

CHAPTER 2

LITERATURE REVIEW

2.1. Role of fluoride on anti-cariogenic effect

Development of caries is known to depend on three factors; susceptible hosts, cariogenic oral microbial floras and fermentable carbohydrate substrates in saliva (Zero,1999). When the factors are imbalance, caries process starts. The dental caries is a dynamic process. The lesion can be demineralization whereas other areas can be remineralization. The demineralization phase starts when organic acid is produced by bacteria after exposure to fermentable carbohydrate. The acid can decrease crystal stability and therefore increases the solubility of enamel (Zero, 1999).

Bacteria such as streptococci and lactobacilli produce mostly lactic acid. Heterofermentative bacteria produces a mixture of organic acids such as propionic, formic, acetic and butyric acid. The acids produced by bacteria result in an increase of hydrogen ion (H^+) concentration. These acids can diffuse into pores of sound enamel and through intercrystalline spaces, interprismatic spaces and defects such as crack. Hydrogen ion attacks apatite crystals of enamel prism (Fejerskov et al.1994; Curzon and Croker, 1978). Mineral phases of enamel begin to dissolve at the attack sites. Consequently, the dissolved minerals raise the degree of saturation of demineralized fluid, as a result the demineralization process stops and reprecipitation of minerals takes place (Marsh 1995).

The formation of subsurface lesions can be reformed by deposition of calcium and phosphate dissolved from substrates and leads to the preservation of the surface enamel. Minerals in the surrounding environment of the tooth can participate in the remineralization process of surface and subsurface enamel. Surface hardness of enamel blocks dropped in early caries lesions formed in laboratory and 20% recovery

was found as a result of brushing with fluoride dentrifice (Zero 1999). Therefore, topical fluoride can prevent caries process at early step. Also it had been shown some occasions that fluoride can effect bacterial carbohydrate fermentation (Arends and Christofferson 1986, Wefel 1990, Bowden 1990).

Calcium phosphates in the form of hydroxyapatite $\text{Ca}_{10}(\text{PO}_4)_6(\text{OH})_2$ are main component in bone and dental hard tissue (Verbeeck,1986). These minerals and many foreign ions that fit in the crystallite structure such as fluoride, strontium, selenium and many other cations can affect the solubility of hydroxyapatite crystalline (Curzon and Crocker, 1978; Duggal et al.,1991, Winter 1990). Enamel is highly mineralized with inorganic content of 96% and also permeable to certain ions so it is susceptible to acid dissolution (Verbeeck,1986). The process is dynamic in nature. When the inorganic material in the tooth surface is loss, the material tries to redeposit. The dental caries lead to the surface loss resulted from chemical reaction by acid dissolution. The cycles of demineralization and remineralization processes are unique and will lead to either cavitation or repair of carious lesion (Silverstone, 1977).

The solubility of apatite mineral is pH dependent. At low pH, calcium and phosphate in the surrounding fluid require more fluoride ions in order to maintain saturation condition. Below pH 5.5, the tooth mineral starts to dissolve. After the pH value crosses the saturation condition, minerals start to reprecipitate. The chemical composition of mineral phase depends on the supply of calcium, phosphate and other ions. One of the most significant findings in the healing cavity is the remineralization process of initial lesion by fluoride. The mechanism is that the fluoride ions incorporate in the apatite crystals and improve the resistance to caries attack. Fluoride ions in the aqueous phase inhibit demineralization and also enhance remineralization.

Fluoride ion (1.36 Å) is smaller than hydroxyl ion (1.40 Å). When hydroxyl ion is taken place, a reduction of unit cell volume occurs and thus makes lattice denser and more stable so that crystal strain decreases and more crystalline apatites are formed (Koulourides et al., 1980; Featherstone et al., 1990). Ogaard et al. (1988) compared

shark enamel (consisted of fluorapatite) with human enamel (contained hydroxyapatite) in high caries model in vivo. They found that shark enamel showed less lesion depth but still had limited resistance to caries attack.

Larsen (1974b) showed that fluoride was crucial for the formation of surface layer during enamel demineralization. The mechanism of fluoride in carious dentin inhibition is twofold. Firstly, fluoride binds calcium and phosphate dissolved from apatite as a result of acid penetration. The precipitation prevents mineral constituents of enamel from leaching away. ten Cate (1983b) found that series of fluoride in the pH ranged from 4-5 relevant to the occurrence of caries. Dentin which normally contains about 5 % of carbonate by weight will double the carbonate content during demineralization and provoke solubility (Featherstone, 1994).

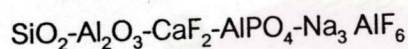
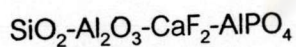
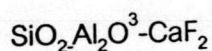
During demineralization process, apatite fraction is dissolved while collagen serves as diffusion barrier. Once the matrix is removed, it can no longer nucleate new apatite crystals. Dentin has double demineralization rate compared to enamel. ten Cate, Damen and Buijs (1998) showed that short term fluoride treatment or continuous uses can inhibit dentin demineralization. A low concentration fluoride has more effect on dentin than on enamel because of smaller crystallite in dentin. Koulouride (1980) found that addition of fluoride in a remineralizing solution can increase the rate of mineral deposition. ten Cate and Duijster (1983a and b), Arends and Chistoffersen (1986), Margolis, Moreno and Murphy (1986) also suggested that the formation of fluorapatite can be anticariogenic and fluoride can be incorporated in the crystal lattice of calcium hydroxyapatite. Fluorohydroxyapatite has lower solubility than hydroxyapatite (HA) because of its higher degree of supersaturation. Kashani et al. (1998) found remineralization process in dentin lesions enriched with fluoride was better than the remineralization process in enamel.

ten Cate and van Duinen (1995) found that dentin with close contact to fluoride releasing material is hypermineralized. Slow releasing fluoride ions have a significant effect on the demineralization and remineralization processes of dentin and enamel.

This result in a shift from negative balance to positive balance. Fluoride also has an antimicrobial effect. Bibby (1940) found that carbohydrate metabolism in pure cultures of oral streptococci and lactobacilli was inhibited by fluoride. Since then, many reports have been published on direct and indirect effects of fluoride on biosynthesis metabolism of oral bacteria (Bowden 1990, Hamilton 1990).

2.2. Fluoride releasing tooth-colored restorative materials, mechanisms and properties

There are four types of fluoride-releasing restorative materials. The first type is designated conventional glass ionomer cement which composes of powder and liquid. The powder is an acid-soluble calcium fluoroaluminosilicate glass (FAS). The types of calcium fluoroaluminosilicate glass may be in the formulations below:



Cement formation is related to glass composition in which $\text{Al}_2\text{O}_3 / \text{SiO}_2$ ratio is required to be 1:2 or more by mass. The liquid was originally solution of polyacrylic acid which may be in the form of a concentrated aqueous solution (40% to 50% by mass). But it was quite viscous and tended to gel over time. The current acid is in the form of copolymer with itaconic, maleic or tricarboxylic acid. When the cement powder and aqueous liquid are brought together, the polyacid reacts to form salt hydrogel which is binding matrix. Then the metal ions are transferred from the glass to the polyacid and this process initiates gelation in the aqueous phase.

There are 5 stages of reaction. The first stage is the decomposition of glass and the release of cement forming metal ions, Al^{3+} , Ca^{2+} and F^- . Secondly, the metal ions migrate into the aqueous phase of the cement. The third stage is gelation (initial

setting). The chain reaction and extension allow for much greater access to polyacid by metal ions leading to setting of the cement. The fourth stage is post-set hardening. When metal ions are increasingly formed by being bound to the polyacid chain, the cement tends to expand under high humidity and susceptible to be damaged by moisture until sufficient hardness has developed. The last stage is maturing (final setting). Over the next 24 hours, the maturation phase occurs during the stage that there are less mobile Al^{3+} ions to be bound within the cement matrix, thus leading to more rigid cross linking between the poly (alkenoic acid) chains. The final set structure is a complex glass core particles sheeted by siliceous hydrogel and bonded together by the matrix phase consisting of hydrated fluoridated calcium and aluminium polyacrylates. A further slow maturation takes place and will last for a few days.

The advantages of glass ionomer cement are fluoride releasing over a long period of time and adhesion to the tooth structure. The most important and clinically relevant properties of glass ionomer cements are working time, setting time, compressive strength, flexural strength and solubility. Glass ionomer cements in the market set rapidly in the mouth (three to eight minutes). They do not represent the fully hardened state when clinicians finish their restorations. The compressive strength is about 120-140 MPa by ISO testing method. The 24 hour flexural strength (Prosser et al. 1984) was found to be between 8.9 to 30.3 MPa which was quite low and showed high variation. That is because the cement properties are affected by its compositions and the temperature during the cement is prepared. The sensitive setting reaction is one of the disadvantages of glass ionomer cement and results in the sensitivity to the balance of water. Thus water contamination can dissolve glass ionomer cement. This cement shows a wide range in properties and also less esthetic compared to resin composite (Akpata, 1996; Shen, 1996).

The second group is resin modified glass ionomer cement which is still consisted of powder and liquid. Mitra (1991) introduced a resin modified glass ionomer cement that combined the fluoride released of a conventional glass ionomer cement and

physical properties of resin based substance by producing favorable physical properties similar to resin composite. The resin modified glass ionomer cement undergoes both polymerization process and acid-base reaction. The powder is comprised of ion leachable glass (FAS) similar to glass ionomer cement and initiators for light or chemical curing.

The basic composition of liquid part is polycarboxylic acid or polyacrylic acid with some carboxylic group modified with methacrylate and hydroxyethyl methacrylate (HEMA) monomer and water. Those monomers are responsible for polymerization. This type of cement was introduced because of its ease of manipulation and less sensitive to moisture while setting. The basic setting reaction of this material is similar to that of the conventional glass ionomer cement but it has additional cross-linking through methacrylate formation. The powder and liquid are mixed and hydrogen ions in the liquid attack the glass surfaces. The metal ions released from the glass particle react with polyacrylic acid while HEMA cures concurrently. This reaction forms a hardened mixture and at the same time the cross-linking can be initiated by light and/or chemical catalysts.

In the light activated materials, the cross linking of methacrylate group progresses rapidly. Water sensitivity could be reduced by incorporation of photopolymerization reaction and rapid setting also provides better color stability. The advantages over the conventional glass ionomer cement are improved setting characteristics, significantly working time and rapid development of early strength that in turn renders set matrix to be less intolerant to moisture. The advantages of this resin modified cement are ease of manipulation, greater strength and also improved esthetics (Tay, 1995). The disadvantages are its mixing procedure, the depth of cure susceptible to degradation and also lower strength compared to resin composite (Shen, 1996).

The third group is called polyacid modified resin composite or Compomer. It composes of carboxylate methacrylate resin and FAS glass fillers. The polyacid modified resin composite is a combination of glass ionomer glass powder or prereacted

glass-ionomer with a polymerizable acidified monomer (Barnes et.al.,1995; Swift and Vann,1995; McLean and Wilson,1994). Polyacid modified resin composite is similar to resin composite in its chemical structure and reaction, ion-leachable glass particles and polymerizable acidic monomer. The importance of mixable polyacid modified resin composite has significantly decreased. The major advantages of this material are excellent handling characteristics, moderate mechanical properties and ability to release fluoride (Christensen, 1997).

de Araujo et al. (1996) found that there were fluoride-releasing characteristics of polyacid modified resin composite (Dyract LD, Caulk, USA) and Miller(1995) also found fluoride approximately 25 $\mu\text{g/ml}$ and after 28 days fluoride release was still maintaining at approximately 6 $\mu\text{g/ml}$. Fluoride released from polyacid modified resin composite is less than resin modified glass ionomer cement because initially there is no water in the composition. Water must diffuse from the environment through the limited hydrophilic matrix to liberate ionic components from glass fillers (Meyer, Cattani-Lorente and Dupuis,1998). Although fluoride release is less but polyacid modified resin composite is still good because of its adequate properties. The only drawbacks are lower wear resistance compared to resin composite and less fluoride release than glass ionomer cement.

The forth group is a new release of resin composite with fluoride releasing properties. The weak point of this type of material is a very low amount of fluoride ions can be released. This is because the addition of the polymer matrix-forming material with the soluble fluoride results in the dissolve in water that diffuses into the material. As a result, fluoride salts are washed out and leave the porous structure thus decreasing physical properties of the material. The acrylic-amine-HF salts have been considered to produce fluoride releasing system. The disadvantages of this material are poor stability and discoloration. These poor properties prevent them for their common uses in restorative dentistry. However, this type of material could be used in adhesive composites for orthodontic brackets and fluoride releasing sealants. This material still

requires extensive development and clinical testing before they can be considered for general uses (Combe, Burke and Douglas, 1999).

The release of fluoride from resin composite was also demonstrated (Arends, Reintsema and Dijkman, 1988, Arends and van der Zee, 1990b). Some resin composite materials exhibited slow fluoride release and resulted in reduction of recurrent caries. Moreover Zimmerman, Rawls and Querens (1984) suggested that a source of fluoride ions was beneficial in terms of protection and capable of barrier. Hattab, Mok and Agnew (1989) assessed various restorative materials in terms of caries resistance using polarized light microscopy. After generating caries like lesions around the restored teeth in vitro, some restorative materials were found to have an ability to inhibit the initiation and progression of caries like lesions. The fluoride releasing materials such as glass ionomer cement and polyacid modified resin composite showed caries inhibition effect and the degree of caries inhibition depended on the type of fluoride releasing materials. (Glasspoole and Erickson, 1993; Dunne et al., 1996; Tam, Chan and Yim, 1997; Dionysopoulos et al., 1998a and b; Millar et al., 1998).

There were many investigations proposed on the inhibition of secondary caries by the glass ionomer materials. Dunne et al, (1996) showed caries inhibition by submerging teeth restored with resin modified and conventional glass ionomer cement in acid gel and then photomicrographed in 14 days. The evidence of caries inhibition on the surface of enamel and dentin was found but there was no significant difference between the two materials. The same result of caries inhibition by glass ionomer cement was also confirmed (Nagamine et al., 1997; Tam et al., 1997; Tanbirojn, Douglas and Versluis, 1997). They all proposed that the glass ionomer cement and resin modified glass-ionomer cement had inhibitive effect on the secondary caries.

2.3. Assessment of the effect of fluoride releasing materials on human enamel and dentin

2.3.1. Hardness test

Hardness test is one of general testing methods. Hardness is the ability of material to resist a permanent indentation on a test surface (Craig, 1993). Hardness changes of the tooth surface may indicate the degree and extent of the carious lesion. Various studies have measured the microhardness of sound dentin or remineralization (Craig and Peyton, 1958; Ryge, Foley and Fairhurst, 1961; Arends, 1980b; Herkstroter, 1989). The hardness test is categorized into two groups, macrohardness (over 1 Kg) and microhardness (less than 1 Kg). The hardness tests commonly used are Brinell, Knoop, Rockwell and Vickers depending on the test materials. In general, hardness can be an index of mechanical strength. The hardness measurement is not expensive. It is also non-destructive and easy to perform. The hardness test evaluates the resistance to deformation of material subjected to a point force and multiple tests can be carried out on a small sample.

2.3.1.1. Microhardness test

Microhardness test or another word microindentation hardness test is commonly used to measure the surface properties of dental materials as it is a simple, reproducible and also non destructive test (Boyer, 1987). The usual unit of hardness is force per unit area of indentation e.g. Kgmm^{-2} or GPa. The commonly used microhardness tests for teeth are Knoop and Vickers tests. The difference between Knoop and Vickers hardness test is the shape of indenter. The Knoop indenter is a diamond pyramidal shape. The indenter produces a rhomboid shape and the dimension of major axis is measured. The lengths of major axis diagonals are measured and reported as Knoop hardness numbers (KHN). The KHN can be calculated from this following formula

$$\text{KHN} = P/C^2$$

Where P is the load applied, l is the length of long diagonal of indentation, C is a constant relating l to the area of the indentation. The unit for KHN is kg/mm^2 (Boyer, 1987; Ekstrand, 1993)

The Vickers hardness test uses 136 degree diamond pyramid indenter which produces a square indentation. The test reports hardness as Vickers hardness number (VHN). VHN is the ratio of load applied to the surface area of indentation calculated from the following formula

$$\text{VHN} = 2 P \sin(\theta/2/d^2) / F \times 1.89 \times 10$$

When P is the load applied, d is the average length of two diagonals and θ is 136. The Unit is kg/mm^2 . Both Knoop and Vickers hardness tests are prone to error from the optical measurement (Boyer, 1987)

The hardness of enamel and dentin has been studied by many investigators using various techniques. The conventional quasi-static test has the limitation of measuring the mechanical properties of enamel and dentin. These are due to the complex structure and the small tooth specimen size. Microhardness indentation techniques were then introduced. The microhardness tests were carried out by uses of spherical indenters (Renson and Bradon, 1975), Vickers indenters (Ryge et al., 1961) and Knoop indenters (Craig and Peyton, 1958; Davidson, Hoekstra and Arends, 1974) and a range of hardness of enamel and dentin was reported. Knoop hardness test has been the most popular method to investigate surface hardness of tooth-colored restorative materials. Some other material properties such as Young's modulus and fracture toughness can also be calculated by this technique (Meredith et al., 1996). Nevertheless, the applied load ranged from 10 to 500 g and the indentation depths from 1 to 10 μm of microhardness tester are too large to investigate some small areas such as small filler particles or peritubular dentin (Willems et al., 1993a).

2.3.1.2. Nanohardness test

The applied load range of 10 to 500 gram of microhardness tester can produce indentation depths of 1 to 10 μm . These indentation depths give large indentation areas that are sometime oversize the proposed testing area of tooth structure (Willem et al., 1993a). Recently, the nanohardness test on the other word a ultra-micro indentation hardness was introduced with the initial force of less than 0.1 mN and the penetration depth can be as small as some nanometers (nm). The new indentation technique is able to investigate the mechanical properties of enamel and dentin in a very small area. This system measures the penetration depth of the indenter by using depth sensing method and then the depth is automatically transformed into area as the indenter geometry is known. The nanoindentation system does not rely on eye measurements. Moreover, the system measures the depth of the actual penetration rather than the size of impression after elastic recovering when using microhardness test (Willem et al., 1992a ; 1993a). Surface hardness and Young's modulus of enamel and dentin can be precisely calculated at some particular areas where the microhardness tester is not possible. Time-consuming precondition of specimen surface is the most important. A well polished surface is necessary and will prevent the indenter from striking surface irregularities. Thus analyzing software will calculate the hardness and the Young's modulus without error.

The nanohardness test is developed to measure near surface properties with nano meter resolution. This testing unit is called UMIS 2000 (The Commonwealth Scientific Industrial Research Organization CSIRO, Sydney, Australia). The operating system of the UMIS machine is totally controlled by computer. Hardness measurement is carried out by the calculation that relates the depth of penetration, applied force to the actual geometry of the indenter. The indenter used is a triangular indenter named a Berkovich indenter (Synton BA, Switzerland). It is a diamond tip with an equilateral triangle base of 65.3 degree face angle to the axis. The diamond tip is the most common used indenter in the determination of hardness because of its great stiffness

and hardness. This triangular indenter or Berkovich indenter is more favorable to the four-sided Vickers or Knoop indenters because it is easier to obtain the sharp tip. The 65.3 face angle geometry is preferred because it provides the similar depth to Vickers indenter that is because geometry is equivalence (Bell 1991/1992). The hardness is defined by the calculation between depth and the indenter load (P) given by this formula

$$H = P/A$$

where A is the contact area and given by

$$A = Kh_p^2$$

where h_p is the depth of penetration and K is a geometric constant which the hardness can be calculated with the indenter geometry, maximum force and the value of h_p (for a Berkovich indenter is 24.5) ($K = \sqrt{3} \tan^2 \theta$, where $\theta = 65.3^\circ$)

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2.3.2. Physical method

In order to physically analyze the composition of any structure, there are many physical methods such as Auger electron spectroscopy, atomic absorption spectroscopy, Raman spectroscopy. X-ray analysis is one of them and it can be performed quickly. The X-ray analysis also requires minimal specimen preparation and the procedure is non destructive to the sample (Postek, 1980). General of X-ray requires the removal of an electron from the inner shell of an atom. There are two ways of removing the electron from the shell. Firstly, electromagnetic radiation (X-ray from an X-ray tube) is used to scatter specimen electrons. Then, scatter specimens in the scanning electron have wavelengths and energies that are characteristic of elements in this specimen. Element evaluation using two different X-ray analysis devices are called WDS (wavelength dispersive spectrometer) and EDS (energy dispersive spectrometer).

Energy dispersive spectrometer (EDS) is one of analytical tools that permits a localization of chemical analysis with the micron range resolution. Fitzgerald, Keil and Heinrich first introduced X-ray detector mounted on an electron probe microanalyzer (Goldstein 1994). As EDS is one type of electron probe microanalysis, it detects the characteristic wavelengths of X-ray spectrum emitted by a solid-state sample which is bombarded by electron beam. X-ray photons from the sample pass to a computer analyzer, where the data is displayed as histogram of intensity by voltage. The EDS is well recognized and it provides useful information.

The advantages of EDS are compact, low cost and rapid. But there is a limitation on X-ray analysis. The lower limit is the light elements which are less electron dense. There are fewer collisions between primary electron and specimen electron and thus fewer X-rays are generated (Postek 1980).