

EROSION POTENTIAL OF WELL WATER, COMMERCIALY AVAILABLE BOTTLED
WATER, AND VEGETABLE JUICE

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ABSTRACT

Upoma Guha: Erosion potential of well water, commercially available bottled water, and vegetable juice
(Under the direction of Terence Donovan)

Objective: The study was conducted to evaluate the dental erosion potential of commercial bottled water, well water, and vegetable juice by pH-metric titration.

Method: 13 bottled water brands, well water from 15 locations, and 9 vegetable juice brands were tested for pH value and titratable acidity (TA). Palatability test and questionnaire survey were performed for bottled water.

Result: 6 bottled and 6 well waters had pH lower or close to critical pH of dentin (6.7) and enamel (5.2-5.5), while all vegetable juices showed low pH (4.0-4.3). Their TA value ranges were 1-14 μ L, 20-103 μ L, and 33.9-81.7mL of 0.1M NaOH, respectively. No significant differences were found in taste preferences for bottled water.

Conclusion: Marked low pH and high TA of all vegetable juice brands indicate significant potential for dental erosion compared to bottled and well waters, and can compromise dental health with long term exposure.

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LIST OF ABBREVIATIONS

Camp Tom	Campbell's Tomato Juice
DS	Degree of Saturation
FL Tom	Food Lion 100% Tomato Juice
GV Veg	Great Value 100% Vegetable Juice
HT Tom	Harris Teeter 100% Tomato Juice
IAP	Ion Activity Potential
MGB Veg	Mott's Garden Blend 100% Vegetable Juice
RWK MegGre	RW Knudsen Organic Tomato
RWK SimNut	RW Knudsen Simply Nutritious Mega Green
RWK VerVeg	RW Knudsen Very Vege
TA	Titrateable Acidity
V8 Veg	V8 Original Vegetable Juice

CHAPTER 1

INTRODUCTION

Erosive tooth wear is becoming more prevalent in contemporary dental practices. Dental erosion is defined as loss of tooth structure due to chemical exposure without any bacterial involvement (Levitch et al., 1994). Loss of tooth structure was first categorized in three types by John Hunter in 1778: erosion, abrasion and attrition (Bartlett and Shah, 2006). Later abfraction was identified as another lesion type (Grippo, 1991). Among all types of tooth wear, **dental erosion** has recently become the most common cause for degradation of aesthetics and mechanical properties (Ganss, 2006; Nunn, 1996). The following table (Table 1.1) has a listing of different types of tooth structure loss and their clinical features.

Table 1. 1 Types of tooth wear and associated causes (Bartlett and Shah, 2006; Grippo, 1991; Imfeld, 1996; Levitch et al., 1994)

Types of wear	Definition	Nature	Clinical location	Cause
Erosion	irreversible loss of dental hard tissue by chemical dissolution process without bacterial acid	Pathological	Non occluding surfaces	Gastoesophageal reflux, bulimia, acidic drinks or beverage, fruit consumption, industry employee
Abrasion	Abnormal loss of tooth structure due to biomechanical friction	Pathological	Cervical areas of tooth	Aggressive tooth brushing, noxious oral habit, pipe stem biting, fingernail biting etc.
Attrition	Wear of tooth structure due to mastication with age	Physiological	Occlusal surface	Masticatory force, normal or parafunctional habits
Abfraction	Stress-induced tooth loss due to biomechanical loading forces	Pathological	Cervical region	Flexure and ultimate fatigue of enamel and dentin

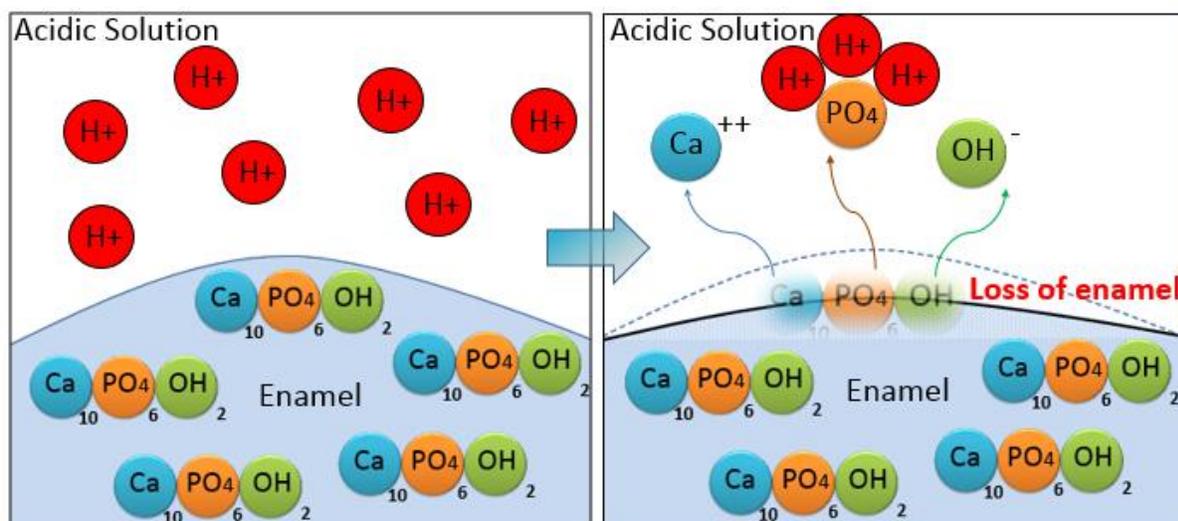


Figure 1.1. Mechanism of erosive tooth wear.

Erosion is associated with the chemical dissolution of enamel and dentin in contact with acidic substances unrelated to bacterial involvement (Imfeld, 1996). Dental hard tissue, (enamel and dentin) are primarily composed of hydroxyapatite, a calcium phosphate compound, chemically written as $\text{Ca}_{10}(\text{PO}_4)_6(\text{OH})_2$. However, carbonate, fluoride, etc. are also present as impurities. In a liquid environment (drinks/water in saliva), the dissolution of enamel occurs if the surrounding liquid (or complex plaque medium) is unsaturated in calcium (Ca^{2+}), phosphate (PO_4^{2-}), and hydroxyl (OH^-) ions. Further if the liquid (or complex plaque medium) pH is acidic, then the hydrogen (H^+) ions can replace Ca^{2+} ions of the matrix, (Figure 1.1) leading to dissolution of enamel until saturation is achieved. Consequent formation of phosphoric acid (H_3PO_4) leads to a more acidic environment. As the pH goes down, more OH^- ions are lost and thereby a significant decrease in saturation occurs. When the pH of surrounding medium drops further below 5.2-5.5, unsaturation becomes predominant and dissolution of enamel progresses. A similar phenomenon happens for root dentin at or below pH 6.7. Thus the **critical pH of erosion** for enamel is designated at 5.2-5.5 and for root dentin at 6.7 (Dawes, 2003; Donovan

and Swift, 2009; Noble et al., 2011). Table 1.2 lists the different types of erosion and their clinical features.

Table 1.2. Classification of dental erosion and clinical features (Bartlett, 2006; Zero, 1996).

Erosion Type	Definition	Clinical location
Extrinsic Erosion	The phenomenon of dissolution of enamel and dentin by acidic challenge from extrinsic or outer sources such as food, drinks etc.	Non occluding surfaces; incisal, buccal, facial, cervical areas
Intrinsic Erosion	Erosive tooth wear induced by intrinsic acids from gastro oesophageal reflux or bulimia	Palatal surface of maxillary teeth

Dissolution mediated dental erosion can be categorized in two classes based on the source of acidic solution: intrinsic and extrinsic. **Intrinsic erosion** is caused when acidic fluids inherently produced inside human body come in contact with the teeth and lowers the surrounding pH. This is attributed to gastroesophageal reflux (Bartlett et al., 1996; Erickson et al., 2013), bulimia patients (Szabo et al., 1991), vomiting (Uhlen et al., 2014) etc . **Extrinsic erosion** on the other hand is the demineralization of dental hard tissue by contact with non-body sources most commonly including acidic foods and drinks (Imfeld, 1989). The presence of citric or other organic acids (e.g. ascorbic acid) in food or carbonated drinks are responsible for the acidity (Lussi, 2006; Lussi et al., 2007; Prati et al., 2003). Foods and drinks with high erosive potential thus include citrus fruits (Grobler et al., 1989), sour vegetables (El Aidi et al., 2011; Pieroni, 2000), sour candies (Wagoner et al., 2009), acidic beverages (Edwards et al., 1999; Lissera et al., 1998; Lussi et al., 1995; Tenovuo and Rekola, 1977), fruit juices (Jensdottir et al., 2006; Larsen, 1975; Lissera et al., 1998) and smoothies (Blacker and Chadwick, 2013), vinegar (Prati et al., 2003) and salad dressings (Lussi et al., 2012), and sports (Cochrane et al., 2012; Coombes, 2005) and energy drinks (Kitchens and Owens, 2007; Pinto et al., 2013). Moreover, sparkling water (Brown et al., 2007) can also produce mild erosive challenges. Swimming pools

with water with a low pH water can contribute to erosive tooth wear for competitive swimmers (Buczowska-Radlińska et al., 2013; Centerwall et al., 1986; Geurtsen, 2000). Further workers in industries dealing with acidic fumes have been shown to demonstrate higher levels of dental erosion (Amin et al., 2001; Lynch and Bell, 1947; Malcolm and Paul, 1961).

The potential of erosion from acidic sources is initially determined by their pH values, titratable acidity, and mineral content (Cochrane et al., 2012; Dawes, 2003). Drinks or foods with the same pH can require different amounts of neutralizing alkalinity and thus can offer different degrees of erosive potential. The larger the alkali requirement, the greater the erosion potential. However, calcium, phosphate, or fluoride presence can elevate the degree of saturation and can decrease the erosion potential of the diet. Calcium or fluoride supplemented foods or drinks have shown the ability to reduce or prevent erosion (Andon et al., 1992; Hara and Zero, 2008; Magalhães et al., 2009). Other important factors for erosion are the viscosity of the substance (Aykut-Yetkiner et al., 2014) and contact time with tooth tissue (van Nieuw Amerongen and Veerman, 1995) . Higher contact time increases the chance of erosion. Frequency of consumption is also an important factor for erosion (Amaechi and Higham, 2005). Increased frequency of consumption also increases the contact time and erosion risk. Another factor is drinking style. Using a straw while drinking acidic beverages may prevent exposure to the surface of the tooth to the acid, and hence may reduce the effect of erosion (Edwards et al., 1998). Wine testers often show significant erosion due to the method of sipping the wine and swishing it in their mouths for a longer time (Gray et al., 1998).

Saliva plays a very significant role in preventing erosion and caries (Meurman and ten Gate, 1996; Zero and Lussi, 2005). The extent of erosion largely depends on the saliva's buffering capacity. Saliva consists of 99% water; the rest is composed of proteins and

electrolytes such as sodium, potassium, calcium, magnesium, bicarbonate, and phosphate. These proteins and electrolytes are essential for the buffering capacity of saliva which helps to keep the pH above the critical erosive pH (Buzalaf et al., 2012). Salivary pellicle covering the enamel also protects the tooth tissue from being eroded (Meurman and ten Gate, 1996; Zero and Lussi, 2005). Erosive lesions are site specific and inversely related to salivary pellicle formation (Amaechi and Higham, 2005). Hence, diminished salivary flow or production is contributory to erosive lesions. Thus, people with low salivary flow or ‘dry mouth’ may potentially be more prone to dental erosion than others.

While soft drinks and fruit juices have been extensively investigated for their erosive effects **bottled drinking water** and **vegetable juices** have not been investigated. These drinks are believed to be sources of safe water and to provide important health benefits.

Consumption of bottled water has increased significantly in recent years. An increase of 6.3% in the consumption of bottled water was observed during the year 2011-2012 (IBWA, 2013). However, manufacturer provided data and media reports regarding pH values and mineral content of these water brands are somewhat ambiguous – ranging from highly acidic (~4.0) to alkaline (~10). Further ultra-purification of the bottled water deprives them of important minerals (e.g. calcium and phosphate) required for prevention of dental erosion. With increasing usage and frequent consumption of these seemingly safer alternatives of drinking water, an increasing oral health risk related to dental erosion is possible. Therefore as mentioned in the second chapter, the erosive potential from 13 different bottled water brands was determined by pH measurement, titration, and reported mineral content. Further, well water from different locations in North Carolina was also assessed for their erosive potential and compared with the bottled water samples to show the importance of the mineral content. Moreover, taste

preference via a palatability test and a questionnaire based survey were performed to determine the popularity of bottled water brands. This study addresses preliminary information regarding enamel and dentin dissolution regular consumption of bottled water and indicates towards the necessity to improve public awareness regarding such oral health concerns.

The third chapter provides similar pH and titratable acidity information for commercially available pure vegetable juices. One hundred percent vegetable juice is a blend from different juiced vegetables including cucumbers, tomatoes, celery, etc. without any fruit additives. These juices are attractive to people with a focus for healthy living due to anti-disease and anti-obesity properties that are theoretically obtained from vegetables (El Aidi et al., 2011; Steinmetz and Potter, 1996). Further, young populations may be inclined to intake more vegetables in a juice formulation rather than the whole form. While media reports suggest these juices are also beneficial for oral health (e.g. teeth whitening purposes), no reports address their dental erosive potential. It has been observed that sour vegetables including cucumber, celery, tomato, etc. have shown some potential to dissolve enamel dissolution (El Aidi et al., 2011). Also, the vegetarian diet has been associated with erosive tooth wear (Linkosalo and Markkanen, 1985; Smith et al., 2008).

It is to be noted that taste and flavor enhancement in pure vegetable juice are performed by the addition of acidic concentrate that reduces the pH of the juice significantly. Low pH values along with the high viscosity of thick vegetable juice may potentially contribute towards tooth dissolution. However, no systematic studies have been conducted to evaluate the contributions of vegetable juices towards dental erosion.

Dental erosion is a chronic attack from extrinsic or intrinsic sources which eventually can lead to aesthetic impairment via lesion formation and reduction of mechanical properties of

enamel and dentin. When unattended during early stages dental erosion can progress to severe tooth wear (Vanuspong et al., 2002), dentin sensitivity (Vanuspong et al., 2002), loss of hardness (Wongkhantee et al., 2006), and impairment of functions (Schlueter et al., 2012). Moreover, the treatment of dental erosion involves a number of investigations, including preventive and remineralization strategies, minimally invasive adhesive restorative strategies and full mouth prosthetic rehabilitation (Sorvari and Rytomaa, 1991). Therefore, prevention of erosive tooth wear by controlling the diet is important critical to retain acceptable esthetics and avoid costly dental care. Through the thesis, an overall effort to address the erosive potential of bottled drinking water and vegetable juice will be performed. The high quality processing of vegetable juice, purification of bottled water, and the manufacturers' packaging and advertisements do not disclose the potential oral health hazards from these drinks. A primary effort to address this discrepancy has been initiated through the completion of the experimental works presented in the subsequent chapters.

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CHAPTER 2

EROSION POTENTIAL OF COMMERCIAL BOTTLED WATER AND WELL WATER AND EVALUATION OF TASTE PREFERENCE OF COMMERCIAL BOTTLED WATER

2.1 INTRODUCTION

Dental erosion is currently recognized as one of the leading causes of loss of tooth structure (Donovan and Swift, 2009; Erickson et al., 2013; Schlueter et al., 2012). Erosion is defined as the chemical loss of tooth structure with no involvement of bacteria (Featherstone and Lussi, 2006). There are 2 forms of erosion, extrinsic and intrinsic (Jarvinen et al., 1991). Extrinsic erosion is caused by the ingestion of acidic food and beverages (Jarvinen et al., 1991; Wongkhantee et al., 2006; Zero, 1996). Intrinsic erosion is the result of expulsion of the contents of the stomach into the esophagus, oro-pharynx and the oral cavity (Erickson et al., 2013; Jarvinen et al., 1991; Scheutzel, 1996; Zero and Lussi, 2005).

Many food and beverages have pH below the critical pH of enamel (5.2 - 5.5) and root dentin (6.7) (Dawes, 2003; Featherstone and Lussi, 2006; Jarvinen et al., 1991; Meurman and ten Gate, 1996), and thus possess the potential to contribute to erosive tooth wear. Some of these food and beverages include fruits, especially citrus fruits and juices (Jarvinen et al., 1991; Larsen and Nyvad, 1999; West et al., 1998; Zero and Lussi, 2005), soft drinks (Fraunhofer and Rogers, 2004; Jarvinen et al., 1991; Jensdottir et al., 2004; Kitchens and Owens, 2007), sports (Cochrane

et al., 2012; Coombes, 2005b; Kitchens and Owens, 2007; Noble et al., 2011) and energy drinks (Kitchens and Owens, 2007; Pinto et al., 2013; Sirimaharaj et al., 2002), white wine and vinegar (Jarvinen et al., 1991; O'Sullivan and Curzon, 1999) and salad dressing (DDS, 2011; Jarvinen et al., 1991; O'Sullivan and Curzon, 1999). Competitive swimmers are at risk for erosion if they swim in pools where the pH is not carefully monitored (Buczowska-Radlińska et al., 2013; Centerwall et al., 1986; Geurtsen, 2000).

Regular consumption of bottled water has become popular in contemporary society. In 2012, almost 12 billion dollars were spent in the USA on bottled water, with 9.7 billion gallons being consumed – which is 15% of the total global consumption of bottled water (IBWA, 2013). During 2011-2012 alone, the consumption increased by 6.5%. It is estimated that consumption of bottled water has tripled since 1991, which demonstrates a staggering increase from 9.8 gallons per capita annual consumption in 1991 to 30.8 gallons in 2012. The global market for bottled water is estimated at 100 billion dollars annually.

It is a common belief by both lay persons and dental professionals that bottled water has a neutral pH, contains all healthy supplements and would not contribute to harmful effects to an individual. However online reports provide ample information demonstrating that pH of bottled water differs from brand to brand. Some of them are acidic (Bottled Water pH List). Moreover, sometimes they are devoid of chemical constituents such as calcium and phosphate (SkipThePie.org). Thus, seemingly benign bottled water may contribute to tooth erosion when consumed on a daily basis for a long time.

This study was conducted first to determine the pH and titratable acidity of 13 popular brands of bottled water available in grocery stores in North Carolina. In order to provide a comparison, samples of well water were collected within a 200 mile radius of Chapel Hill, and

their pH and titratable acidities were determined. The second objective was to determine taste preferences of different commercially available bottled drinking water through a palatability test using a visual analogue scale (VAS).

2.2 MATERIALS AND METHODS

Thirteen different brands of bottled water were selected based on popularity and availability in grocery stores in North Carolina. The brands of water sampled and their manufacturers are listed in Table 2.1. Three bottles of each brand (N = 39) were obtained from 3 different stores.

Samples of well water were collected within a 200 mile radius of Chapel Hill, with approximately 5 miles between sample sites. Attention was paid to geographic complexities of the landscape, and wells were selected to represent a wide range of water tables and underground reservoirs. These included locations on both eastern and western sides of the Appalachian Mountains, representing a variety of rock substrate bases as sources of mineral content in water.

Table 2.1. Name of the bottle water brands and their manufacturing companies.

Bottled Water brands	Manufactures and Addresses
Penta	Penta Water Company, Colton, CA
Dasani	The Coca-cola Company, Atlanta, GA
Aquafina	Pepsi-cola, Purchase, NY
Deer Park	Nestlé Waters North America, Stamford, CT
Resource	Nestlé Waters North America, Stamford, CT
Nestle Pure Life	Nestlé Waters North America, Stamford, CT
Smart Water	The Coca-cola Company, Atlanta, GA
Volvic	Volvic, Allentown, PA
Fiji	Fiji water, Los Angeles, CA
Icelandic Glacial	Icelandic Glacial, Los Angeles, CA
Eternal	Eternal Water, Smokey Mountain, TN
Iceland Natural Spring Water	Iceland Spring, West Palm Beach, FL
Alkalife 10	Alkalife, Miami, FL

One hundred mL of untreated well water were collected from each sample site in two 50 ml. vials. The vials were then numbered and labeled with the location and date of sampling, and then were sealed with Parafilm (Pechiney Plastic Packaging Co., Chicago, IL).

In order to determine the pH of both the bottled water and the well water, 100 mL of each sample was poured into a glass beaker. A pH meter with a pH probe (Mettler Toledo MP230 pH meter and Mettler Toledo Inlab 413 pH probe, Mettler Toledo Int., Columbus, OH) was calibrated using buffer solutions of pH 4, 7 and 10. Then the calibrated pH probe was immersed in each water sample to measure the pH. This was replicated 3 times for each sample.

Titrateable acidity was determined by adding freshly prepared 0.1 M NaOH solution with a micro pipette in 10 μ L increments until the pH reached 7.0. Titrateable acidity determinations were only done on water samples with a recorded pH below 7. The water sample was magnetically stirred during the NaOH additions using a Corning PC-320 Hot Plate Stirrer (Corning Inc., Corning, NY) to ensure proper mixing. The cumulative volume of NaOH added to reach the level of pH 7 was the titrateable acidity of the water sample. A sample data collection chart to determine pH and TA is given in Appendix 2.1.

The pH values of the water samples were compared with the critical pH of enamel (5.2 – 5.5) and dentin (6.7) to evaluate the erosive potential of the bottled and well water. The erosive potential was further evaluated by considering the effect of minerals suspended in the water samples using a dissociation equation and resulting degree of saturation. The method for calculating the degree of saturation is presented in Appendix 2.2 (Cochrane et al., 2012).

The palatability study was performed to determine any related brand specific test preference of 8 commercial bottled water with 52 participants (N=52) including students, faculty, and staff of the School of Dentistry, University of North Carolina at Chapel Hill. The study

protocol was exempted by the Institutional Review Board (IRB study #14-2187; exempted 10/30/2014). The consent form and taste evaluation form are provided in Appendix 2.3. The eight brands were selected based solely on the pH study results including 3 low pH brands, 2 medium pH brands and 3 high pH brands of water bottles. Every interested participant was asked to taste 30 mL samples from each of 8 different commercial brands of bottled drinking water in a blinded manner at an interval of 10 seconds in between each sample tasted. After tasting each sample, participants were asked to determine the pleasantness or unpleasantness by marking a point on the Visual Analogous Scale (VAS) with 0 for most unpleasant and 10 for most pleasant sample. Statistical analyses of the obtained data were performed with One-way ANOVA with reported measures (parametric method) in SAS (SAS, Cary, NC).

A survey questionnaire was completed to obtain the demographic information of the 52 participants of the palatability test (Appendix 2.4). This included questions to identify the factors affecting the preference of various brands of bottled water, behavioral information indicating health consciousness etc. Participation was completely voluntary in the survey.

2.3 RESULTS

The measured pH values of bottled water ranged from 5.29 to 9.79, and the pH of the well water samples ranged from 5.5 to 7.7. The recorded pH values of the bottled water are displayed in Table 2.2 and shown graphically in Figure 2.1a. Five of the brands had pH values below the critical pH of root dentin and 1 was near or below the critical pH of enamel. The pH values of the well water samples are displayed in Fig. 2.1b. Water from 5 of the wells sampled had pH values below the critical pH of root dentin.

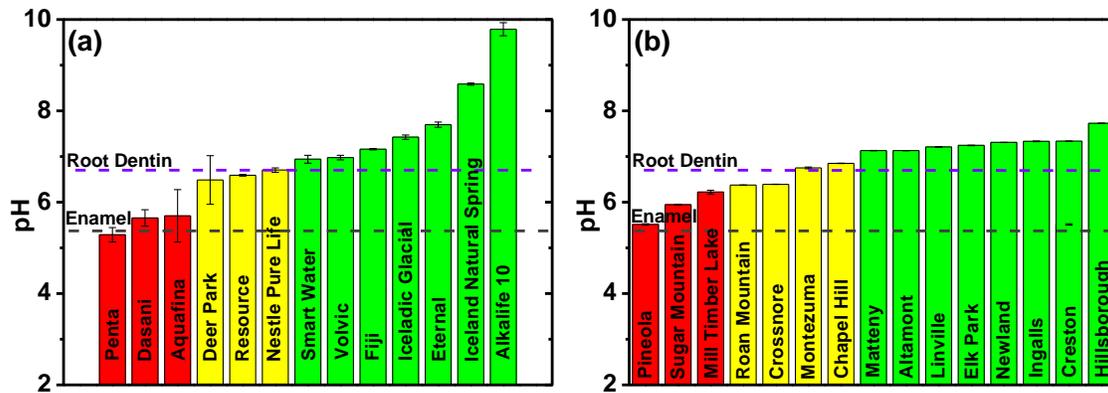


Figure 2.1. Measured mean pH values with respective standard deviation of (a) 13 commercially available bottled water and (b) well water from 15 different regions of North Carolina. Critical erosive pH of enamel is 5.2-5.5 and of root dentin is 6.7 as indicated by dashed lines. Red bars indicate higher potential for enamel and root dentin dissolution. Yellow bars indicate higher potential of root dentin demineralization. Green bars have pH above the critical value for potential erosive tooth wear.

Table 2.2. Comparison of measured pH values of bottled water brands with their online reported values.

Name of the brands and manufacturers	Reported pH variation	Measured pH
Penta (<i>Penta Water Company</i>)	7.8, 6.7, 4, 8, 6*	5.29
Dasani (<i>The Coca-cola Company</i>)	5.6, 7.2, 4, 4.1	5.65
Aquafina (<i>Pepsi-cola</i>)	5.5, 5.9, 4, 3.5	5.70
Deer Park (<i>Nestlé Waters North America</i>)	7.8, 6.3, 7, 5.6-8.3*	6.49
Resource (<i>Nestlé Waters North America</i>)	6.5-7.1*	6.59
Nestle Pure Life (<i>Nestlé Waters North America</i>)	7.3, 6.6-8.0*	6.70
Smart Water (<i>The Coca-cola Company</i>)	7.6, 7	6.94
Volvic (<i>Volvic</i>)	7.5, 7.1, 7, 6.7*	6.98
Fiji (<i>Fiji water</i>)	7.3, 6.7, 7.3, 7.7*	7.16
Icelandic Glacial (<i>Icelandic Glacial</i>)	8.4*	7.52
Eternal (<i>Eternal Water</i>)	8.0, 7.8-8.2*	7.70
Iceland Natural Spring Water (<i>Iceland Spring</i>)	8.9*	8.59
Alkalife 10 (<i>Alkalife</i>)	10*	9.79

* pH mentioned in the company websites and quality reports.

Reported pH values of the bottled water brands on the manufacturers' websites were compared with the measured pH values as displayed in Table 2.2. There are considerable differences between values reported by the manufacturers and the measured values in the study.

For acidic bottled water samples, the titratable acidity ranged from 20 μL of 0.1 mL NaOH (Penta) to 103 mL (Deer Park). Compared to the bottled water samples, the acidic well water samples required less NaOH and thus generally had lower titratable acidities. The titratable acidities of both groups are displayed in Figure 2.2a and b.

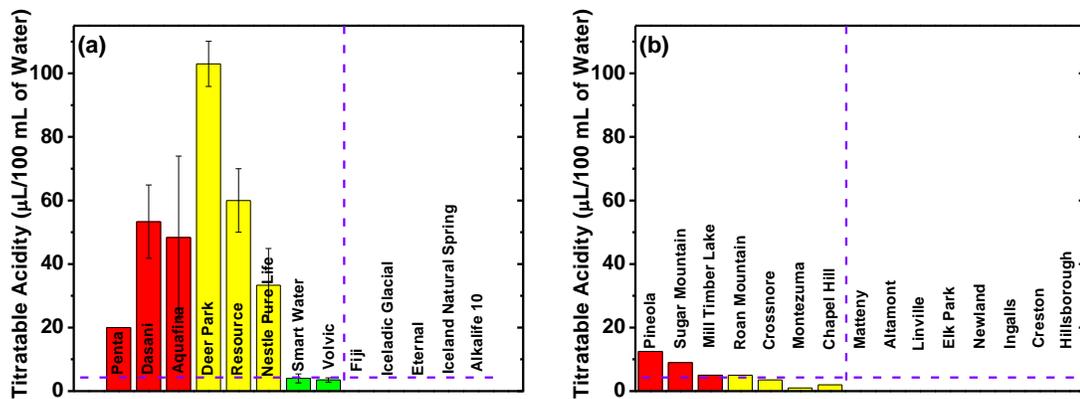


Figure 2.2. Measured titratable acidity as μL of 0.1 M NaOH needed to neutralize 100 mL of samples from (a) 13 commercially available bottled water and (b) well water from 15 different regions of North Carolina. Only the samples with $\text{pH} < 7$ were titrated.

The palatability test results (Figure 2.3) present with no significant difference in taste preferences for bottled water brands. The survey questionnaire data revealed (figure 2.4) that the two mutually non-exclusive groups, 63% people preferred bottled water brands with pH lower than the critical pH of enamel (5.2 – 5.5) and dentin (6.7) and only 37% people preferred bottled water with pH higher than 6.7.

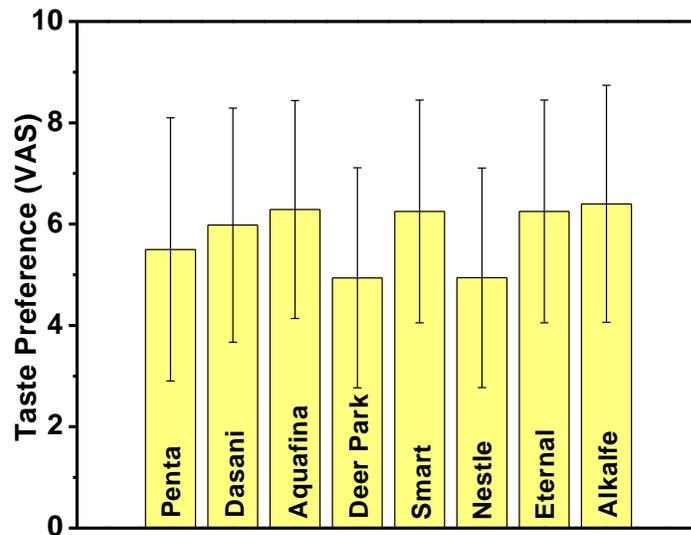


Figure 2.3. Taste preferences of commercial bottled water brands obtained by blinded palatability test with 52 individuals.

The questionnaire results also showed that the public concept of convenience of commercial bottled water is mainly due to two reasons – availability (34%) and ease of use (34%) in comparison with other factors such as cleanliness, taste, price, healthy, and mineral content (Figure 2.6). Moreover, the considerations to decide for the final selection of the purchase finally depend on price (45%), flavor (25%) and availability (15%) (Figure 2.7). One key finding was that the significant populations (82%) do not consider drinking alkaline water with safer pH range (Figure 2.8).

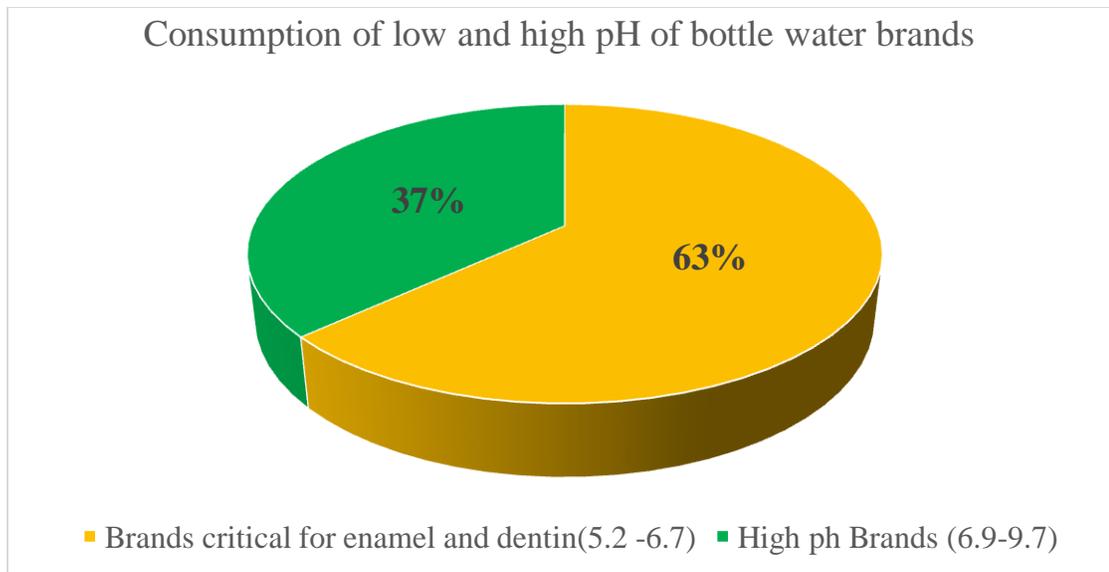


Figure 2.4. Consumption of bottled water brands with low and high pH range

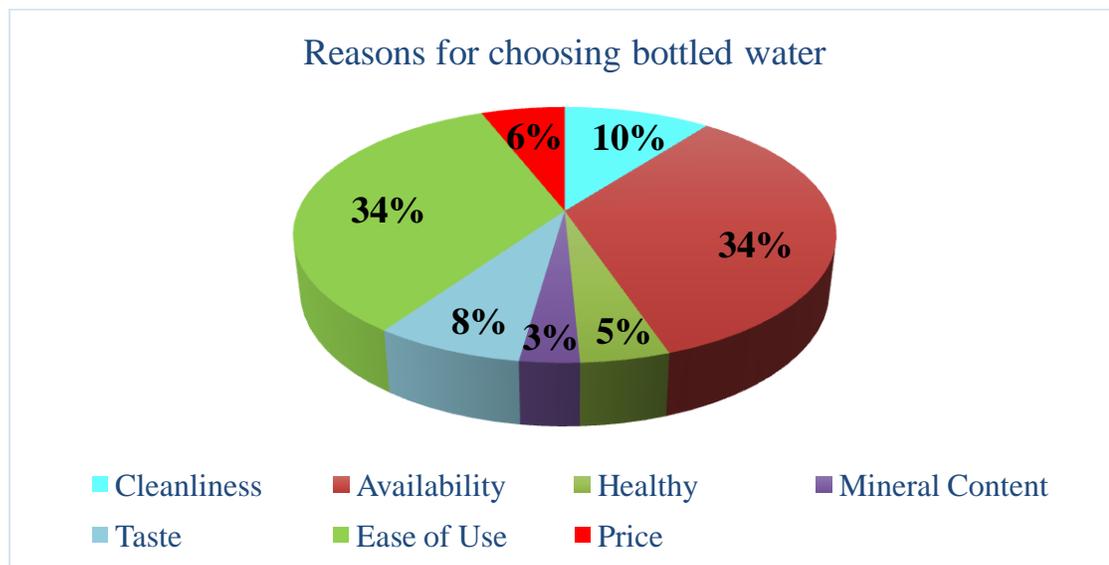


Figure 2.5. Reasons for choosing bottled water

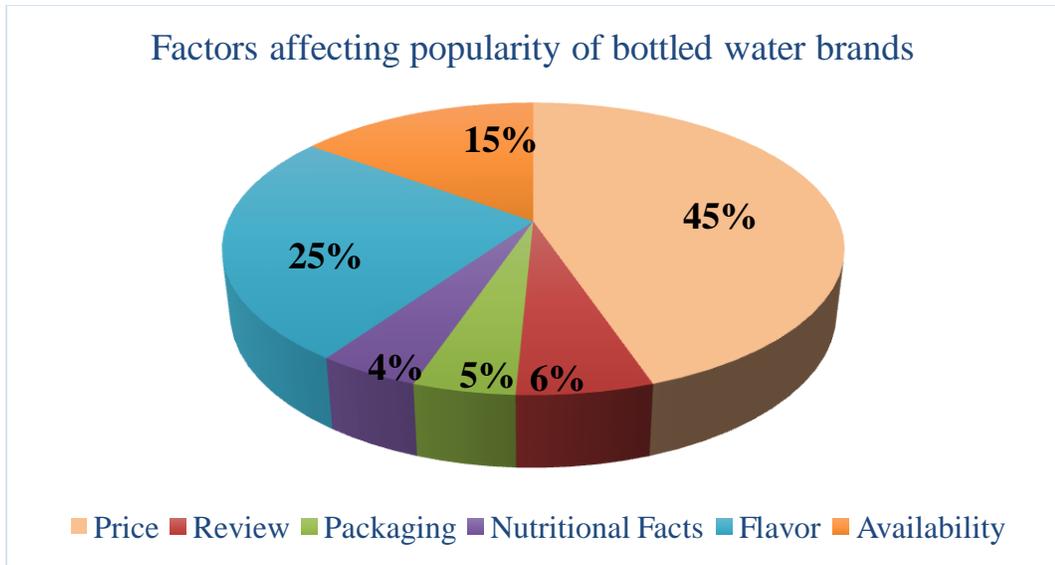


Figure 2.6. Factors affecting popularity of bottled water brands

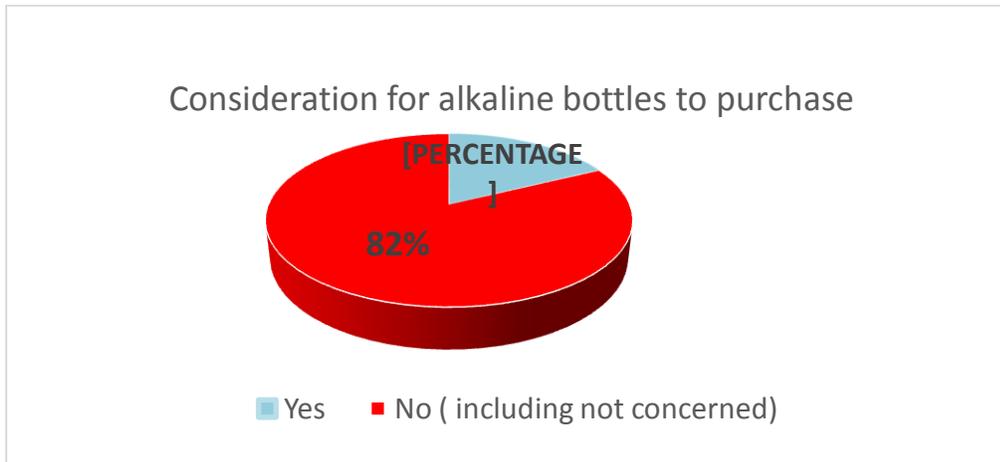
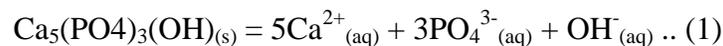


Figure 2.7. Significantly high percentages of population are not concerned or aware of pH values of bottled waters and do not consider purchasing high pH containing water products.

2.4 DISCUSSION

The results of this study are interesting primarily because both lay persons and dental professionals likely assume that bottled waters have a neutral pH. Clearly, neither bottled water nor well water can be assumed to have a neutral pH, and several brands and wells had water with a pH below the critical pH of root dentin, and 1 bottled water sample had a pH about the same as the critical PH for enamel. It should not be construed by the reader that these findings indicate the public is at high risk for erosion by consuming certain brands of bottled water. The risk for significant dental erosion from these products for most patients is minimal. However, it could be significant for an individual who consumes copious amounts of certain brands of bottled water and also has compromised salivary flow rates (Zero and Lussi, 2005). The brands with lower pH that have been intentionally demineralized seem to pose the most risk.

Tooth erosion does not entirely depend on the pH of the surrounding solution, but is controlled by the degree of the saturation provided by the minerals present in the suspension. Tooth enamel, otherwise known as calcium hydroxyapatite and having the chemical formula $\text{Ca}_5(\text{PO}_4)_3(\text{OH})$, when exposed to a suspension or water follows a dissociation reaction as presented in equation 1 (Dawes, 2003).



The dissociation reaction will continue until the solution or water sample is saturated with the calcium, phosphate, and hydroxyl ion when compared to hydroxyapatite (Cochrane et al., 2012; Dawes, 2003). The reaction constant is known as the solubility product of the hydroxyapatite and is given using the formula $k_{sp} = \{\text{Ca}^{2+}\}^5 \{\text{PO}_4^{3-}\}^3 \{\text{OH}^{-}\}$ where right hand terms inside parenthesis correspond to the active concentrations of calcium, phosphate, and hydroxyl ions. The k_{sp} value for enamel has been determined as 5.5×10^{-55} .

The potential of the enamel to dissolve can be determined by having the ion activity product (IAP) which can be calculated from the same equation (1) but with the actual concentrations of the ions present in the solution. Enamel will dissolve in a solution only if $IAP > k_{sp}$ or $DS = (IAP/k_{sp})^{1/n} > 1$, where DS=degree of saturation and n = the total of the coefficients. An estimation of the degree of saturation from the calcium (from their companies' quality reports) and phosphate concentration data in the bottled water and hydroxyl ion concentration from the measured pH values was made. However, it was not possible to find phosphate concentration data in the bottled waters. Most of the bottled water companies declared no phosphorus or phosphate concentration data. Generally phosphate concentration needs to be less than 0.1 mg/L concentration to avoid microbial growth in drinking water (Water Quality). Therefore, a threshold concentration of 0.1 mg/L for phosphate concentration was used.

As mentioned before, a sample calculation for the degree of saturation is presented in Appendix 2.2, the degree of saturation values for bottled water samples are presented in table 2.3, and also the whole data set for the calculation is presented in Table 2.4. It is important to note the degree of saturation values were not dominated by the pH but the overall composition of the water. Only the more acidic bottled water samples (Penta, Aquafina, and Dasani) present with unsaturated conditions due to the absence of calcium ions. Other samples show supersaturation with degree of saturation values greater than 1. Thus, the pH along with the mineral content determines the bottled water erosive potential. This is also crucial as the bottled water brands with the greatest erosive potential (Penta, Aquafina, and Dasani) are produced by ultra-purification of previously treated drinking water to remove the mineral content. While such high purity has been appealing to the consumers, such mineral content elimination creates the risk of enamel dissolution and possible tooth loss with long-term exposure.

Actual enumeration of the degree of saturation for these samples requires exact determination of anion and cation concentrations in the bottled water samples via state-of-art ion chromatographic and mass spectroscopy techniques (Cochrane et al., 2012). However, it is to be noted that the estimations were based on a high phosphate concentration (0.1 mg/L). If phosphates in several orders of magnitude lower concentrations are present in near neutral to alkaline bottled water, the degree of saturation will go down and may reach unsaturation in the absence of phosphate and there will be potential for causing tooth erosion.

It would be desirable to determine the degree of saturation value for the whole set of well water samples. However this was not possible due to the unavailability of the mineral content data which will require determination of the mineral content in the well water samples using above mentioned techniques. Yet, in order to have a presumption about the well waters' erosive potential, it was estimated as the degree of saturation of the well water sample collected from Hillsborough NC, which had the highest pH of 7.7 and also with the sample from the Old Well of Chapel Hill which had a pH slightly below 7 (i.e., 6.84).

Table 2.3. Estimated degree of saturation for the 13 bottled water brands.

Bottled Water Brands	Degree of Saturation
Penta	0
Dasani	0
Aquafina	0
Deer Park	62.97
Resource	23.99
Nestle Pure Life	29.28
Smart Water	21.29
Volvic	28.38
Fiji	39.91
Icelandic Glacial	19.13
Eternal	50.67
Iceland Natural Spring	21.67
Alkalife 10	29.23

Table 2.4. Calculated values for degree of saturations for bottled water samples from measured pH values and compositional information.

Brand Name	pH	Total Dissolved Solids (mg/L)	Ionic Strength	Ca ²⁺ Conc. (M)	(PO ₄) ³⁻ Conc. (M)	OH ⁻ Conc. (M)	Ca ²⁺ Activity Coefficient	(PO ₄) ³⁻ Activity Coefficient	OH ⁻ Activity Coefficient	Ion Activity Potential (IAP)	Degree of Saturation
Penta	5.29	1	0.000025	0	1.052E-06	1.949E-09	1.0232	1.0529	1.0057	0	0
Dasani	5.65	20	0.0005	0	1.052E-06	4.467E-09	1.1059	1.2543	1.0255	0	0
Aquafina	5.703	9	0.000225	0	1.052E-06	5.047E-09	1.0704	1.1655	1.0172	0	0
Deer Park	6.49	115.5	0.0028875	0.0017	1.052E-06	3.09E-08	1.2647	1.6962	1.0604	8.57E-39	62.97
Resource	6.59	95.5	0.0023875	0.0003	1.052E-06	3.89E-08	1.2393	1.6205	1.0551	1.45E-42	23.99
Nestle Pure Life	6.7	80.5	0.0020125	0.000425	1.052E-06	5.012E-08	1.2186	1.5603	1.0506	8.69E-42	29.28
Smart Water	6.94	36	0.0009	0.00025	1.052E-06	8.709E-08	1.1435	1.3523	1.0341	4.96E-43	21.29
Volvic	6.98	150	0.00375	0.0003	1.052E-06	9.549E-08	1.3044	1.8183	1.0687	6.57E-42	28.38
Fiji	7.16	220	0.0055	0.00045	1.052E-06	1.445E-07	1.3743	2.0449	1.0827	1.41E-40	39.91
Icelandic Glacial	7.42	62	0.00155	0.00015	1.052E-06	2.630E-07	1.1906	1.4806	1.0446	1.89E-43	19.13
Eternal	7.7	105	0.002625	0.000675	1.052E-06	5.012E-07	1.2516	1.6569	1.0577	1.21E-39	50.67
Iceland Natural Spring	8.59	48	0.0012	0.000115	1.052E-06	3.890E-06	1.1667	1.4147	1.0393	5.81E-43	21.67
Alkalife 10	9.8	43	0.001075	0.000115	1.052E-06	6.309E-05	1.1574	1.3895	1.0372	8.55E-42	29.23

For this analysis, the average calcium and phosphate concentration data obtained from the Orange County well water report was used as Chapel Hill and Hillsborough are within Orange County (Cunningham and Daniels, 2001). The estimated degrees of saturation was found to be around 47 and 38 which are much higher than 1 indicating extremely low potential of tooth dissolution. It is likely well water will probably provide remineralization due to the presence of calcium and phosphate even though their pH values are not highly alkaline.

Therefore, it is evident that acidic bottled water has the potential of erosive tooth wear not only due to the low pH but also due to the absence of the minerals calcium or phosphate. An increase in pH along with addition of mineral can prevent the potential tooth erosion due to continuous exposure.

The palatability test and questionnaire survey results suggest that the popularity of the bottled water is not solely dependent on taste preference. Rather the ease of availability and lower prices of low pH water brands are the major factors affecting brand selection. From a market overview the lower pH brands from popular companies are readily available in any super store, gas station or vending machines with a considerable low price which makes it more accessible to the public in general. Additionally the sense of cleaner and safer water from a bottled source will continue to increase their consumption which can possibly lead to a habit of daily intake which in turn will raise the frequency of exposure to a maximum level to develop significant erosive impact.

2.5 SUMMARY AND CONCLUSIONS

The results of this study indicate that:

1. Some brands of bottled water have pH values below that of the critical pH for root dentin.
2. The brands of bottled water that have the lowest pH values with low mineral contents, which might increase their erosive potential.
3. Constant use of the low pH brands of bottle water may increase the erosive risk.
4. Well water has variable pH, some of which are below the critical pH for root dentin. However, they tend to have relatively low titratable acidities and contain significant concentrations of minerals. Well water is more likely to assist in the remineralization of tooth structure than demineralization.

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CHAPTER 3

EROSION POTENTIAL OF COMMERCIALY AVAILABLE VEGETABLE JUICE

3.1 INTRODUCTION

Erosive tooth wear is a significant problem in contemporary practices. (Donovan and Swift, 2009; Erickson et al., 2013; Lussi, 2006; Nunn, 1996). Erosion is caused by acidic dissolution of dental hard tissues, i.e. enamel or root dentin, via replacement of the calcium ion by hydrogen ions generated at low pH without assistance from bacterial community (Donovan and Swift, 2009; Imfeld, 1996). Continuous exposure to acidic fluids can lead to such deterioration of tooth structure which can ultimately result in aesthetic and functional impairment due to lesion formation and loss of mechanical strength (Lussi, 2006). The two major types of erosion are intrinsic which include bulimia and gastroesophageal reflux disease (Erickson et al., 2013; Jarvinen et al., 1991), and extrinsic erosion resulting from the congestion of acidic foods and beverages (DDS, 2011; Imfeld, 1989; Zero, 1996).

Foods and beverages with a pH lower than the critical erosive pH of enamel (5.2 - 5.5) and root dentin (6.7) have shown to significantly contribute towards erosive tooth wear (Dawes, 2003; Donovan and Swift, 2009). Consumption of acidic foods including citrus fruits (Aquad and Moynihan, 2007; Grobler et al., 1989; Moynihan, 2002), sour vegetables (El Aidi et al., 2011), salad dressings (Lussi et al., 1993; Lussi et al., 2012), etc. has shown to be responsible for dental erosion. Similarly, low pH drinks including fruit juices (Larsen, 1975; Lussi et al., 1993; Smith and Shaw, 1987), smoothies (Blacker and Chadwick, 2013), soft beverages (Grando et al., 1996;

Jensdottir et al., 2006; Kitchens and Owens, 2007; Prati et al., 2003), energy (Kitchens and Owens, 2007; Pinto et al., 2013) and sports drinks (Cochrane et al., 2012; Coombes, 2005; Kitchens and Owens, 2007; Noble et al., 2011), alcohol (Gray et al., 1998; Lissera et al., 1998), etc. can cause erosion. Moreover, the vegetarian diet has recently been identified to be associated with an increased level of dental erosion (HermanABD et al., 2011; Linkosalo and Markkanen, 1985; Rafeek et al., 2006; Sherfudhin et al., 1996; Smith et al., 2008).

The increasing awareness regarding healthy living has led to increased consumption of fruits and vegetables and their derivatives. Vegetables are the sources of many health benefits including protection against obesity (Slavin and Lloyd, 2012), cardiovascular complications (Appel et al., 1997), or even cancer (Block et al., 1992; Steinmetz and Potter, 1996) and also by increasing anti-inflammatory (Giugliano et al., 2006) and cleansing abilities (Pieroni, 2000). They are consumed directly and also as juices. Juice intake has been identified as one of the easiest methods of vegetable intake and has been promoted to encourage young people and adults to increase vegetable intake (Kimmons et al., 2009). Though nutritious values of vegetable juices remain important, their contributions to oral health have not been investigated thus far. Moreover, acidic additives are included in vegetable juices to improve the flavor and taste (FAO). Therefore, systematic studies regarding dental erosion from consumption of these seemingly healthy drinks are warranted.

The present study was performed to determine the pH and titratable acidity (TA) of commercially available vegetable juices in different stores in North Carolina. A pH metric titration methodology was employed to determine the dental erosion potential of these vegetable juice samples.

3.2 METHODS AND MATERIALS

Nine widely available vegetable juice brands were selected based on popularity and contents. Among these, eight brands were selected only with pure vegetable contents while one contained a mixture of fruit juices and was considered as a negative control. The brands and their manufacturers along with ingredient information are listed in Table 3.1.

Three different samples of each selected brand were collected from 3 different stores ($N = 27$). 100 mL of each vegetable juice was poured into a clean and dry glass beaker. A Mettler Toledo MP230 pH meter with Inlab 413 pH probe was used for the pH measurement (Mettler Toledo Int., Columbus, OH). The pH meter was calibrated using buffer solutions of pH 4, 7 and 10. The pH probe was immersed into the sample completely to obtain pH value. The pH level of each sample was obtained in at least three replicates. The average of the obtained pH value was recorded as the pH of the vegetable juice sample.

A freshly prepared 0.1 M NaOH solution was used for determining the TA of the samples. Incremental addition of NaOH solution neutralized the vegetable juice samples and corresponding gradual increase of pH were recorded until the pH reached 7. During the titration procedure the proper mixing of vegetable juice samples with 0.1 M NaOH solution was confirmed by using a Corning PC-320 Hot Plate Stirrer (Corning Inc., Corning, NY). The required cumulative volume of NaOH solution for neutralizing the samples (pH 7) was recorded as the TA. A sample of the data collection process is provided in Appendix 3.

The recorded data of the pH level of the vegetable juice samples were compared with the critical pH of enamel (5.2 – 5.5) and dentin (6.7) to evaluate the erosive potential of the vegetable juice.

3.3 RESULTS

The measured pH values of all the vegetable juice samples were found to be in the range of 4.0-4.2. The recorded pH and titratable acidity (TA) values of all the vegetable juice brands are listed in Table 3.1 along with their ingredient information. A comparison of the critical erosive pH of enamel and root dentin with the obtained vegetable juice pH values is shown in Figure 3.1. All brands of vegetable juices had pH values significantly lower than the critical pH of root dentin and enamel.

The TA values ranged from 33.9 mL of 0.1 M NaOH (V8 Original Vegetable Juice) to 81.67 mL (RW Knudson Very Vege) as presented in Table 3.1 and figure 3.2. The TA values of vegetable juices are three orders of magnitude higher than the titratable acidity of previously tested commercially available bottled drinking waters and well waters. Thus neutralization of such acidic juices will take a longer time and a high buffering capacity which indicate significant erosive challenge and large contact time.

Table 3.1. pH and titrable acidity (TA) of vegetable juice samples

Sample No.	Brands	Ingredients	pH	TA (ml of 0.1 M NaOH)
1	RW Knudsen Simply Nutritious Mega Green	Filtered water, apple juice concentrate, banana puree, pineapple juice concentrate, peach and mango purees, spirulina, chlorella, broccoli and spinach powders, ascorbic acid, gellan gum, natural flavor, peach juice concentrate.	4.03 (± 0.03)	38.2(± 4.70)
2	V8 Original Vegetable Juice	Reconstituted vegetable juice blend (water and concentrated juices of tomatoes, carrots, celery, beets, parsley, lettuce, watercress, and spinach), ascorbic acid, citric acid, natural flavors.	4.13 (± 0.04)	33.9(± 1.9)
3	Campbell's Tomato Juice	Tomato juice from concentrate (water, tomato concentrate), ascorbic acid, citric acid, malic acid, salt.	4.09 (± 0.03)	36.8(± 1.11)
4	Harris Teeter 100% Tomato Juice	Tomato juice from concentrate (water, tomato concentrate), ascorbic acid, citric acid, salt.	4.16 (± 0.19)	45.5(± 12.73)
5	Food Lion 100% Tomato Juice	Tomato juice from concentrate (filtered water, tomato concentrate), salt, ascorbic acid, citric acid	4.04 (± 0.13)	51.77(± 9.62)
6	Mott's Garden Blend 100% Vegetable Juice	Water, tomato paste, pepper juice, celery juice, vinegar, carrot juice concentrate, clarified carrot juice concentrate, sea salt, onion juice, ascorbic acid, dill juice, parsley juice, carrot juice, natural flavor, onion and garlic powder, soy lecithin	4.13	49.5
7	RW Knudsen Organic Tomato	Filtered water, organic tomato concentrate, organic lemon juice concentrate, sea salt.	4.1(± 0.09)	79.17(± 10.27)
8	RW Knudsen Very Vege	Filtered water, organic tomato paste, organic vegetable juice blend (carrot, celery, parsley and beet juices; organic lettuce, Watercress and spinach juices, citric acid), organic grain vinegar, organic lemon juice concentrate, organic dehydrated green bell peppers, sea salt.	4.05(± 0.06)	81.67(± 7.05)
9	Great Value 100% Vegetable Juice	Filtered water, tomato paste, reconstituted vegetable juice blend (filtered water, concentrated juice of carrots, celery, beets, parsley, lettuce, watercress, and spinach), salt, ascorbic acid, citric acid, natural flavor.	4.27(± 0.09)	45.17(± 4.91)

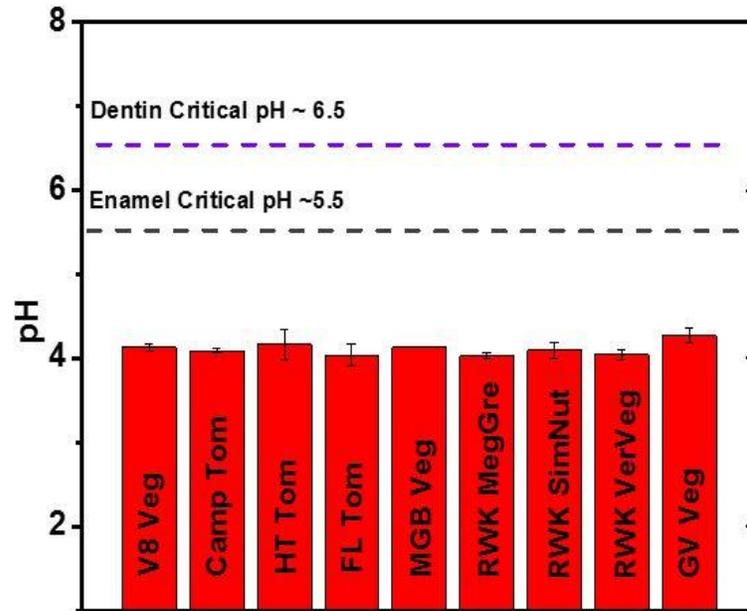


Figure 3.1. pH values of 9 vegetable juice brands and comparison with critical erosive pH of enamel (5.2-5.5) and root dentin (6.7). All brands indicate higher potential for enamel and root dentin dissolution.

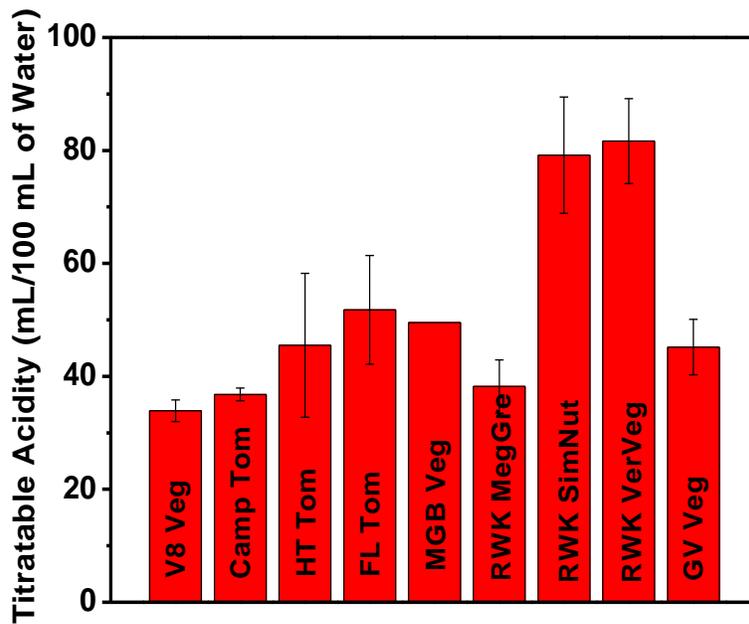


Figure 3.2. Titratable acidity (TA) of 9 vegetable juice brands in mL 0.1M NaOH

3.4 DISCUSSION

Dental hard tissue loss via dissolution has been attributed to low pH values of acidic drinks. Thus, it is evident that acidic vegetable juice samples certainly have a higher potential to cause erosive tooth wear. This risk for erosion can be coupled with a slurry thickness and a high viscosity of the vegetable samples as compared to soft drinks or water. Higher viscosity of drinks has been shown to cause more enamel loss (Aykut-Yetkiner et al., 2014). The high viscosity increases the contact time of the vegetable juice with the tooth surface.

The composition of all vegetable juice samples includes a wide range of various vegetable groups, while the negative control (RW Knudsen Simply Nutritious Mega Green) contains a blend of different fruits. However, pH levels of all nine juice samples are relatively close to each other with a low standard deviation of 0.02-0.1. This similarity in the average pH range of all vegetable juice samples is likely due to the addition of extra acidic substance such as citric acid in the juice to elevate the palatability of the samples. Online reports show that additional citric acid addition is a common practice. This high citrus content can create additional risk factors of chelating the calcium substances from the vegetable juice contents thus binding the calcium and the solution remains unsaturated contributing towards dental erosion.

It is to be noted that the company provided composition of the vegetable juice samples does not include calcium ions. Presence of calcium ions or minerals in diet or drinks has shown to reduce the dental erosion via enhancing the degree of saturation. Dissociation of hydroxyapatite, $\text{Ca}_5(\text{PO}_4)_3(\text{OH})$, only happens if the suspension or saliva is unsaturated i.e. destitute of calcium or phosphate ions. Therefore, the absence of calcium in the vegetable juice samples can predict a high risk of enamel and dentin dissolution. However, the measurement of free calcium ions needs to be pursued in future research to confirm such a mechanism.

Vegetable juices have been increasingly popularized and promoted to serve the purpose of healthy living. Not only lay persons, but also physicians and nutritionists frequently recommend vegetable juices, not knowing the possible relation with erosive tooth wear. Generally five or more portions of fruits or vegetables have been associated as part of a healthy diet for most people; replacement of solid vegetables with liquid formulations (juices) has increased due to the ease of use and as an encouragement for young people. Such a high frequency of usage of vegetable juice with low pH, high TA, high viscosity, and contact time may contribute to a high risk of erosive tooth wear.

2.5 SUMMARY AND CONCLUSIONS

The study results indicate:

1. All vegetable juice brands tested have pH values (4.0-4.2) that are well below the critical erosive pH of enamel and dentin
2. Titratable acidity (TA) values are variable for all of the tested samples, however, all are extremely high
3. Compositions indicate presence of acidic additives which may enhance the tooth dissolution process

Further in vitro and in vivo studies are required to determine the actual erosion ability of these commercial vegetable juices.

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CHAPTER 4

CONCLUSIONS AND FUTURE RECOMMENDATIONS

The overall premise of the present investigation is directed towards the evaluation of the dental erosive potential from drinks including vegetable juice and bottled drinking water. It was evident that while some brands of bottled water presented with a pH lower than the critical erosive pH of enamel and root dentine, all vegetable juice samples had significantly lower pH values. However, the high titratable acidity measured for vegetable juice indicates a high erosive potential while bottled water presented with minimal titratable acidity indicating probable erosion only due to continuous and long-term exposure. Well water samples on the other hand presented with low pH but had extremely low titratable acidity and presence of mineral content to provide protection from teeth demineralization. The study indicates that some bottled water brands that have a pH lower than the critical pH of root dentine may raise potential risks for the individuals who drink the brands very frequently especially those with xerostomia or dry mouth conditions. The lack of minerals in the low pH group of bottled water may cause a higher rate of dental hard tissue dissolution due to lower degree of saturation.

On the contrary, the constant low pH values of vegetable juice in different commercial brands indicate the higher risk of dental erosion coupled with excessively higher titratable acidity. Taste enhancement via additions of acidic concentrates may have been at the core of the acidic nature of vegetable juice.

Therefore, while campaigns for healthy living in modern days recommend intake of commercial vegetable juice or purified bottled drinking water, their dental erosion potential cannot be ignored. Future in vitro or in vivo studies are required to assess the exact erosion capabilities of these studied drinks. Moreover, the effect of additional calcium or other mineral content in bottled water and vegetable juice brands on erosion should also be evaluated to deduce preventive techniques. Parameters including consumption frequency, salivary flow rate, viscosity of vegetable juice, etc. should be incorporated in the experimental matrices for systematic and mechanistic understanding of the erosive impacts on tooth.

Finally, awareness among the general population regarding such erosive effects of seemingly benign and healthy drinks should be raised.

APPENDIX 2.1: SAMPLE DATA COLLECTION FOR BOTTLED WATER

Sample data collection for pH and titratable acidity of Penta water

Ph 8.A(i)-Penta : 5.377

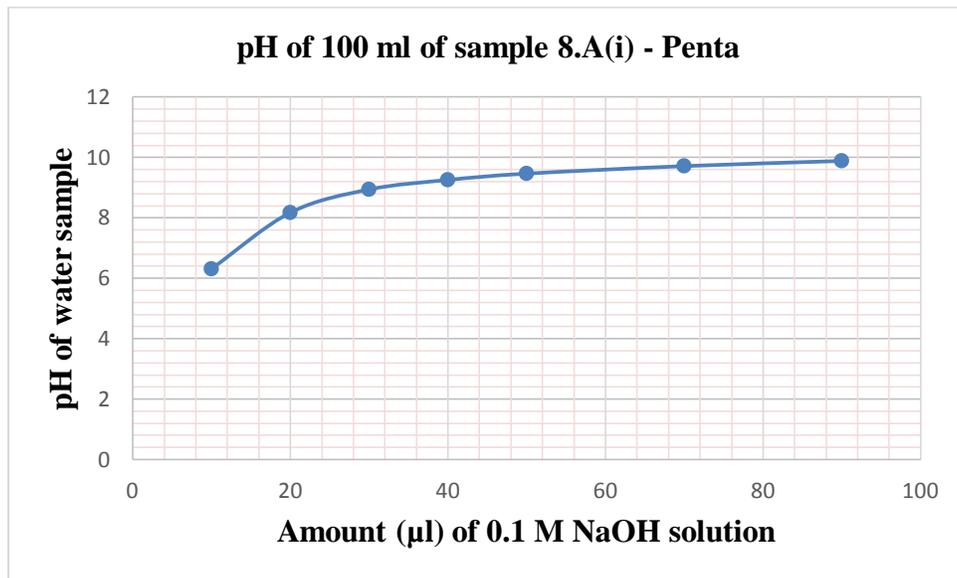
5.36

5.344

5.259

Average pH: 5.335

<u>Added NaOH volume in μl</u>	<u>pH of 100 ml 8.A(i)</u>
10	6.313
10	8.176
10	8.944
10	9.26
10	9.47
20	9.719
20	9.89



APPENDIX 2.2: SAMPLE CALCULATION FOR DEGREE OF SATURATION

Bottled Drinking Water Brand: Icelandic Glacial

Measured pH: 7.42; OH⁻ concentration, [OH⁻] = 2.63×10⁻⁷M

Total Dissolved Solids (TDS): 62 mg/L (Icelandic Glacial Water Quality Report)

Calcium Concentration, [Ca²⁺] = 6 mg/L (Icelandic Glacial Water Quality Report) = 0.00015 M

Phosphate concentration, [PO₄³⁻] = 0.1 mg/L = 1.05×10⁻⁶ M (assumed, not given in water quality report but this is the regulatory limit for pollution as per Environmental Protection Agency)

Ionic strength of water, $I = 2.5 \times 10^{-5} \times \text{TDS} = 0.00155$

If activity coefficient of an ion is γ_i , then $\log \gamma_i = \frac{0.5 (Z_i)^2 I^{1/2}}{1 + I^{1/2}}$

Where Z_i = valence of the ion

Activity Co-efficient of Calcium, $\gamma_{\text{Ca}} = 10^{\log \gamma_{\text{Ca}}} = 1.191$

Activity Co-efficient of Phosphate, $\gamma_{\text{PO}_4} = 10^{\log \gamma_{\text{PO}_4}} = 1.48$

Activity Co-efficient of hydroxyl, $\gamma_{\text{OH}} = 10^{\log \gamma_{\text{OH}}} = 1.045$

Active Concentration of Calcium, $\{\text{Ca}^{2+}\} = \gamma_{\text{Ca}}[\text{Ca}^{2+}]$

Active Concentration of Phosphate, $\{\text{PO}_4^{3-}\} = \gamma_{\text{PO}_4}[\text{PO}_4^{3-}]$

Active Concentration of Hydroxyl Ion, $\{\text{OH}^-\} = \gamma_{\text{OH}}[\text{OH}^-]$

Ion Activity Product, $\text{IAP} = \{\text{Ca}^{2+}\}^5 \{\text{PO}_4^{3-}\}^3 \{\text{OH}^-\}$

Putting all the values, $\text{IAP} = 1.89 \times 10^{-43} \text{ mole}^9 \text{L}^{-9}$

Solubility Product of Enamel, $k_{\text{sp}} = 5.5 \times 10^{-55} \text{ mole}^9 \text{L}^{-9}$

Degree of Saturation, $\text{DS} = (\text{IAP}/K_{\text{sp}})^{(1/9)} = 19.13283$

APPENDIX 2.3: TASTE PREFERENCE SURVEY OF BOTTLED WATER

Consent Form IRB study #14-2187; exempted 10/30/2014

You are taking part in the research survey of bottled drinking water. Please read this form carefully and ask any questions you may have before agreeing to take part in the study.

Study background:

The aim of this study is to determine taste preferences of different commercially available bottled drinking water.

What we will ask you to do:

If you agree to be in this study, you will be asked to taste 30 mL samples from each of 8 different commercial brands of bottled drinking water. The brands on the samples will not be disclosed to you before the test and will be coded with numbers. After each sample tasting you will be asked to mention the pleasantness or unpleasantness by marking a point on the Visual Analogous Scale (VAS) with 0 for most unpleasant and 10 for most pleasant sample. Please mention the respective sample code number on your response paper. It may take about 5 minutes to participate in this study.

Risks and benefits:

We do not anticipate any risks to you participating in this study other than those encountered in day-to-day life.

There are no personal benefits to you. Through this study we hope to learn more about the factors affecting the population to choose different brands of bottled drinking water.

Confidentiality of the participants:

This is an anonymous survey. To keep your records confidential no information regarding your identity will be disclosed in any situation. During any type of publication we will not disclose your identifying information. Research records will be kept in a locked file for 2 years; only the researchers will have access to the records.

Completely voluntary participation:

It is completely voluntary to participate in this study. You have right to withdraw from the study at anytime for any reason. If you decide not to take part or to withdrawal yourself from this study, it will not affect your current or future relationship with the researchers.

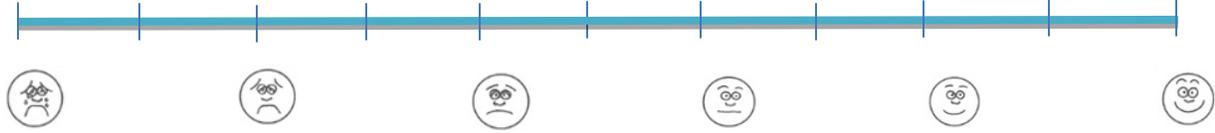
Contact us:

Dr. Upoma Guha (upoma_guha@unc.edu Phone: 919-537-344) and Dr. Terence Donovan (terry_donovan@unc.edu Phone: 919-537-3983) are conducting this research from the Department of Operative Dentistry, University of North Carolina at Chapel Hill. Please contact the researchers in any question regarding the research.

Taste assessment of different samples of drinking water: Visual Analogous Scale (VAS)

1. Sample code

0 1 2 3 4 5 6 7 8 9 10



Unpleasant

Pleasant

2. Sample code

0 1 2 3 4 5 6 7 8 9 10



Unpleasant

Pleasant

3. Sample code

0 1 2 3 4 5 6 7 8 9 10

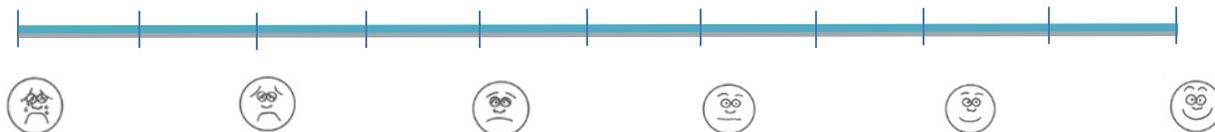


Unpleasant

Pleasant

4. Sample code

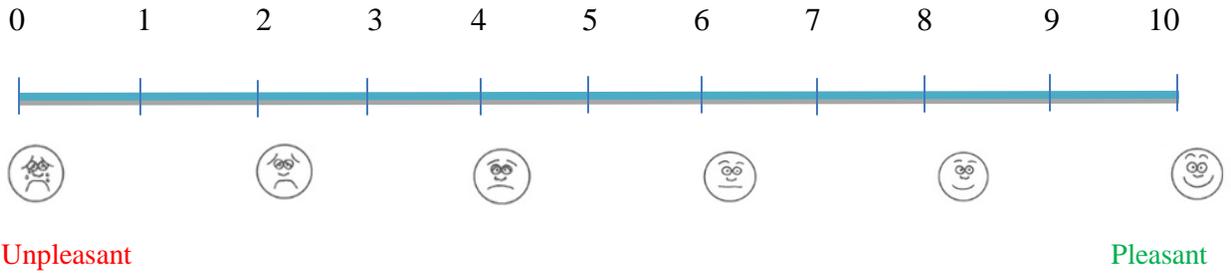
0 1 2 3 4 5 6 7 8 9 10



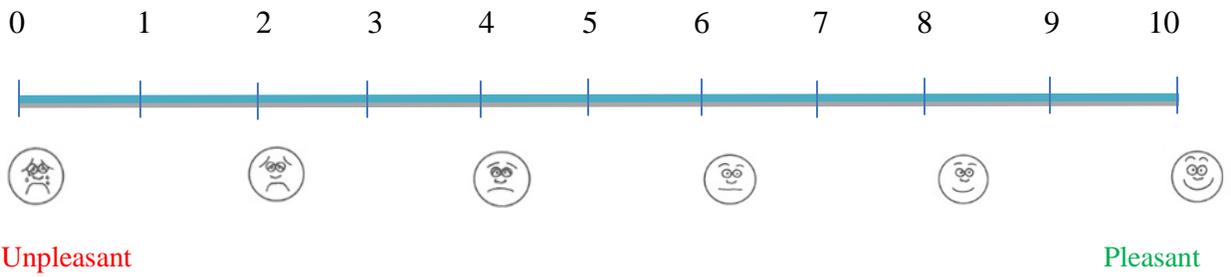
Unpleasant

Pleasant

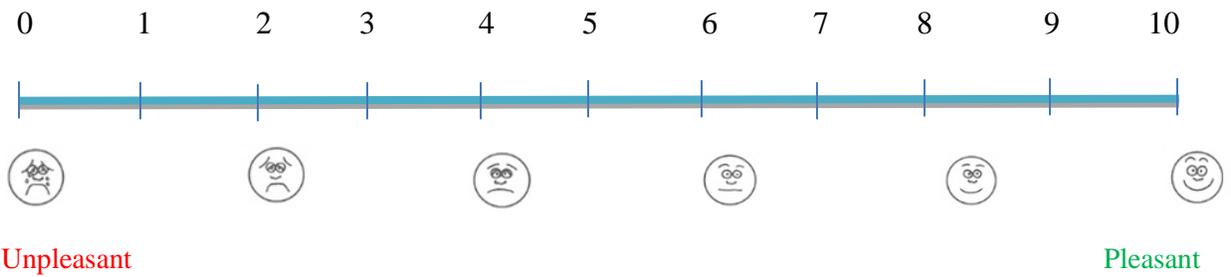
5. Sample code



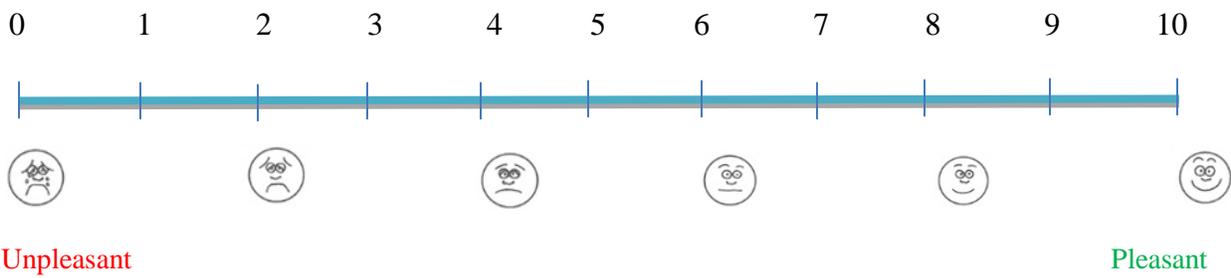
6. Sample code



7. Sample code



8. Sample code



APPENDIX 2.4: DEMOGRAPHIC QUESTIONNAIRE SURVEY

Drinking Water Study Consent Form

You are taking part in the research survey of bottled drinking water. Please read this form carefully and ask any questions you may have before agreeing to take part in the study.

Study background:

The aim of this study is to determine the factors of popularity of bottled drinking water.

What we will ask you to do:

If you agree to be in this study, you will be asked to answer 25 questions regarding your water consumption habit. It may take about 10 minutes to participate in this study.

Risks and benefits:

We do not anticipate any risks to you participating in this study other than those encountered in day-to-day life.

There are no personal benefits to you. Through this study we hope to learn more about the factors affecting the population to choose different brands of bottled drinking water.

Confidentiality of the participants:

This is an anonymous survey. To keep your records confidential no information regarding your identity will be disclosed in any situation. During any type of publication we will not disclose your identifying information. Research records will be kept in a locked file for 2 years; only the researchers will have access to the records.

Completely voluntary participation:

It is completely voluntary to participate in this study. You have right to withdraw from the study at anytime for any reason. If you decide not to take part or to withdrawal yourself from this study, it will not affect your current or future relationship with the researchers.

Contact us:

Dr. Upoma Guha (upoma_guha@unc.edu Phone: 919-537-344) and Dr. Terence Donovan (terry_donovan@unc.edu Phone: 919-537-3983) are conducting this research from the Department of Operative Dentistry, University of North Carolina at Chapel Hill. Please contact the researchers in any question regarding the research.

1. Please specify your gender
 - Male
 - Female
 - Not interested to mention

2. Your age
 - Under 18 years
 - 18-24 years
 - 25-34 years
 - 35-44 years
 - 45-54 years
 - 55-64 years
 - 65 years or older

3. Highest level of education you have completed
 - Less than high school
 - High school
 - Some College
 - 2 year College degree (associates)
 - Bachelors degree
 - Masters degree
 - Doctoral degree

4. Your ethnicity
 - White
 - Hispanic or Latino
 - Black or African American
 - Native American or American Indian
 - Asian / Pacific Islander
 - Other

5. Annual income in dollars
 - Below 10K
 - 10K +
 - 20K+
 - 30K+
 - 40K+
 - 50K+

6. Which on in the following describes your physical activity? (Select each that applies)
 - Play any form of sport (Specify:-----)

- Work out in gym
 - Other form of exercise
 - No exercise at all
7. How fit are you?
- Very unfit
 - Unfit
 - Fit
 - Very fit
8. How much water do you drink per day?
- Less than 1 L
 - 1-2 L
 - 2-3 L
9. Where do you get the drinking water from? (Select all that apply)
- Well
 - Bottled drinking water
 - Tap water
 - Fountain
 - Filtered water
10. Do you drink bottled drinking water?
- Yes
 - No (Completion of the survey)
11. Which brand of bottled drinking water do you prefer? (Select all that apply)
- Dasani
 - Aquafina
 - Penta water
 - Nestle pure life
 - Nestle Deer park
 - Smart water
 - Resource
 - Fiji
 - Evian
 - Icelandic Glacial
 - Icelandic Natural spring water
 - Volvic
 - Eternal
 - Other.....

12. Do you think bottled water is convenient for drinking?
- Yes
 - No (Skip question no 14)
13. Why do you think bottled water is convenient for drinking?
- Cleanliness
 - Availability
 - Healthier
 - Contains minerals
 - Taste
 - Ease of use
 - Price
14. Where do you buy the bottled drinking water from?
- Walmart
 - Harris teeter
 - Food lion
 - Vending machine
 - Gas station
 - Other.....
15. How many bottles do you buy at a time?
- 1 bottle
 - 1-5 bottles
 - 6-12 bottles
 - 13-24 bottles/value pack
16. Do you carry the bottle when you go out (e.g. school, gym, shopping)?
- Yes
 - No
17. What do you consider for selecting the brand of the bottle? (Select all that apply)
- Price
 - Review
 - Packaging
 - Nutritional information
 - Flavor/taste
 - Availability
18. Do you look for the information regarding nutritional composition on the packaging before buying?
- Yes
 - No

19. Do you think the bottled waters contain fluoride?
- Yes
 - No
 - I am not concerned about it
20. How many bottles do you drink daily (500 ml)?
- 1
 - 1-3
 - 3-5
 - More than 5
21. How much are you ready to pay for a water bottle?
- \$ 1 - \$1.5
 - \$2 - \$2.5
 - \$3 - \$3.5
 - \$4 - \$6
22. Which of the following media advertisements attract you? (Select all that apply)
- Local newspaper
 - Magazine
 - TV
 - Internet
 - Poster/ flyers
 - Cinema
 - Public Event
 - Sponsorships
 - Other
23. Do you use the bottled drinking water for other purposes other than drinking (e.g. cooking)?
- Yes
 - No
24. Are there any babies under 5 years of age who drink bottled drinking water in your family on a regular basis?
- Yes
 - No
25. Do you consider to buy alkaline bottled drinking water?
- Yes
 - No
 - I am not concerned

APPENDIX 3: SAMPLE DATA COLLECTION FOR VEGETABLE JUICE

pH
 4.141
 4.147
 4.151

Average. pH: 4.15

<u>Added NaOH vol μl</u>	<u>Cumulative NaOH vol μl</u>	<u>pH of 100 ml I.A</u>	<u>Temp -C</u>
0	0	4.151	22.5
200	200	4.158	22.1
400	600	4.181	22
500	1100	4.198	21.7
600	1700	4.223	21.8
600	2300	4.24	22
700	3000	4.275	21.9
800	3800	4.272	22
900	4700	4.283	21.3
1000	5700	4.328	21.9
1000	6700	4.369	22
1000	7700	4.412	22
1000	8700	4.449	22.1
1000	9700	4.498	21.4
1100	9800	4.551	21.9
1200	11000	4.613	22
1300	12300	4.67	20.9
1400	13700	4.734	21.3
1500	15200	4.844	21.8
1600	16800	4.933	21.2
1700	18500	5.041	21.2
1800	20300	5.175	21.5
1900	22200	5.368	21.8
1900	24100	5.548	21.5
1900	26000	5.761	21.7
1900	27900	6.04	21.8
1500	29400	6.342	21.7
1000	30400	6.514	21.7
900	31300	6.902	21.7
500	32300	7.166	21.5

900	33200	7.374	21.6
1000	34200	7.591	21.6
1500	35700	7.948	21.5
1500	37200	8.209	21.5
1500	38700	8.402	21.5
1500	40200	8.563	21.3
1500	41700	8.683	21.2
1500	43200	8.829	21.4
1500	44700	8.944	21.2
1500	46200	9.037	21.3
1500	47700	9.123	21.3

