Mosby–Elsevier, Indian*, 319-56.

NANCI, A. 2017. *Ten Cate's Oral Histology-E-Book: Development, Structure, and Function*, Elsevier Health Sciences.

NARANG, A., MAGUIRE, A., NUNN, J. & BUSH, A. 2003. Oral health and related factors in cystic fibrosis and other chronic respiratory disorders. *Archives of Disease in Childhood,* 88, 702-707.

NATHWANI, N. S. & KELLEHER, M. 2010. Minimally destructive management of amelogenesis imperfecta and hypodontia with bleaching and bonding. *Dental Update,* 37, 170-179.

NIGAM, P., SINGH, V. P., PRASAD, K. D. & TAK, J. 2015. Amelogenesis imperfecta: a case report and review of literature. *Int J Sci Stud,* 2, 146-149.

ODELL, E. W. 2017. *Cawson's essentials of oral pathology and oral medicine e-book*, Elsevier Health Sciences.

ORTIZ, L., PEREIRA, A. M., JAHANGIRI, L. & CHOI, M. 2019. Management of amelogenesis imperfecta in adolescent patients: clinical report. *Journal of Prosthodontics,* 28, 607-612.

OSORIO, R., PISANI-PROENCA, J., ERHARDT, M. C. G., OSORIO, E., AGUILERA, F. S., TAY, F. R. & TOLEDANO, M. 2008. Resistance of ten contemporary adhesives to resin–dentine bond degradation. *Journal of Dentistry,* 36, 163-169.

PARRY, D. A., MIGHELL, A. J., EL-SAYED, W., SHORE, R. C., JALILI, I. K., DOLLFUS, H., BLOCH-ZUPAN, A., CARLOS, R., CARR, I. M. & DOWNEY, L. M. 2009. Mutations in CNNM4 cause Jalili syndrome, consisting of autosomal-recessive cone-rod dystrophy and amelogenesis imperfecta. *The American Journal of Human Genetics,* 84, 266-273.

PASHLEY, D. H. 1991. Dentin bonding: overview of the substrate with respect to adhesive material. *Journal of Esthetic and Restorative Dentistry,* 3, 46-50.

PATEL, M., MCDONNELL, S., IRAM, S. & MF WY, C. 2013. Amelogenesis imperfecta-lifelong management. Restorative management of the adult patient. *British Dental Journal,* 215, 449.

PATEL, R.-R. A., HOVIJITRA, S., KAFRAWY, A. H. & BIXLER, D. 1991. X-linked (recessive) hypomaturation amelogenesis imperfecta: a prosthodontic, genetic, and histopathologic report. *Journal of Prosthetic Dentistry,* 66, 398-402.

PERDIGAO, J., GOMES, G., GONDO, R. & FUNDINGSLAND, J. W. 2006. In vitro bonding performance of all-in-one adhesives. Part I--microtensile bond strengths. *Journal of Adhesive Dentistry,* 8.

PINKHAM, J. R., CASAMASSIMO, P., FIELDS, H., MCTIGUE, D. & NOWAK, A. 2005. Pediatric dentistry. *Infancy through adolescence,* 4.

POCIUS, A. V. 2012. *Adhesion and adhesives technology: an introduction*, Carl Hanser Verlag GmbH Co KG.

POUSETTE LUNDGREN, G., MORLING VESTLUND, G., TRULSSON, M. & DAHLLÖF, G. 2015. A randomized controlled trial of crown therapy in young individuals with amelogenesis imperfecta. *Journal of Dental Research,* 94, 1041-1047.

POULSEN, S., GJØRUP, H., HAUBEK, D., HAUKALI, G., HINTZE, H., LØVSCHALL, H. & ERRBOE, M. 2008. Amelogenesis imperfecta–a systematic literature review of associateddental and oro-facial abnormalities and their impact onpatients. *Acta Odontologica Scandinavica,* 66, 193-199.

POUSETTE LUNDGREN, G., WICKSTRÖM, A., HASSELBLAD, T. & DAHLLÖF, G. 2016. Amelogenesis imperfecta and early restorative crown therapy: an interview study with adolescents and young adults on their experiences. *PLoS ONE,* 11, e0156879.

QUANDALLE, C., BOILLOT, A., FOURNIER, B., GARREC, P., DE LA DURE-MOLLA, M. & KERNER, S. 2020. Gingival inflammation, enamel defects, and tooth sensitivity in children with amelogenesis imperfecta: a case-control study. *Journal of Applied Oral Science,* 28.

RAJPAR, M. H., HARLEY, K., LAING, C., DAVIES, R. M. & DIXON, M. J. 2001. Mutation of the gene encoding the enamel-specific protein, enamelin, causes autosomal-dominant amelogenesis imperfecta. *Human Molecular Genetics,* 10, 1673-1677.

RANGANATH, V., NICHANI, A. S. & SOUMYA, V. 2010. Amelogenesis imperfecta: A challenge to restoring esthetics and function. *Journal of Indian Society of Periodontology,* 14, 195.

RATBI, I., FALKENBERG, K. D., SOMMEN, M., AL-SHEQAIH, N., GUAOUA, S., VANDEWEYER, G., URQUHART, J. E., CHANDLER, K. E., WILLIAMS, S. G. & ROBERTS, N. A. 2015. Heimler syndrome is caused by hypomorphic mutations in the peroxisome-biogenesis genes PEX1 and PEX6. *The American Journal of Human Genetics,* 97, 535-545.

ROBINSON, C., KIRKHAM, J., BROOKES, S., BONASS, W. & SHORE, R. 1995. The chemistry of enamel development. *The International journal of developmental biology,* 39, 145.

ROBINSON, C., KIRKHAM, J., BROOKES, S., BONASS, W. & SHORE, R. 2003. The chemistry of enamel development. *International Journal of Developmental Biology,* 39, 145-152.

ROBINSON, C., KIRKHAM, J., BROOKES, S. J. & SHORE, R. C. 2017. Chemistry of mature enamel. *Dental enamel formation to destruction.* CRC Press.

ROGERS, H., YESUDIAN, G. & RODD, H. 2016. Unusual extrinsic staining following microabrasion in a girl with amelogenesis imperfecta. *European Archives of Paediatric Dentistry,* 17, 271-275.

ROMA, M. & HEGDE, S. 2016. Amelogenesis Imperfecta: A Review of the Literature. *Journal of Pharmaceutical Sciences and Research,* 8, 1042.

ROWLEY, R., HILL, F. & WINTER, G. 1982. An investigation of the association between anterior open-bite and amelogenesis imperfecta. *American Journal of Orthodontics,* 81, 229-235.

RUAN, Q. & MORADIAN-OLDAK, J. 2015. Amelogenin and enamel biomimetics. *Journal of Materials Chemistry B,* 3, 3112-3129.

SABANDAL, M. M., DAMMASCHKE, T. & SCHÄFER, E. 2020. Restorative treatment in a case of amelogenesis imperfecta and 9-year follow-up: a case report. *Head & face medicine,* 16, 1-8.

SABANDAL, M. M. & SCHÄFER, E. 2016. Amelogenesis imperfecta: review of diagnostic findings and treatment concepts. *Odontology,* 104, 245-256.

SABATINI, C. & GUZMÁN-ARMSTRONG, S. 2009. A conservative treatment for amelogenesis imperfecta with direct resin composite restorations: a case report. *Journal of Esthetic and Restorative Dentistry,* 21, 161-169.

SÁNCHEZ-QUEVEDO, M., CEBALLOS, G., GARCIA, J., LUNA, J., RODRIGUEZ, I. & CAMPOS, A. 2004. Dentine structure and mineralization in hypocalcified amelogenesis imperfecta: a quantitative X-ray histochemical study. *Oral Diseases,* 10, 94-98.

ŞAROĞLU, I., ARAS, Ş. & ÖZTAŞ, D. 2006. Effect of deproteinization on composite bond strength in hypocalcified amelogenesis imperfecta. *Oral Diseases,* 12, 305-308.

SARR, M., KANE, A. W., VREVEN, J., MINE, A., VAN LANDUYT, K. L., PEUMANS, M., LAMBRECHTS, P., VAN MEERBEEK, B. & DE MUNCK, J. 2010. Microtensile bond strength and interfacial characterization of 11 contemporary adhesives bonded to bur-cut dentin. *Operative Dentistry,* 35, 94-104.

SCHMIDLIN, P. R., ZEHNDER, M., PASQUALETTI, T., IMFELD, T. & BESEK, M. J. 2004. Penetration of a bonding agent into De-and remineralized enamel in vitro. *Journal of Adhesive Dentistry,* 6.

SEOW, W. & AMARATUNGE, A. 1998. The effects of acid-etching on enamel from different clinical variants of amelogenesis imperfecta: an SEM study. *Pediatric Dentistry,* 20, 37-42.

SEOW, W. K., FORD, D., KAZOULLIS, S., NEWMAN, B. & HOLCOMBE, T. 2011. Comparison of enamel defects in the primary and permanent dentitions of children from a low-fluoride District in Australia. *Pediatric Dentistry,* 33, 207-212.

SEZINANDO, A. 2014. Looking for the ideal adhesive–a review. *Revista Portuguesa de Estomatologia, Medicina Dentária e Cirurgia Maxilofacial,* 55, 194-206.

SHIBATA, S., TAGUCHI, C., GONDO, R., STOLF, S. & BARATIERI, L. 2016. Ceramic veneers and direct-composite cases of amelogenesis imperfecta rehabilitation. *Operative dentistry,* 41, 233-242.

SHIVHARE, P., SHANKARNARAYAN, L., GUPTA, A. & SUSHMA, P. 2016. Amelogenesis Imperfecta: A Review. *Journal of Advanced Oral Research,* 7, 1-6.

SHOLAPURKAR, A. A., JOSEPH, R. M., VARGHESE, J. M., NEELAGIRI, K., ACHARYA, S., HEGDE, V., PAI, K. M. & BHAT, M. 2008. Clinical diagnosis and oral rehabilitation of a patient with amelogenesis imperfecta: a case report. *The journal of contemporary dental practice,* 9, 92-98.

SHORE, R., BÄCKMAN, B., ELCOCK, C., BROOK, A., BROOKES, S. & KIRKHAM, J. 2010. The structure and composition of deciduous enamel affected by local hypoplastic autosomal dominant amelogenesis imperfecta resulting from an ENAM mutation. *Cells Tissues Organs,* 191, 301-306.

SILVERSTONE, L., SAXTON, C., DOGON, I. L. & FEJERSKOV, O. 1975. Variation in the pattern of acid etching of human dental enamel examined by scanning electron microscopy. *Caries Research,* 9, 373-387.

SIMMER, J. & FINCHAM, A. 1995. Molecular mechanisms of dental enamel formation. *Critical Reviews in Oral Biology and Medicine,* 6, 84-108.

SIMMER, S., ESTRELLA, N., MILKOVICH, R. & HU, J. 2013. Autosomal dominant amelogenesis imperfecta associated with ENAM frameshift mutation p. Asn36Ilefs56. *Clinical genetics,* 83, 195.

SINGH, A., AGRAWAL, S. K., SHRESTHA, A. & BHAGAT, T. 2018. Amelogenesis Imperfecta: A Case Series from the Community. *Journal of the Nepal Medical Association,* 56.

SLAVKIN, H. C. & BAVETTA, L. A. 2017. *Developmental aspects of oral biology*, Elsevier.

SMITH, C. 1998. Cellular and chemical events during enamel maturation. *Critical Reviews in Oral Biology and Medicine,* 9, 128-161.

SMITH, C. E., POULTER, J. A., ANTANAVICIUTE, A., KIRKHAM, J., BROOKES, S. J., INGLEHEARN, C. F. & MIGHELL, A. J. 2017. Amelogenesis imperfecta; genes, proteins, and pathways. *Frontiers in Physiology,* 8, 435.

SMITH, R., ELCOCK, C., ABDELLATIF, A., BÄCKMAN, B., RUSSELL, J. & BROOK, A. 2009. Enamel defects in extracted and exfoliated teeth from patients with Amelogenesis Imperfecta, measured using the extended enamel defects index and image analysis. *Archives of Oral Biology,* 54, S86-S92.

SÖNMEZ, H., YILDIRIM, G. & BEZGIN, T. 2013. Putative factors associated with molar incisor hypomineralisation: an epidemiological study. *European Archives of Paediatric Dentistry,* 14, 375-380.

SÖNMEZ, I. Ş., ARAS, Ş., TUNÇ, E. Ş. & KÜÇÜKEŞMEN, Ç. 2009. Clinical success of deproteinization in hypocalcified amelogenesis imperfecta. *Quintessence International,* 40.

SOUZA, J. F. D., FRAGELLI, C. M. B., PASCHOAL, M. A. B., CAMPOS, E. A., CUNHA, L. F., LOSSO, E. M. & CORDEIRO, R. D. C. L. 2014. Noninvasive and multidisciplinary approach to the functional and esthetic rehabilitation of amelogenesis imperfecta: a pediatric case report. *Case reports in dentistry,* 2014.

SOVIERO, V., HAUBEK, D., TRINDADE, C., DA MATTA, T. & POULSEN, S. 2009. Prevalence and distribution of demarcated opacities and their sequelae in permanent 1st molars and incisors in 7 to 13-year-old Brazilian children. *Acta Odontologica Scandinavica,* 67, 170-175.

SOXMAN, J. A., WUNSCH, P. B. & HABERLAND, C. M. 2019. Anomalies of tooth formation. *Anomalies of the Developing Dentition.* Springer.

STERNE, J., SAVOVIĆ, J., PAGE, M., ELBERS, R., BLENCOWE, N., BOUTRON, I., CATES, C., CHENG, H., CORBETT, M. & ELDRIDGE, S. 2019. RoB 2: A revised Cochrane risk-of-bias tool for randomized trials. *BMJ,* 366, 1-24.

STRAUCH, S. & HAHNEL, S. 2018. Restorative treatment in patients with amelogenesis imperfecta: A review. *Journal of Prosthodontics,* 27, 618-623.

SUNDFELD, R. H., SUNDFELD-NETO, D., MACHADO, L. S., FRANCO, L. M., FAGUNDES, T. C. & BRISO, A. L. F. 2014. Microabrasion in tooth enamel discoloration defects: three cases with long-term follow-ups. *Journal of Applied Oral Science,* 22, 347-354.

SUPPA, P., BRESCHI, L., RUGGERI, A., MAZZOTTI, G., PRATI, C., CHERSONI, S., DI LENARDA, R., PASHLEY, D. H. & TAY, F. R. 2005. Nanoleakage within the hybrid layer: a correlative FEISEM/TEM investigation. *Journal of Biomedical Materials Research Part B: Applied Biomaterials: An Official Journal of The Society for Biomaterials, The Japanese Society for Biomaterials, and The Australian Society for Biomaterials and the Korean Society for Biomaterials,* 73, 7-14.

TAY, F. R. & PASHLEY, D. H. 2001. Aggressiveness of contemporary self-etching systems: I: Depth of penetration beyond dentin smear layers. *Dental Materials,* 17, 296-308.

TEN CATE, A. & NANCI, A. 2013. *Ten Cate's oral histology: development, structure, and function*, Elsevier.

THESLEFF, I. & JUURI, E. 2015. Dental Epithelial Stem Cells. *Stem Cell Biology and Tissue Engineering in Dental Sciences.* Elsevier.

TOUPENAY, S., FOURNIER, B. P., MANIÈRE, M.-C., IFI-NAULIN, C., BERDAL, A. & DE LA DURE–MOLLA, M. 2018. Amelogenesis imperfecta: therapeutic strategy from primary to permanent dentition across case reports. *BMC oral health,* 18, 1-8.

TUMMERS, M. & THESLEFF, I. 2008. Observations on continuously growing roots of the sloth and the K14-Eda transgenic mice indicate that epithelial stem cells can give rise to both the ameloblast and root epithelium cell lineage creating distinct tooth patterns. *Evolution & development,* 10, 187-195.

URZÚA, B., ORTEGA-PINTO, A., MORALES-BOZO, I., ROJAS-ALCAYAGA, G. & CIFUENTES, V. 2011. Defining a New Candidate Gene for Amelogenesis Imperfecta: From Molecular Genetics to Biochemistry. *Biochemical Genetics,* 49, 104-121.

VAN LANDUYT, K. L., SNAUWAERT, J., DE MUNCK, J., PEUMANS, M., YOSHIDA, Y., POITEVIN, A., COUTINHO, E., SUZUKI, K., LAMBRECHTS, P. & VAN MEERBEEK, B. 2007. Systematic review of the chemical composition of contemporary dental adhesives. *Biomaterials,* 28, 3757-3785.

VAN MEERBEEK, B., DE MUNCK, J., YOSHIDA, Y., INOUE, S., VARGAS, M., VIJAY, P., VAN LANDUYT, K., LAMBRECHTS, P. & VANHERLE, G. 2003. Buonocore Memorial Lecture. Adhesion to enamel and dentin: Current status and future challenges. *Operative Dentistry,* 28, 215.

VAN MEERBEEK, B., YOSHIHARA, K., YOSHIDA, Y., MINE, A., DE MUNCK, J. & VAN LANDUYT, K. 2011. State of the art of self-etch adhesives. *Dental Materials,* 27, 17-28.

VENEZIE, R. D., VADIAKAS, G., CHRISTENSEN, J. R. & WRIGHT, J. T. 1994. Enamel pretreatment with sodium hypochlorite to enhance bonding in hypocalcified amelogenesis imperfecta: case report and SEM analysis. *Pediatric Dentistry,* 16, 433-433.

VIOLICH, D. & CHANDLER, N. 2010. The smear layer in endodontics–a review. *International Endodontic Journal,* 43, 2-15.

VISWESWAR, V., AMARLAL, D. & VEERABAHU, R. 2012. Prevalence of developmental defects of enamel in children and adolescents with asthma: A cross-sectional study. *Indian Journal of Dental Research,* 23, 697-698.

VOLODARSKY, M., ZILBERMAN, U. & BIRK, O. S. 2015. Novel FAM20A mutation causes autosomal recessive amelogenesis imperfecta. *Archives of Oral Biology,* 60, 919-922.

WEERHEIJM, K., JALEVIK, B. & ALALUUSUA, S. 2001. Molar-incisor hypomineralisation. *Caries Research,* 35, 390.
WENZEL, R. N. 1936. Resistance of solid surfaces to wetting by water. *Industrial and Engineering Chemistry,* 28, 988-994.
WHITE, S., LUO, W., PAINE, M., FONG, H., SARIKAYA, M. & SNEAD, M. 2001. Biological organization of hydroxyapatite crystallites into a fibrous continuum toughens and controls anisotropy in human enamel. *Journal of Dental Research,* 80, 321-326.
WILLIAM, V., MESSER, L. B. & BURROW, M. F. 2006. Molar incisor hypomineralization: review and recommendations for clinical management. *Pediatric Dentistry,* 28, 224-232.
WINTER, G. & BROOK, A. 1975. Enamel hypoplasia and anomalies of the enamel. *Dental Clinics of North America,* 19, 3.
WITKOP JR, C. 1988. Amelogenesis imperfecta, dentinogenesis imperfecta and dentin dysplasia revisited: problems in classification. *Journal of Oral Pathology,* 17, 547.
WITKOP JR, C. J. 1967. Partial expression of sex-linked recessive amelogenesis imperfecta in females compatible with the Lyon hypothesis. *Oral Surgery, Oral Medicine, Oral Pathology,* 23, 174-182.
WOGELIUS, P., HAUBEK, D. & POULSEN, S. 2008. Prevalence and distribution of demarcated opacities in permanent 1st molars and incisors in 6 to 8-year-old Danish children. *Acta Odontologica Scandinavica,* 66, 58-64.
WRIGHT, J., DEATON, T., HALL, K. & YAMAUCHI, M. 1995. The mineral and protein content of enamel in amelogenesis imperfecta. *Connective Tissue Research,* 32, 247-252.
WRIGHT, J., HART, P., ALDRED, M., SEOW, K., CRAWFORD, P., HONG, S., GIBSON, C. & HART, T. 2003. Relationship of phenotype and genotype in X-linked amelogenesis imperfecta. *Connective Tissue Research,* 44, 72-78.

WRIGHT, J. T. 2002. The etch-bleach-seal technique for managing stained enamel defects in young permanent incisors. *Pediatr Dent,* 24, 249-252.
WRIGHT, J. T. 2006. The molecular etiologies and associated phenotypes of amelogenesis imperfecta. *American journal of medical genetics Part A,* 140, 2547-2555.
WRIGHT, J. T. 2015. Amelogenesis Imperfecta: Current Understanding of Genotype-Phenotype. *Planning and Care for Children and Adolescents with Dental Enamel Defects.* Springer.
WRIGHT, J. T., TORAIN, M., LONG, K., SEOW, K., CRAWFORD, P., ALDRED, M. J., HART, P. S. & HART, T. C. 2011. Amelogenesis imperfecta: genotype-phenotype studies in 71 families. *Cells Tissues Organs,* 194, 279-283.
XU, H., SMITH, D., JAHANMIR, S., ROMBERG, E., KELLY, J., THOMPSON, V. & REKOW, E. 1998. Indentation damage and mechanical properties of human enamel and dentin. *Journal of Dental Research,* 77, 472-480.
YAMAN, B. C., OZER, F., CABUKUSTA, C. S., EREN, M. M., KORAY, F. & BLATZ, M. B. 2014. Microtensile bond strength to enamel affected by hypoplastic amelogenesis imperfecta. *Journal of Adhesive Dentistry,* 16.
YANG, Y., REICHL, F.-X., ILIE, N., SHI, J., DHEIN, J., HICKEL, R. & HÖGG, C. 2019. Antioxidants as a novel dental resin-composite component: Effect on elution and degree of conversion. *Dental Materials,* 35, 650-661.
YOSHIDA, Y. & INOUE, S. 2012. Chemical analyses in dental adhesive technology. *Japanese Dental Science Review,* 48, 141-152.
ZAGDWON, A., FAYLE, S. & POLLARD, M. 2003. A prospective clinical trial comparing preformed metal crowns and cast restorations for defective first permanent molars. *European Journal of Paediatric Dentistry,* 4, 138-142.
ZIMMERMANN, M., KOLLER, C., HICKEL, R. & KÜHNISCH, J. 2016. Chairside treatment of amelogenesis imperfecta,

including establishment of a new vertical dimension with resin nanoceramic and intraoral scanning. *The Journal of prosthetic dentistry,* 116, 309-313.

8. Appendix

Appendix 1: Service evaluation registration form

Eastman Dental Hospital Clinical Audit - Project Registration Form

Project Title: The Burden and Impact of Amelogenesis Imperfecta care at Paediatric Department, Eastman Dental Hospital		
Start Date: 18/03/2019	**Expected End Date: 1/08/2019**	**Department:** Paediatrics
Project Lead: Husam Al siyabi, Dr. Paul Ashely , Dr. Susan Parekh & Dr. Joana Monteiro	**E-mail:**	**Phone:**
✓ **Approved by Departmental Audit Lead**	✓**Approved by Departmental Clinical Lead**	
Other Personnel Involved (please include contact details)**:** NIL		

Reasons for audit:

- To assess the burden of care for child patients with Amelogenesis Imperfecta (AI) attending the Paediatric Dental Dept at EDH

Aims and objectives with an overview of the project:

- To determine the type of treatment, duration and number of appointments for patients receiving specialist care for A.I.

Standards:

This is service evaluation, therefore there is no standard available.

Materials and method for measuring against standards (include sample size & timescale):

Study design

This is a retrospective audit of AI patient records within the Eastman Dental Hospital.

Study population

Uses existing databases of patients with AI from the anomalies database and asking colleagues

Data collection

Data collection will be from both patient clinical notes in paper and electronic if available

Data collection

Form for each patient and transferred to Microsoft Excel to allow analysis

Data Protection

All collected data will be kept in the postgraduate office with secure entry office, electronic data will be stored in password protected computer within EDH/EDI on share drive

Statistical analyses

Descriptive statistics will be undertaken including means and standard deviations.

Please return this completed form electronically by

e-mail it to either EDH Audit Lead: Sonita.koshal@nhs.net or EDH Audit Coordinator Tracy.masterson@nhs.net

Clinical Governance, 2nd Floor, Victoria Wing, Eastman Dental Hospital, 256 Gray's Inn Road, London, WC1X 8LD

July 2019

APAT SCORE: This gives an indication of the quality of the proposed audit. If there is a low score, then a redesign may be required. *Double Click the Icon for the APAT calculator* APAT SCORE CALCULATOR			SCORE 18 / 25
Interdepartmental audit: More than one department involved	**X No**	☐ **Yes,** specify departments:	
Multi-professional audit: E.g. Dentists/ Dental Nurses/ Admin team/ Medics	**X No**	☐ **Yes,** specify professions:	
Type of audit project:	**Structure**	**Process**	**X Outcome**
Scope of audit project: (tick one or more)	**National**	**Regional**	**X Local**
Does this project link with research?	**X No**	☐ **Yes,** give details:	
Does this project link with the clinical audit priorities in the annual report?	**X No**	**Yes**, please specify: If No, please give good reason for audit:	
Audit Office support required: (*e.g.* project design, database creation, Formic)	**X No**	☐ **Yes,** please specify:	

Guidance Notes

The audit project you undertake can be part of the local audit, hospital wide or national audit programmes.

Types of Audit Project

Structure (What you need): Structure standards refer to the resources required. They may include the numbers of staff and skill mix, organisational arrangements, and the provision of equipment and physical space.

Process (What you do): Process standards refer to the actions and decisions taken by practitioners together with users. These actions may include communication, assessment, education, investigations, prescribing, surgical and other therapeutic interventions, evaluation and documentation.

Outcome (What you expect): Outcome standards are typically measures of the physical or behavioural response to an intervention, reported health status, and level of knowledge and satisfaction.

Audit Project Assessment Tool (APAT)

Assessment of all audit projects with APAT prior to start was mandated by the Trust in an effort to standardise and improve the quality of audit projects undertaken across the organisation. It is a tool that has been extensively validated and demonstrated to not only standardise audit practice, but result in a multidisciplinary focus for each project, increase the amount of re-audit, facilitate change in practice and generally re-energise audit.

If a project is assessed and it scores low, this indicates the proposed project is of inadequate quality, may not be "true" audit and the methodology and standards should be re-considered.

Support for your Audit

The Clinical Governance team offers support via the EDH Audit Lead and Audit Coordinator and Clinical Governance lead. For more information, or if you are unsure about any aspect of your audit, please do not hesitate to contact. Please also see the audit information guide on UCLH insight pages.

Registration Process

Once you have completed this form including as much detail of your audit proposal and the standards by which your audit will be measured, you should submit it to initially your departmental audit lead for approval and then the EDH audit lead or EDH audit coordinator. If you have already designed any data collection forms you should submit these at the same time (do not worry if you haven't). You should not start your audit at this point.

The registration form will be finally approved at the following departmental audit leads meeting at which point you can start your audit. You will be notified only if your registration is not approved and the recommendations required for adjustment will be advised.

Completion of Audit Project

Once you have completed your audit your departmental audit lead should be informed. A completed audit report form (available from insight pages) should be submitted to the EDH audit coordinator or EDH audit lead, **along with your audit report and presentation (if present).** The easiest way is to send them electronically via e-mail (.doc, .pdf, .ppt file types preferred). The report form should include the action plan for your audit. An audit cannot be considered complete until all documentation has been submitted to the Audit team.

Terminating an Audit

If for some reason you are unable to complete an audit, or an audit is placed on hold, please inform the EDH audit coordinator/audit lead as soon as possible along with the reasons why you are unable to complete the audit.

Implementation of Recommendations

The Clinical Governance team should be kept informed of any recommendations made as a result of an audit, and the progress (or otherwise) in implementing these recommendations. A second cycle re- audit will need re registration to show implementation of the action plan from the first cycle. A record is kept of all recommendations and progress toward their implementation is monitored.

Checklist

To ensure approval of your audit as quickly as possible please ensure you have done the following:

- Completed the registration form and set the standards.
- Defined any terms where necessary.
- Ensured you are collecting appropriate data to allow measurement of activity and comparison against the standard.
- Your registration should include sample population criteria, the time period of the audit, and a schedule for re-audit.
- Included your contact information.
- Got approval from your department Clinical Audit Lead.
-

- Have you contacted Library Services to check the evidence base on your topic?
  Please email edi-library@ucl.ac.uk or come into Eastman Dental Library.

Please ensure you do not start your audit before an approved registration form is submitted to the EDH Audit lead or Audit Coordinator.

UCLH Trust Directives: Clinical Audit Priorities

Divisional Clinical Directors and Divisional Managers are responsible for ensuring a programme of clinical audit is undertaken in their division which addresses the activity detailed below as a minimum:

- clinical audits identified to mitigate clinical risks included on the divisional risk register
- audits in response to Trust or divisional objectives and quality priorities
- clinical audits specified and monitored by the Clinical Audit & Quality Improvement Committee (e.g. local systems for radiology imaging results)
- clinical audits in response to incidents, complaints, safeguarding issues, consent
- clinical audits of compliance with key clinical policies including: medicines management, blood transfusion, resuscitation, infection control, discharge and venous thromboembolism prevention
- clinical audits relating to compliance with NICE guidance relevant to the division
- clinical audits included on the National Clinical Audit & Patient Outcomes Programme (NCAPOP) and any required for the Quality Account

- clinical audits arising from National Clinical Audit reports and National Confidential Enquiries such as NCEPOD
- clinical audits of divisional clinical procedures, guidelines and pathways *including* relevant antimicrobial guidelines
- clinical audits arising from measures of patient satisfaction, as an adjunct to Patient Reported Outcome Measures (PROMs)
- re-audits

Junior staff/ postgraduates needing to undertake an audit project should be directed to the Divisional Audit Programme in the first instance. The Clinical Audit policy and Trust information is available on Insight.

The Eastman divisional audit programme for each year is submitted on an annual basis and can be accessed if required by contacting the Audit lead directly (sonita.koshal@nhs.net)

Appendix 2: Data Collection Form

Paediatric Department
Audit form

Patient identification No:	

Gender:	Male ☐	Female ☐	Other ☐	Post code:

Medical History:		Type of AI: _____

Current age:		Age first seen: _____ Date: _____ How many times re-referred: _____ Reason: _____
Current patient:	☐	Where: _____
Discharge: Date (MM/YY):	☐ _____	Where to: _____ Reason: -!6y+ ☐ -Family circumstances/wishes ☐ -Other _____ -Multiple DNA's
Lost to follow up: Last seen (MMYY)	☐ _____	
No. of DNA's	____over ____years	
No. of Cancellation		

Family History: Unknown: ☐	Parents affected: Mum ☐ Dad ☐ Siblings affected: Brothers ☐ Sisters ☐ Both ☐ Other in the family:
Referred originally by:	
Clinics received treatment	Round trip mileage o _____ BY Car _____ By Public transport _____
Total number of operators:	_____ Level of operators: _____

Cons.	Senior Reg.	STR	PG	Specialist speciality Doctor	DCT	Therapist

NO. of total appointments	Paeds	Ortho	Ortho-Paeds	Other (Speciality)

No. of years in total been seen within service	

Treatment				
	Primary Dentition 0-5 y 11m	Mixed Dentition 6-11 y 11m	Permanent Dentition 12-18 yr	Total
Lead clinician identified	cons □ Specialist □ Other □	cons □ Specialist □ Other □	cons □ Specialist □ Other □	
No of LA appt				
No of I.S				
No of RA appt				
No of GA's Treatment under GA: No of teeth treated under G.A				
No of tx episodes (at a 3\12 R\V gap between appt)				
No of XLA'S				
No of comps on posterior teeth				
No of comps on anterior teeth				
No of onlays				
No of PMC's Hall crown				
Conventional				
No of bleaching episode				

Appendix 3: Published paper at European Archives of Paediatric Dentistry

European Archives of Paediatric Dentistry
https://doi.org/10.1007/s40368-021-00638-x

ORIGINAL SCIENTIFIC ARTICLE

The burden of dental care in Amelogenesis Imperfecta paediatric patients in the UK NHS: a retrospective, multi-centred analysis

F. Lafferty[1] · H. Al Siyabi[3] · A. Sinadinos[4] · K. Kenny[2] · A. J. Mighell[2] · J. Monteiro[5] · F. Soldani[4] · S. Parekh[3] · R. C. Balmer[2]

Received: 18 December 2020 / Accepted: 24 May 2021

Abstract

Purpose The burden of dental care in Amelogenesis Imperfecta (AI) has not been well described. This condition results in weak, discoloured and often sensitive teeth. Specialist paediatric care is available for AI patients in the UK, but treatment protocols and care provided are inconsistent. The aim of this study was therefore to analyse the provision of treatment and burden of care for children and families with AI across four Paediatric Dentistry centres in the UK.

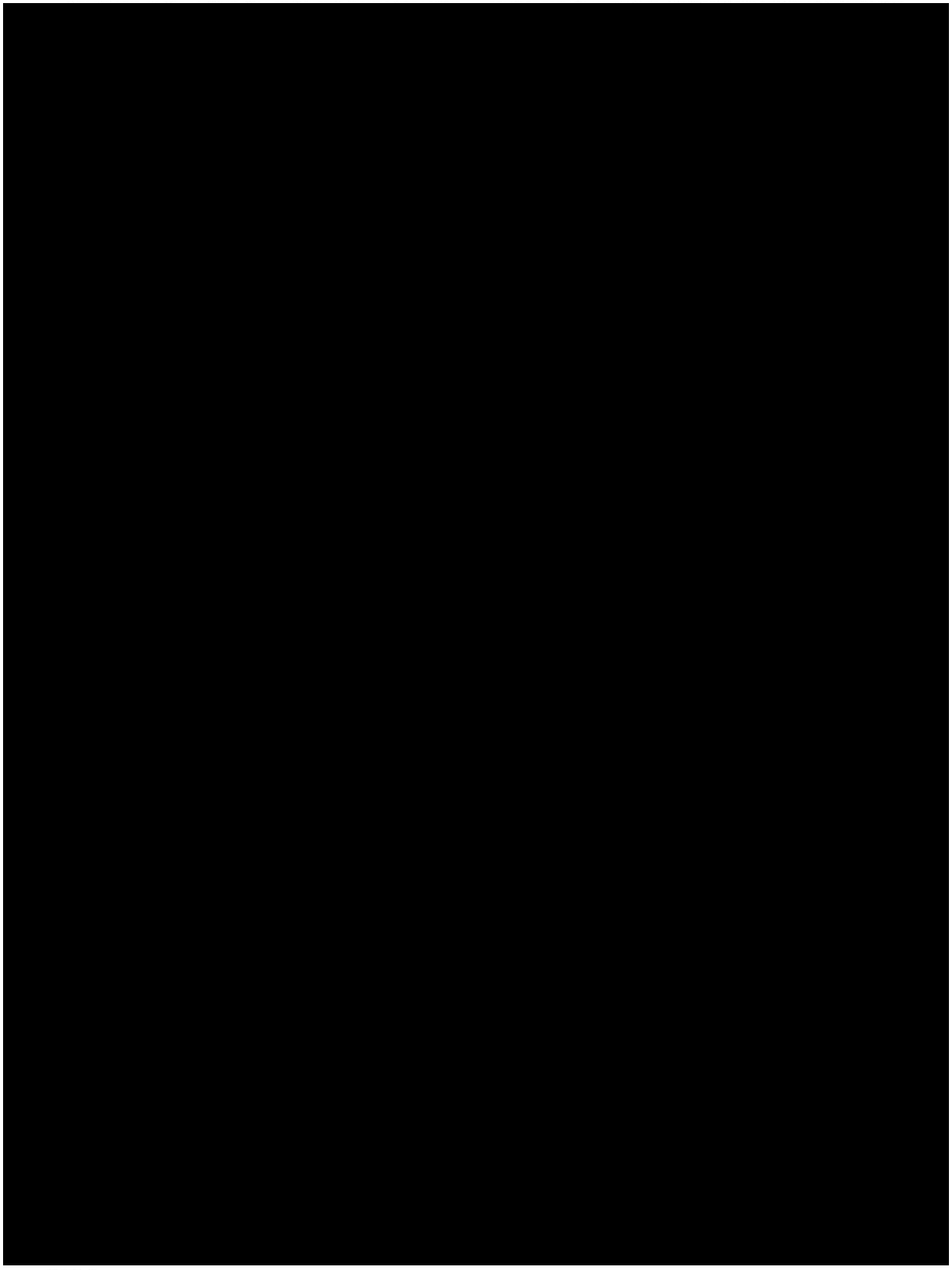
Methods A retrospective evaluation of AI patient clinical records across four UK consultant-led Paediatric Dentistry centres was completed. Frequency and duration of care were recorded along with treatment and experience of inhalation sedation, local and general anaesthetic.

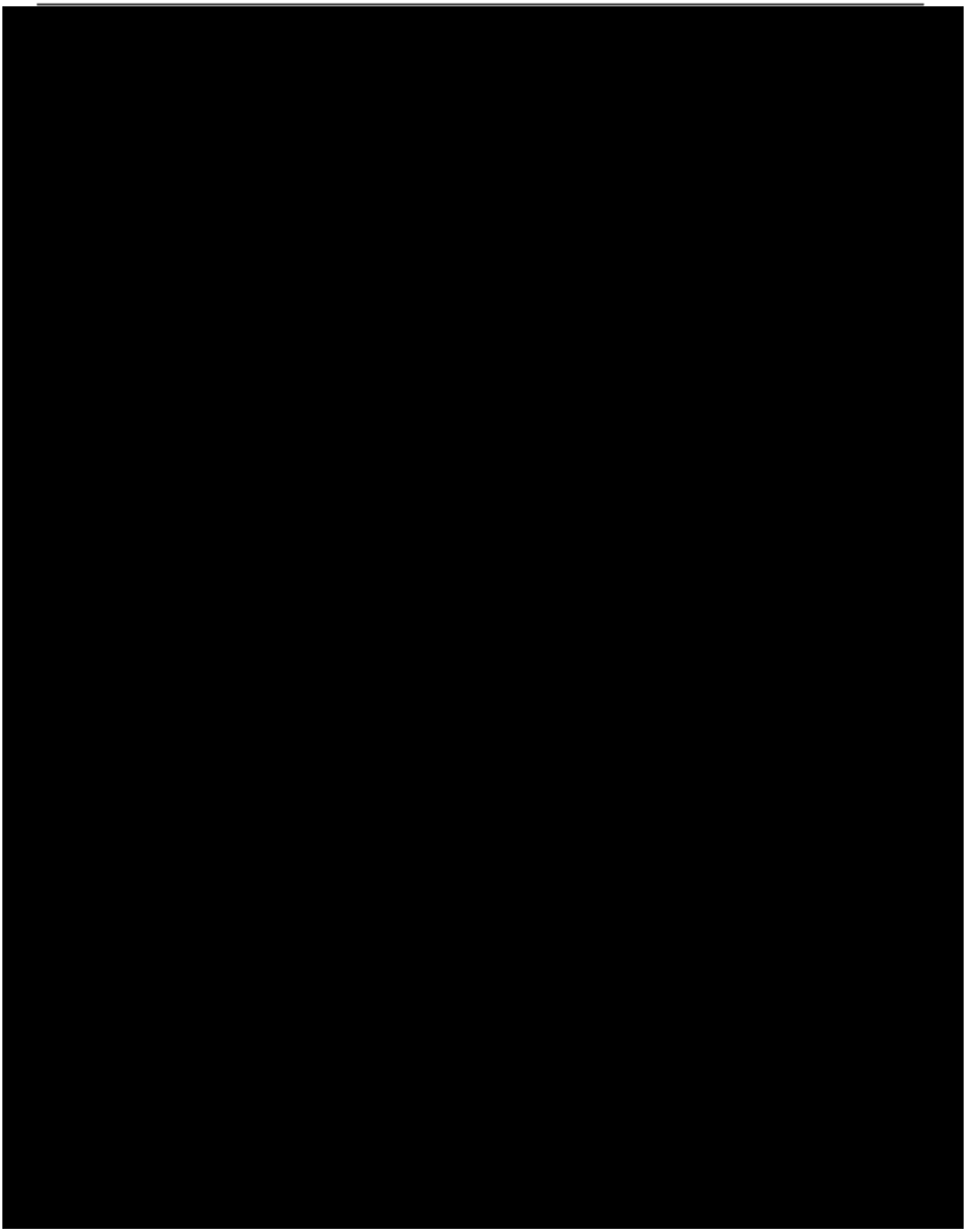
Results In total, 138 records were available for analysis. The average patient age at first referral was 7.7 years (range 1–16 years) and families travelled an average 21.8 miles per appointment (range 0.2–286 miles). Patients attended on average 4.5 appointments per year for 5.8 years. In total, 65.2% had experience of local anaesthetic, 27.5% inhalation sedation and 31.9% general anaesthetic. Dental treatment including restorations and extractions were commonly required on multiple teeth per patient.

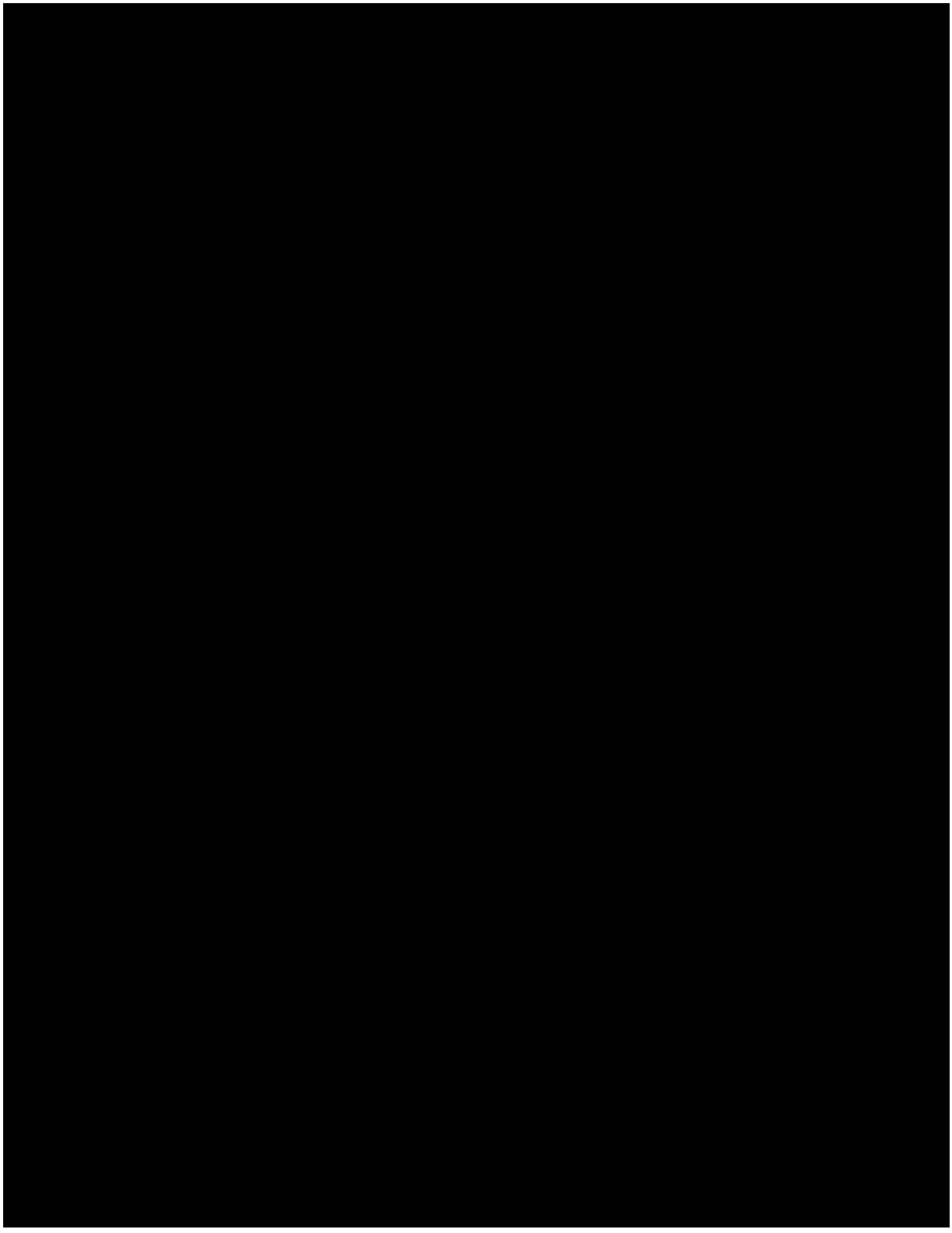
Conclusion AI carries a high burden of specialist dental care to patients and families. Specialist centres are required to provide longitudinal, comprehensive care.

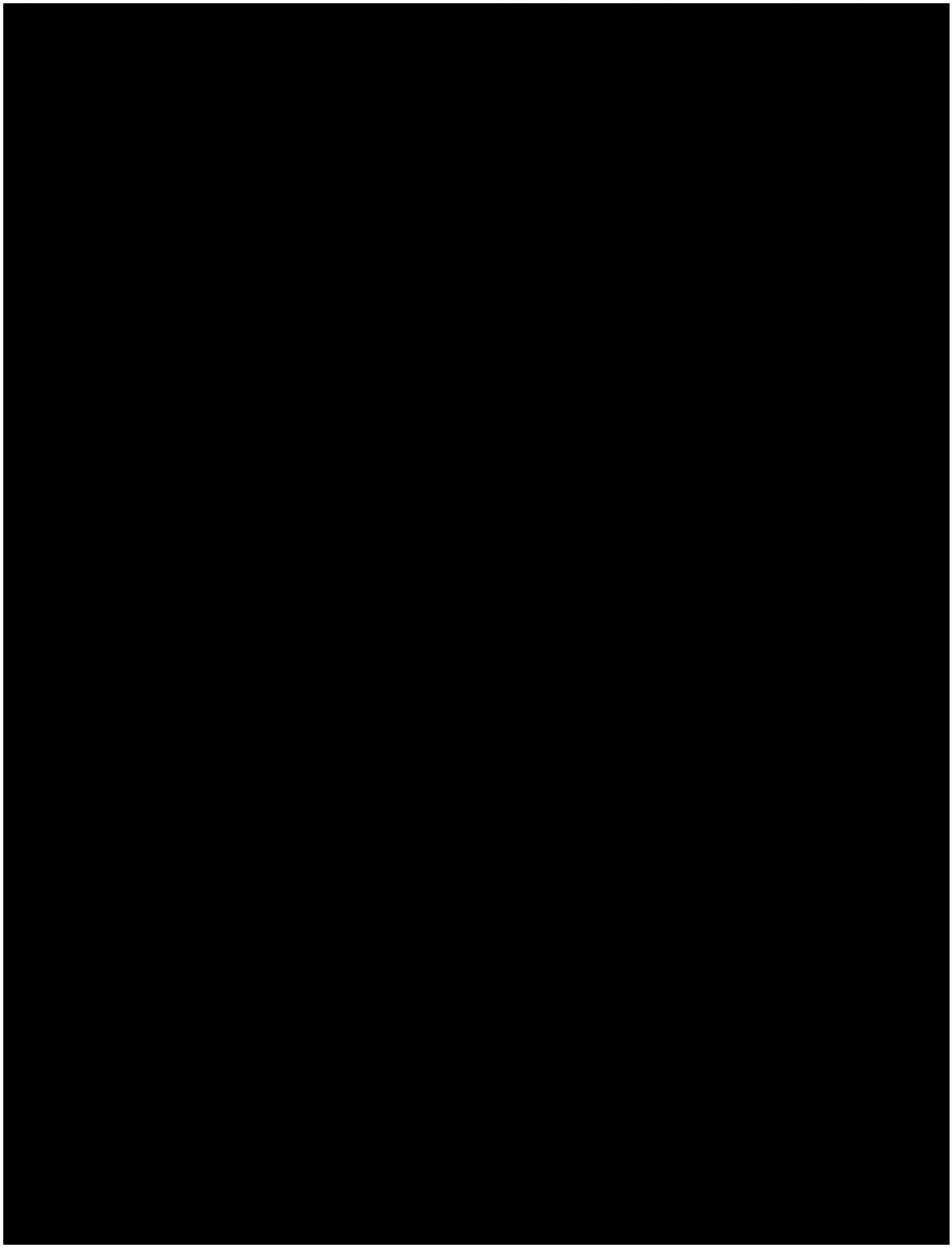
Keywords Amelogenesis Imperfecta · Burden of Care · Paediatric Dentistry

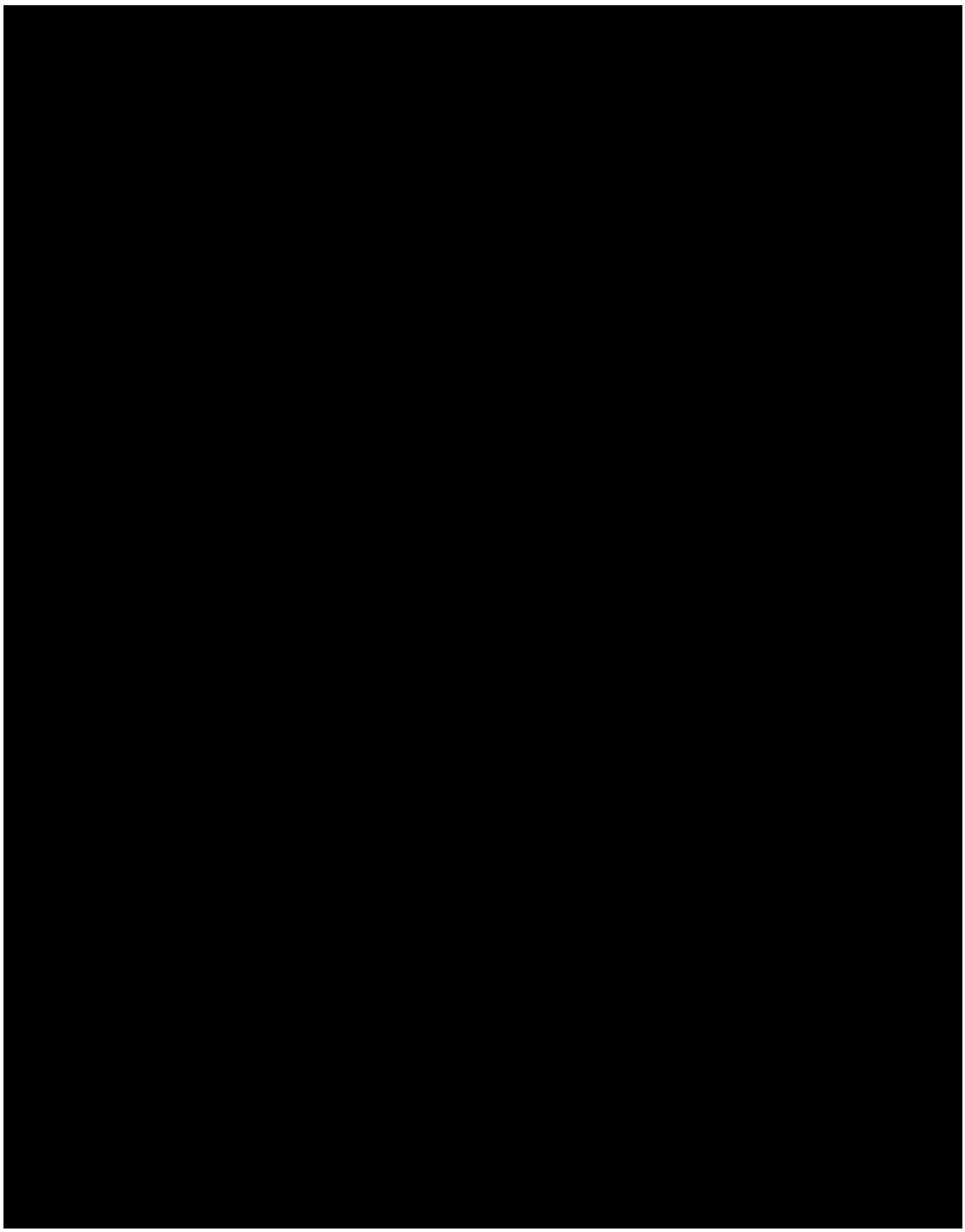
Published online: 19 June 2021

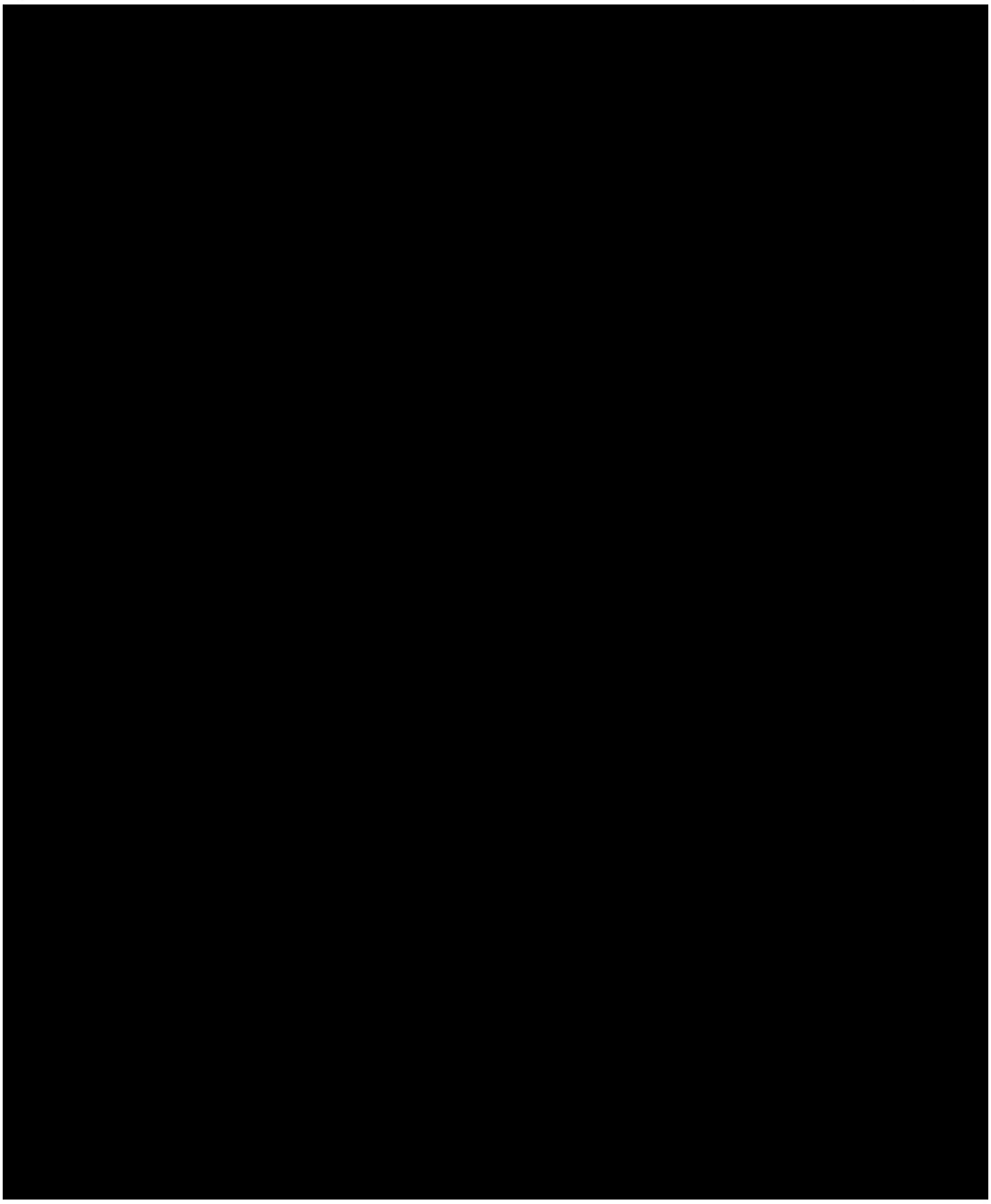


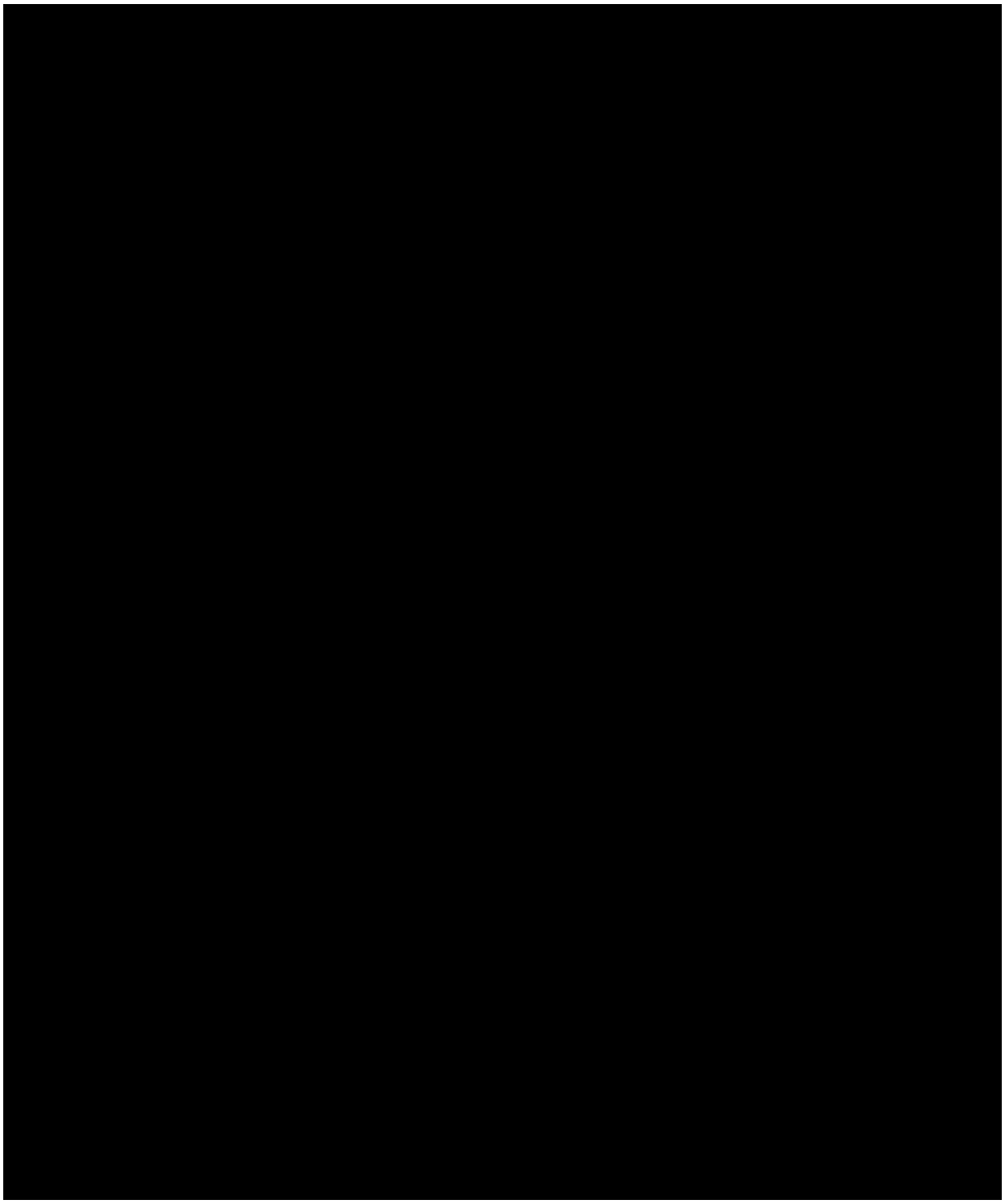












Poster presentation at the 15th congress of the European Academy of Paediatric Dentistry, July 2020.

The burden and impact of Amelogenesis Imperfecta care at the Eastman Dental Hospital

Department of Paediatric Dentistry, UCL Eastman Dental Institute,
256 Gray's Inn Road, London WC1X 8LD • www.ucl.ac.uk/eastman

Husam AL Siyabi , Joana Monteiro , Paul Ashley, Susan Parekh

Introduction: Amelogenesis Imperfecta (AI) is a heterogeneous group of generalised developmental enamel defects affecting both dentitions. Patients may complain of discoloured teeth, tooth sensitivity, difficulties eating and maintaining oral hygiene which can affect their oral health quality of life. The burden of dental care on these children and their families has not been widely reported.

Aim of study To assess the burden of care for child patients with Amelogenesis Imperfecta (AI) attending the Paediatric Dental Department at the Eastman Dental Hospital (EDH).

Materials and methods

Study design: This is a retrospective audit of AI patient records within the Eastman Dental Hospital.

Study population: This audit used existing databases of patients with AI and asking colleagues in the Paediatric department.

Inclusion criteria

- Documented AI definitive or provisional diagnosis in the clinical notes.
- Patients with at least six months care in the service.

Data Collection

- Information from patient records was obtained using a data collection form.
- Data was collated on a spreadsheet and descriptive statistics were produced using Microsoft Excel.

Results

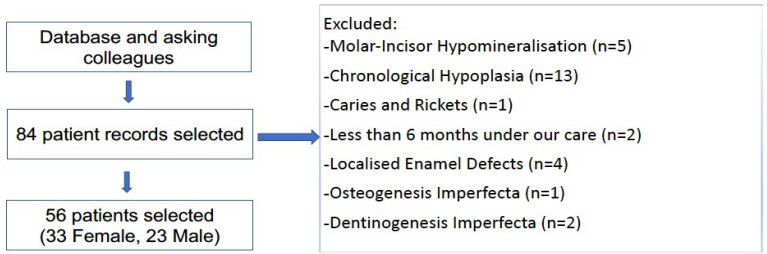


Figure 1 Flow chart : Process of patient record selection for study

The burden and impact of care

- 5 = average number of appointments per year
- 67.5 = average round trip mileage to attend an appointment with a range 8 – 268 miles
- 3.4 years = average duration of care

Results

	Age	Age at 1st appointment	Duration of specialist care	Duration of specialist care (Discharge patients)	Number of appointments / year	Round trip mileage
Average	12.8	9.2	3.4	4.2	5	67.5
Range	4 - 25	4 - 16	6 months – 12 years		1 - 11	8 – 268

Table 1: Demographics data

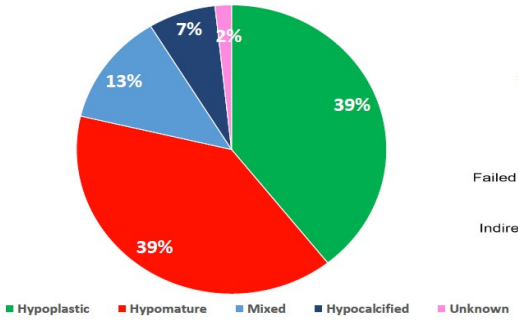


Figure 2: Types of AI as explored by PI

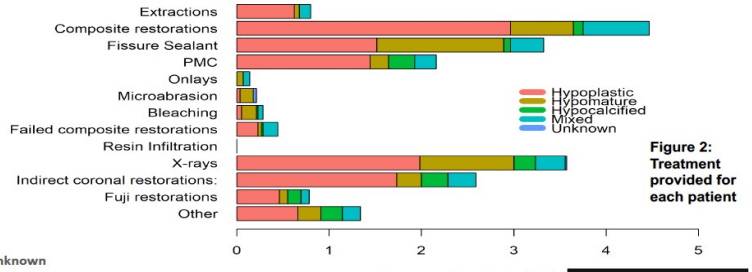


Figure 2: Treatment provided for each patient

Acknowledgements : Lafferty F. and Balmer R at University of Leeds for conceiving the study