Fall 12-11-2017

# The Analytical Methods Used to Detect Fluoride in Dental Pharmaceuticals

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Bachelor of Arts: Chemistry

Honors 4998

11 December 2017

## INTRODUCTION

Access to care is a widespread issue in dentistry, especially in the United States. The underserved communities in cities such as Detroit are particularly battling this issue, contributing to further oral health problems. This issue among many others impacts the oral health of individuals of all ages.[1]

The oral cavity is exposed to various forms of bacteria through the consumption of food. Teeth are crucial in breaking down food, that is consumed, into smaller pieces that are more easily digestible for humans. However, most food that is ingested contains harmful components that can ultimately deteriorate structure of the teeth.[2] The oral cavity contains hundreds of different types of bacteria, both good and bad. Dental plaque is a thing, colorless, sticky film of bacteria that is present on the surface of teeth.[2] The consumption of various types of foods and drinks cause the bacteria to turn the sugar and starch components into acid, which eats away at the hard outer surface of teeth. The enamel is composed of calcium phosphate crystals (hydroxyapatite).[3] After consumption of various foods, the acidity in an individual's mouth increases due to the continual production of acid. The acid that is produced reacts with the hydroxyapatite in the tooth enamel and causes damage to the structure of the tooth. When teeth are constantly exposed to this acid, the enamel begins to lose minerals, sometimes causing the appearance of a white spot. This defines the signs of early tooth decay.[2] As this process continues, more minerals are lost and the enamel eventually becomes weak and/or destroyed leading to the formation of a cavity. Often times, dental caries or cavities are the first signs of tooth decay. A cavity is damage to the tooth that is irreversible and must be corrected by a dental professional, often times with a composite restoration. When fluoride is present in the oral cavity, it substitutes to form hydroxyfluoroapatite which is less soluble than hydroxyapatite under acidic conditions, preventing a great loss of minerals from the enamel.[4] Fluorapatite, due to its acid resistant nature, allows for the incorporation of layers of fluoride onto tooth enamel. This strengthens the enamel, causing a decrease in pH in order to prevent demineralization of the outer surface of the teeth.[4] Tooth decay continues to be one of many prevalent issues in oral healthcare. It is known to be the most common and most expensive oral health problem in both children and adults. This disease is one of many causes that lead to the loss of teeth beginning from early childhood and continuing throughout adulthood.[2] Various forms of fluoride are beneficial in prevention of this disease, allowing for better preservation of tooth enamel. Many studies have been completed in an attempt to determine the amount of fluoride necessary to prevent tooth decay and the various benefits of fluoride. Each study differs with respect to factors such as cost, availability of materials, complexity of instrumentation, etc. and the methods and results of each will be further discussed.

Preventive dentistry, a sector of dentistry aimed at caring for and keeping teeth healthy in order to avoid disease, is significant when discussing tooth decay and caries. It is common knowledge that each individual is required to brush their teeth a minimum of two times a day for at least two minutes each time. In addition to brushing, it is equally as important to floss and use mouthwash, however, the majority of people do not practice this daily habit. The group of individuals who do practice normal preventive techniques are not necessarily aware of the "proper" technique and much of this is due to dental professionals not properly educating patients about the correct methods. While children who are in school are required to receive dental checkups regularly due to the "School Health Law," adults receive dental checkups less consistently, most often depending on the awareness of the importance of oral health in the workplace. A study was done by one dentist and three dental hygienists on the employees of an airline company as requested by the associated health insurance group in order to compare the differences between dental checkups that incorporate tooth-brushing instruction (TBI) verses those that do not (conventional dental checkups).[5] Over a span of five years (2001-2005), 3854 patients received checkups, 284 of which occurred annually. A specific routine examination was given for each patient. First, pre-examination brushing was done prior to an oral exam performed by the dentist. After the oral exam each patient received a tablet they were required to chew in order to stain the remaining dental plaque. After recording the results, the dental hygienist gave the patients instructions on properly brushing their teeth. This normally required 10-15 minutes per patients and required the use of a hand mirror in order to insure proper understanding. The recommendation of using floss and various other supplementary devices was given in addition to the explanation of the destructive affects of periodontal diseases. At the end of each instructional session, the dentist kept a record of the patient chart and also gave a copy to the patient so they were also able to review details and follow proper dental hygiene instructions. Follow-up appointments were highly suggested for patients whose charts showed scores greater than or equal to sixty percent, which demonstrated high plaque levels. Scores were categorized according to O'Leary's plaque index as less than or equal to thirty percent as being good plaque control, greater than thirty percent but less than sixty percent as moderate control and anything greater than or equal to sixty percent as poor plaque control.[5] After completing the five-year period, it was seen that patients who received checkups every year, which was about thirty-seven percent of overall patients, were seen to have good plaque control. Additionally, the number of patients who demonstrated less than thirty percent of plaque present increased over time. Patients who received tooth-brushing instruction and applied it to their oral health routines scored significantly higher (according to O'Leary's plaque index) during their oral examinations, meaning the percentage of plaque decreased over time. This illustrated the effectiveness of the program and allowed for an increase in oral health awareness among individuals. In turn, improved preventive dentistry techniques were indicated.

## BACKGROUND

Fluoride ions are a form of fluorine, an element that is abundant in the earth's crust. Its main form is a gas, however, this gas never occurs in its free state in nature but it exists along with other elements as a mixture. Minerals found in rocks and soil in the earth are composed of this fluoride compound and when they are exposed to water, the fluoride component dissolves and fluoride ions are discharged leading to the trace amounts of fluoride found in water. Various water sources contain different concentrations of fluoride based on how deep the water is and how much fluoride-containing materials are surrounding it. Lakes and rivers are examples of surface bodies of water that contain minimal amounts of fluoride. For example, Lake Michigan's fluoride level is 0.17 ppm while in other parts of the United States, the amount of fluoride ranges from very little amounts to just over 4 ppm.[2] Larger bodies of water, including oceans, have fluoride levels ranging between 1.2 to 1.4 ppm.[2] Fluoride compounds are found in food and drinks that are consumed daily, however, the concentration of fluoride found in these items varies.

Fluoride is an important aspect of dental care and has been widely used for several years. Fluoride protects teeth in two ways- systemically and topically.[2] Systemic fluoride is what we ingest, it is found in things such as fluoridated water, fluoride tablets/drops, and in food and beverages. As a child, when teeth are being formed, the fluoride that is consumed becomes part of the tooth structure. This intake of fluoride provides protection that is more efficient than that of fluoride which is applied topically by a dental professional.[2] This is because systemic fluorides are ingested and can be found in saliva which is continuously surrounding the teeth, providing them with an ample amount of fluoride that is incorporated into the tooth, preventing tooth decay.[2] Topical methods of incorporating fluoride into one's oral cavity are done after teeth have completely developed in the mouth (posteruptively).[2] Fluoride is integrated onto the surface of the teeth resulting in an increased resistance to tooth decay. Topical applications of fluoride include application from various toothpastes, mouth rinses, and fluoride foams which are applied by a hygienist. During the recommended biannual cleanings done by a hygienist, topical fluoride is provided after a cleaning is completed. However, only children receive this application of topical fluoride because it is important to keep their young teeth healthy and strong. Children are less likely to brush their teeth twice a day and also less likely to do so properly. Adults receive this form of topical fluoride, however, not as frequently, so they must use toothpaste and mouthwash that contains fluoride in order to remove plaque as much as possible. Fluoride incorporated in the oral cavity, both systemically and topically, provides a few of many benefits to an individual's oral cavity further causing benefits to overall health.

The controversy regarding the benefits of fluoride in dental hygiene products has been prevalent for over fifty years. Beginning in the early 1940s, dietary fluoride supplements were tested for their effectiveness in the reduction of caries in the oral cavity. The study found that caries reduction was significant in both initial and permanent teeth.[6] This conclusion was then tested in an experiment performed by Arnold et al. in which concentrated sodium fluoride solutions or tablets were placed in drinking water of the participants. This was estimated to be about 1.0 mg of fluoride intake daily. This experiment was proposed two years' prior by the

American Dental Association. It required dissolving a 2.2 mg sodium fluoride tablet in one quart of water. This fluoridated water was then used to prepare food and as drinking water for children under two years of age. Older children received the addition of 1.0 mg of fluoride to their water and juice daily while children between the age of two and three years received 1.0 mg of fluoride every other day.[6] The Council on Dental Therapeutics (CDT), when analyzing the previous methods of the delivery of fluoride to adults and children, concluded that the delivery of fluoride is most effective during the period of tooth development. It was recommended that fluoride be distributed to children until the age of ten, which is roughly the time when the growth of the enamel surrounding the second molars is complete.[6] In the 1960s and 1970s, the CDT recommended creating water with fluoride for young children to consume. The ADA suggested many alternatives to lower the 1.0 mg of fluoride every other day for children three years of age or younger, however, dental professionals still believed fluoride was necessary for consumption regardless of the opposition's view. In 1972, the American Academy of Pediatrics included fluoride in the category of nutrients, encouraging physicians to prescribe a necessary amount of fluoride for intake daily.[6] Continuing on through the rest of the 1970s, various studies and experiments were performed in order to perfect the amount of fluoride intake for adults and children starting as young as twenty-four months. In 1977 a proposed schedule was made which allowed for supplementation beginning at birth and dividing the intake between various age ranges over a span of six years and not providing any fluoridation beyond that period.[6] Many alternatives to this schedule were presented in the late 1970s and some of which included plans for the developmentally disabled, being that they were at higher risk for dental caries then other individuals. Differing views were presented regarding the intake of fluoride due to the complex methods presented to professionals. Although many oral benefits are presented, the question of fluoride digestion and impacts on overall health are still present. From the time of the schedules and methods of proposal for necessary amount of fluoride intake in the 1970s until current day proposals, methods and recommended dosage is still varying.

The components of various toothpastes have progressed over the years, allowing for better preservation of oral health. In ancient times dentifrice (toothpaste/powder) was mainly used as a cleaning agent, but its use has recently advanced, incorporating fluoride into its formula. This change has permitted a significant improvement in oral health, globally.[7] The first toothpastes that contained fluoride were ineffective due to the interaction of calcium-based abrasives with fluoride. These abrasives caused fluoride to become insoluble and ineffective in toothpaste.7 Prevention of caries (cavities) was unable to occur because fluoride must be freely soluble in order to control the progression of cavities over time. In order to incorporate useful fluoride in toothpaste, an effective fluoride was developed, sodium monofluorophosphate salt. This salt ionizes in salivary fluids allowing for the release of the monofluorophosphate ion further causing the fluoride to covalently bind to the phosphate group. This prevents the reaction with calcium ions from the abrasives and causes the incorporation of fluoride into the oral cavity in order to replenish and protect it.[7]

A student at Harvard University conducted research regarding the regulation of toothpaste in 1997, further defining the distinct differences between a dental “cosmetic” and a dental” drug.” [8] Dental hygiene products used in maintaining good oral health such as toothpaste, mouth wash, fluoride foams, etc. are regulated by the Food and Drug Administration (FDA) in order to protect individual health.[8] Individuals are accustomed to simply going to the store and having the option to choose between various brands and types of dental hygiene products, however, the process which allows these products to reach the market is complex. The type and amount of certain ingredients is regulated as well as the labels and various other components of the products.[8] There is a small distinction between something that is a drug as opposed to a cosmetic. Dental hygiene products fall under the drug category due to the presence of fluoride. This classification is used because fluoride is incorporated as an “anti-cavity” component of dental products and is responsible for the long-term change in the structure of teeth. Also, many products regulated by the FDA that are considered drugs are used by consumers for their long term pharmacological effects, just as fluoridated dental hygiene products are. Although many toothpastes consist of the same ingredients in the same combinations, many new manufacturers have incorporated new ingredients in order to distinguish their products from the various other brands, allowing them to succeed in the market. Prior to releasing a product to the market, the FDA considers factors such as the size of the package, type of package, necessary ingredients, and warnings issued for the product. Safety, ingredients, effectiveness, toxicity, and other factors contribute to the strict regulation of fluoride by the FDA.[8] Although the issue is controversial with regards to how safe fluoride really is for adults and most importantly young children, there have been various methods studied in order to educate individuals about the “safe” levels of fluoride for all ages.

Water fluoridation is one of many effective methods used to help prevent dental decay. Various studies have revealed the positive outcomes of fluoride in preventing cavities and tooth decay in children and adults.[2] The first city in the world to have a fluoridated water supply was Grand Rapids, Michigan. A study indicated that children who consumed this fluoridated water had 50-63% less tooth decay than children from various other cities who did not consume fluoridated water.[2] Over the last several years, the consumption of bottled water has increased in the United States. Consequently, individuals who consume bottled water as their primary source are at a higher risk for tooth caries and decay than those who consume water from a community water supply.[2] Bottled water does not contain the necessary amount of fluoride a person is recommended to consume in order to protect their oral cavity. However, the amount of fluoride in different brands of water varies, some containing more than others. For example, data was conducted in 2005 by USDA that collected the amount of fluoride in different brands of bottled water. The average amount of fluoride present in twelve different brands of water was 0.11 parts per million (ppm). Brands such as Propel Fitness Water had fluoride levels as low as 0.02 ppm and other brands such as Perrier had fluoride levels as high as 0.31 ppm.[9] With the issues many cities are battling with having a clean water supply, people are opting to use bottled water more than before. This is contributing to the presence of tooth decay in the form of caries in younger generations. People may state that drinking fluoridated water may increase chances of diseases such as cancer, inhibition of human enzymes, decrease thyroid gland activity, etc., however, there has been no scientific evidence that proves such correlation is present. Studies performed at the National Cancer Institute analyzed the causes of death of over two million patients. No findings had any correlation with the patient's consumption of fluoridated water. Additionally, when considering enzyme levels, it is shown that consumption of fluoridated water has no influence on human enzyme activity. There are two primary mechanisms that maintain a stable concentration of fluoride in the human body. The rapid removal of fluoride is done by the kidneys and calcified tissues are responsible for the intake of fluoride. Another study was conducted in order to compare the affects of the intake of fluoridated water on the thyroid gland. The first group consumed water with extremely high fluoride levels of 3.48 ppm while the other group consumed water with extremely low fluoride levels of 0.09 ppm.[2] The individuals which were tested in different communities had been residents of that community for ten or more years.

This allowed for the conclusion that ingestion of fluoride for long periods of time has no effect on thyroid gland diseases. Although differing beliefs may still be present, tests performed by national institutes for prolonged periods of time suggest the lack of hazards associated with the consumption of fluoridated water.

A genetic study exploring the actions of fluoride resistance or susceptibility was conducted by the University of North Carolina School of Dentistry in 2011. This experiment required the use of fluoride concentrations of about 400 μg/mL in order to isolate fluoride-resistant mutants of *Caenorhabditis elegans.* Studies of these mutants have allowed for the identification of fluoride-resistant (flr) genes, which include *flr1*, *flr3,* and *flr4*. Each gene was classified with specific plasmids or activities.[10] Additional studies were performed using inbred strains of mice and focusing on the development of tooth enamel development and bone homeostasis. Inbred strains of mice are used in genetic studies because of their heterogenicity between inbred strains and isogenicity within individual strains. The diversity that is present between these strains of mice is beneficial for testing things such as cancer susceptibility, obesity, blood disorders, aging, susceptibility to infectious diseases, neurosensory disorders, and atherosclerosis. The enamel in mice is similar to that of human enamel allowing for experiments to be accurately measured. However, humans and mice differ in the fact that mouse incisors erupt continuously allowing for the facilitation of fluoride's effects on the development of tooth enamel at any time throughout the life of an animal. It was seen that both environmental and genetic factors had similar influence on mineralization of the teeth. Fluoride metabolism differs between the mouse strains, some having the capability to retain water more than others. Despite these differences being present, the resistance to dental fluorosis remains constant. Dental fluorosis is a detrimental defect of tooth enamel that is due to an overexposure of systemic fluoride during periods of enamel formation, mainly in children. Complex traits are under the control of multiple genes, some being genetic while others are environmental. The quantitative trait loci (OTL) contribute to the discrepancy in phenotypic traits which can in turn be mapped using traditional genetic methods. The mice are crossed in a two-generation cross which are varied in the fluoride distributed to one strain as opposed to the other. Recently, many genome-wide contributions have been associated with studies in humans.

## **ANALYTICAL METHODS**

Analytical chemistry methods have been used to determine the various types of fluoride in toothpaste. Being that fluoride is now more widely accepted as a component of toothpastes, several forms have been incorporated in dental products over the years. Among many of the fluorides used are stannous fluoride, sodium fluoride, amine fluoride, and sodium monofluorophosphate, all of which are biologically and chemically unique.[11] Stannous fluoride was the first type of fluoride used in toothpastes in the United States at the end of the 1960s. Sodium fluoride and stannous fluoride supply bio-active fluoride due to their high solubility in water. Amine fluoride on the other hand provides bacterial protection in order to reduce plaque on teeth. It further dissociates in water to fluoride anions and an organic cation. [11] Due to the fact that toxic and therapeutic fluoride are present simultaneously, a method is required to determine the dissolved, ionic, and total fluoride concentrations of each in items such as toothpaste. This allows for the determination of storage properties, incorporations of specific healthcare requirements, and controlling of specific components. Two steps must be considered when analyzing the chemical components of toothpaste. First, total fluoride is considered in order to control the quantity in any given formula. Then, the bioactive free fluoride available in the suspension is examined because it determines the efficiency of cavity prevention.[11]

Various methods have been used to determine fluoride in toothpastes such as fluoride ion selective electrodes (F-ISE) and ion chromatography (IC).[11] Fluoride ion selective electrodes are easy to use, cost effective, and sensitive. The pH required to run this reaction is between 5.2 and 5.5 in order to avoid the interaction with $OH^-$ and further conversion of $F^-$ to HF and $HF_2^-$. Additionally, the samples must have the same ionic strength and must avoid interference of ions that form precipitates with fluoride.[11] The conditions for pH and ionic strength can be controlled more effectively with the addition of the total ionic strength adjustment buffer (TISAB) to neutral toothpaste. However, it is more difficult to control the interference of ions that form precipitates because many toothpastes consist of Al, Si, Ca and many water-soluble organic compounds that form precipitates with fluoride.[11] Ion chromatography allows for the determination of several fluoride ions in one sample. Unfortunately, it has a lower sampling rate and a higher cost when compared to F-ISE and also requires the use of particle-free samples causing the mandatory use of a 0.15-0.45 micrometer filter.[11] Lower conductivity detection is highly recommended for ion chromatography because the ion is weak and the matrix components such as bicarbonate can cause disturbances and produce a large peak.[11]

An experiment was performed in order to investigate the concentrations of fluoride present in different toothpaste brands in varying forms. Colgate Total and Colgate Max (each available in concentrations of 70%, 100%, and 130%) were prepared by the manufacturers for this specific investigation and Eurodent, Elmex, Signal, and Amin Med were purchased from a local supermarket.[11] The sample containing 100% concentration can be assumed to be the “standard” used in commercially available toothpaste of each brand. The sample with a concentration of 70% had a 30% lower fluoride content, while the sample with a concentration of 130% had a 30% higher fluoride content with respect to the standard.[11] Each sample was stored for one year before measurements of total and dissolved fluoride content were taken. In order to determine the amount of total fluoride present, 10-20 mg of each toothpaste was weighed into cups and filled up to the 50 ml mark with deionized water. These mixtures were then diluted 10x with de-ionized water with no additional adjustments to pH and no use of filtration. The content of fluorine was then determined by the HR-CS GF MAS. Samples were also centrifuged for five minutes and the supernatant was used for further investigations in order to determine soluble fluorine.[11] Among completion of the study, it was seen that the most commonly used forms of fluoride in toothpaste are sodium fluoride, amine fluoride, and sodium monofluorophosphate (MFP). However, in order to test the length of storage, samples of Colgate Total and Colgate Max were prepared with various concentrations and stored for one year. All results obtained proved to be consistent with the assumptions made by the manufacturers, illustrating the presence of fluoride even after long periods of storage. The fluoride species initially tested were present in stable concentrations even after long storage periods. The proposed HR-CS GF MAS method is proven to be more efficient when compared to the GC-MS (gas chromatography- mass spectrometry) method.[11] Although both methods are reliable in terms of accuracy and precision and no significant differences in sensitivity has been detected, GC-MS requires more work and takes five times longer to complete the same number of samples. Also, the HR-CS method can be used in order to determine dissolved fluoride. A limited number of methods are available that are as efficient as the HR-CS method, therefore this proposed method could further be expanded and used to test other samples such as blood, food, urine, and pharmaceuticals.

The faculty of Dentistry at the Jordan University of Science and Technology performed an experiment to analyze the forms of fluoride ($F^-$) in a number of different toothpastes from large retail outlets through the use of buffers and electrodes. They collected six brands of toothpaste (used three tubes per brand) and created mixtures whose concentration had a theoretical yield of 200-210 ppm.[12] These slurries were then analyzed in order to find the ionic, soluble, and total fluoride concentration. The pH of the mixtures was also measured using a glass electrode. Prior to measuring the fluoride content, the suspensions of toothpaste were diluted 50x with deionized water. A parallel experiment was performed using an undiluted sample of fluoride. However, the diluted sample was mixed with a sodium acetate buffer. The concentration of fluoride was determined by using a $F^-$ -electrode. Portions of the soluble F were transferred to tubes where 0.125 ml of 4 M HCL was added, resulting in a concentration of about 0.8 M. These samples were then left to stand overnight (12 hours) at room temperature and then neutralized with 0.125 ml of 4 M sodium hydroxide. Deionized water was added allowing the mixture to consist of a total of 1 ml before mixing with a sodium acetate buffer (10 per cent by volume). After constructing the correct mixture for determination of fluoride, the various concentrations were tested. Total fluoride in the suspension ranged from 192- 207 μg/g and fluoride in the paste ranged from 987- 1046 μg/g.[13] The study revealed five major conclusions. First, the determination of the ionic fluoride concentration in the suspensions were found to be inaccurate regardless of which buffer was combined. Second, the determination of the total F concentration in supernatants of the slurry of MFP- toothpastes resulted in complete hydrolysis of MFP. Also, the use of the acid-HMDS diffusion method was used in order to separate the fluorine bound to the solid particles. Additionally, the toothpastes which contained amine F-sodium metaphosphate, aluminum silicate, or MFP-calcium carbonate presented the least solubility of fluoride. Lastly, abrasives in toothpaste formulas inactivated the fluoride. This method has gained prominence over the methods which use the traditional chemical components and colorimetric methods due to its sensitivity, specificity, speed, and ability to withstand a large range of concentrations. Eliminating a solvent is not only beneficial for the chromatographic column, but it is also beneficial for reducing the large amounts of hazardous solvents that accumulate over time. The reduction in the amount of reagents used and considerably shorter time allows for sufficient precision and accuracy in the measurement of fluoride concentration.

Wejnerowska, Karczmarek, and Gaca performed an experiment employing a new analytical method to determine the fluoride content of numerous brands of toothpaste. They used a solid-phase microextraction (SPME) method in order to determine its usefulness and compare it to the liquid-liquid extraction (LLE) method which has a low-cost and does not require the use of solvent.[13] A similar procedure was conducted for both methods and a linear calibration curve was constructed. Both procedures required 800 mg of the sample, however the LLE procedure required 2 ml of HCl, 2 ml of TMCS, 5 ml of solvent, 15 minutes of reaction time, 35 minutes of extraction, resulting in a total of 50 minutes. The SPME procedure consisted of 1 ml HCl, 30 μl of TMCS, no solvent, 10 minutes reaction time, 10 minutes extraction time, for a total of 20 minutes.[13] The results illustrate that the two procedures presented similar results. The presence of TMFS appeared after two and a half minutes of the reaction running, and the presence of TMCS appeared five minutes into the reaction. This method also allowed for the determination of fluoride in toothpastes which contain amine fluoride as well as sodium monofluorophosphate. The solid-phase microextraction method was seen to be advantageous over the traditional liquid-liquid extraction method not only because it was solvent free, considering the hazards associated with the use of organic solvents, but also because it is fast, easy, and eliminates costs of incorporating an organic solvent.

In a study performed by Gajic et al., a combined fluoride-selective electrode named “Jenway” was used in order to determine fluoride content. The active membrane was composed of a $LaF_3$ monocrystal that also had europium(II) fluoride that was used to reduce the resistance and to increase the movement of ionic charge. The ions of fluoride were able to bind to the crystalline surface. The internal component of the fluoride-selective electrode was composed of 0.1 M NaCl and 0.1 M NaF.[3] This internal activity controlled the electrodes inner potential. After combining necessary reagents and taking potentiometric measurements, fluoride content was determined. Analyzed samples consisted of toothpastes and mouthwash from various brands. Additionally, different fluoride salts were combined in the sample matrix allowing for a rapid and simple preparation of the experiment. High yields (94-103%) of fluoride content were confirmed for the accuracy of the method.[3] There were agreements between the specified and calculated contents of fluoride in various samples of mouthwash. Many advantages were observed for the proposed method, one being that this allowed for adapting the sample to pretreatment approaches depending on the type of fluoride in the desired dentifrice. The authors also suggested that there be exchangeable use of hydrolysis of fluoride sources and hexamethyldisiloxane diffusion due to the inaccuracies in fluoride determination. The procedure was seen to be reliable due to the good recovery of the non-fluoride samples as well as the fluoride- containing samples. Due to the various hazards and levels of toxicity fluoride possesses, stricter control of the type and amount of fluoride present in pharmaceutical products in oral healthcare is necessary.

**FOOD AND DRINKS**

Much of the food and drink an individual consumes has an acidic pH, making it easier for bacteria to wear down tooth enamel, leading to the formation of cavities. A study directed by Lussi and Carvalho tested the pH levels of different liquids individuals consume, some of which include soft drinks, sports drinks, fruit juices, alcoholic drinks, medication, milk, mineral water, tea, coffee, and salad dressing. Soft drinks, including Pepsi, Coca Cola, Sprite, Ice tea, Fanta, etc. have an average pH of 3.0. Sports and energy drinks such as Gatorade, Powerade, and Red Bull have an average pH of 3.5. Fruit juices such as apple juice, carrot juice, orange juice, etc. have an average pH value of 3.5. Alcoholic drinks such as Champagne, wine, and vodka have an average pH value of 3.3. Medication such as Neocitran, Alka-Seltzer fizzy tablets, Siccoral, Vitamin C fizzy tablet, etc. have an average pH value of 4.8. Mineral water such as Henniez, Valser, and Valser Viva Lemon have an average pH value of 5.7. Tea has an average pH value of 6.0 while coffee (espresso) has an average pH of 5.8. Salad dressings both classic and light have an average pH 0f 4.0.[14] This study demonstrated the true acidic nature of most liquids individuals regularly consume whether it be drinks, dressings, or medication, most of what an individual consumes is harming the tooth enamel. The pH of each group of liquids was found to be highly acidic, causing erosion to the teeth when introduced into the oral cavity. Milk is the only liquid tested that had a neutral pH of exactly 7.0, having less of a harmful affect in relation to other common drinks. In contrast, most water that is bottled, whether it be flavored, sparkling, or even regular, has a pH lower than neutral (7.0).[14] Tap water is more beneficial to consume due to the fluoride content. Although not all tap water is at a neutral pH, it is generally near neutral and contains fluoride, a necessary component in the protection of teeth that is present in miniscule amount or not present at all in various other drinks. Most drinks, such as coffee, sports drinks, soft drinks, etc. are continuously consumed over a span of a few hours or even an entire day. This habit causes the reintroduction of the acidic substance to the oral cavity, allowing bacteria to survive much longer, thus causing teeth to be more prone to decay and the formation of caries.

## CONCLUSION

As defined by Lussi and Carvalho, the softening process during dental erosion is described as "near-surface demineralization" and it refers to the erosion that occurs between the liquid and enamel as well as within the demineralized enamel layer which has become softened. This process clarifies the fact that dental erosion due to an acidic component is not just superficial, it occurs in the softened layer of the tooth as well. Lowering the extent at which a substance erodes dental enamel is possible through a variety of mechanisms. These processes increase the concentration of the inhibitor which in turn reduces the acidity of the substance. For example, the addition of calcium carbonate ($CaCO_3$) increases the pH of the solution as well as the level of saturation.[14] This mechanism allows the erosive potential of the solution to be lowered, reducing the harmful affects on the teeth. Research has been conducted regarding non-fluoridated dental products which contain protective agents such as polyvalent metal ions (tin and titanium), or even products which contain amino acids, peptides, or even proteins. Dental products that contain tin ions are known to provide protection against demineralization of the tooth structure. This is due to the interaction of tin with hard tissue on the teeth forming salts such as $Sn_2OHPO_4$, $Ca(SnF_3)_2$, and $Sn_3F_3PO_4$ which enable the formation of a protective layer that is resistant to erosion from acid. Although both tin and fluoride individually cause a similar reduction in deterioration of enamel, when present simultaneously, they inhibit a loss of minerals much more significantly. Dentifrices do not necessarily remineralize the tooth surface, but rather continue to hinder the loss of enamel in further acidic exposures. However, when tin is present in dentifrice, the mechanism of action includes the remineralization of the eroded enamel and the inhibition of further softening of the enamel, making tin a beneficial addition to the oral cavity. Similar to the benefits of tin ions, titanium ions (titanium tetrafluoride ($TiF_4$)) allow for the formation of a protective layer on the surface of teeth. A glaze-like protective layer is formed, which is ultimately dependent on the low pH of the titanium tetrafluoride. The amino acid arginine plays a significant role in the erosion of enamel. Arginine, when introduced to surface enamel, increases the affinity of $CaCO_3$ for the surface of the teeth. Other proteins directly impact the acquired enamel pellicle (AEP), which is a film that covers the surfaces of teeth and assists in protection against erosion. An example of this is the presence of a combination of casein and mucin. These proteins are able to increase the ability of AEP to become resistant to acid, decreasing the amount of erosion. Casein has also been used in commercial soft drinks in order to reduce the erosion of enamel in the oral cavity. The casein not only aids in erosion prevention, but also interacts with the pellicle and results in a film that protects the enamel layer from mineral loss. Although fluoride is widely used among the majority of populations, the number of patients with enamel erosion and caries is increasing. If preventative actions are taken in the future such as the incorporation of polyvalent metals in dentifrice in addition to fluoride, then more beneficial pre-cautionary methods are possible.

The analytical methods used to determine concentration of fluoride present in dental pharmaceuticals such as the fluoride- selective electrode (Jenway), solid-phase microextraction, liquid-liquid extraction, high-resolution continuum source graphite furnace molecular absorption spectrometry (HR-CS GF MAS), ion chromatography (IC), and fluoride ion selective electrodes (F-ISE) have been suggested to be effective methods for analytical chemists working in plants that manufacture dental pharmaceuticals which are used in dentistry as well as by the general public. However, considering the outcomes of the various studies which incorporate certain methods, some have been proven more accurate due to certain factors such as cost, outcomes, etc. For example, the Jenway (fluoride-selective electrode) has a high recovery rate (between 94 and 103%) while the solid-phase microextraction method was seen to be advantageous over the traditional liquid-liquid extraction method because it was solvent free, fast, easy, and eliminates the costs and hazards of organic solvents. Ion chromatography, on the other hand, has a low sampling rate and requires the use of a micrometer filter, making it less efficient in detection of fluoride then other methods such as the fluoride ion selective electrode. From the various studies performed using different methods of detection, it was seen that high-resolution continuum source graphite furnace molecular absorption spectrometry (HR-CS GF MAS) was the most efficient, not only in current detection of fluoride, but also in future expansion of the technique to measure various other samples. This method does not require and extensive preparation of samples and can easily be used. Additionally, this method as opposed to other which can only determine ionic fluoride, is able to detect dissolved fluorides. Future use of this method can be used for samples of blood, urine, pharmaceuticals, and an assorted number of foods. Properly measuring quantities and types of fluoride is important not only in an attempt to control dental caries and plaque build up, but also when taking into consideration one's overall health. Although fluoride is beneficial in strengthening enamel, too much exposure to the wrong types of fluoride can adversely affect health, both oral and overall.

Maintaining oral health is a crucial aspect of overall health. Dental hygiene methods have progressed significantly over the years and will continue to improve in the future. Fluoride is not only a component of dental pharmaceuticals such as toothpaste and mouthwash, but it can also be found in water and food. Although the side effects of fluoride are controversial, many studies have been performed illustrating the several benefits of the presence of fluoride in the oral cavity. Because individuals consume a variety of foods and drinks that contain large amounts of sugar and starch, the oral cavity is constantly introduced to the harmful bacteria that causes tooth decay, one form being dental caries (cavities). Various analytical methods such as solid phase microextraction (SPME) and fluoride-selective electrodes are used to detect the amount of fluoride present in oral hygiene products, thus confirming that the benefits are greater than the drawbacks. Fluoride, whether applied topically or systemically, aids in cavity prevention by building layers around the tooth enamel, allowing it to become stronger and less susceptible to attack and decay by bacteria. Given that children are more prone to tooth decay in the form of caries due to their poor oral hygiene routines, it is especially important for children from the ages of two until ten to receive fluoride whether it is applied by a dental hygienist in the form of fluoride foam (topically), through the consumption of fluoridated water, or through the use of toothpaste and mouthwash which contain fluoride. While it is difficult to avoid tooth decay and cavities completely, incorporating fluoridated dental hygiene products as well as fluoridated food and drinks that contain even a trace amount of fluoride can be beneficial in shielding teeth from the harmfulness of bacteria. Decay is not a superficial form of tooth destruction, but rather harms the deeper lays of the tooth, ultimately softening inner layers, making teeth more susceptible to erosion by acidic substances. Protection of the oral cavity, more specifically tooth enamel is possible through the combination of fluoride, polyvalent metals (tin and titanium), amino acids, and proteins.

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