



School of  
Dental Medicine

**The Effect of Different Beverages on the Microhardness of UltraSeal XT<sup>®</sup> Hydro:  
An In Vitro Study.**

A Thesis

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By

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## **DEDICATION**

To my parents Abdullah and Masuda, who's their affection, love, encouragement, and prays made me able to reach this stage

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## **Abstract**

### **Aim**

The aim of this study is to evaluate the influence of different beverages on the microhardness of UltraSeal XT<sup>®</sup> hydro.

### **Materials and Methods**

Samples of the resin sealants were prepared and immersed in milk, Coca-Cola, orange juice, Gatorade, or, as control groups, in artificial saliva and samples without immersion. Specimens were divided into six groups by beverage type (n = 10 per group). Specimens were then immersed in the beverage for 14 days. An indenter was used to perform the Vickers microhardness test on the surface of the resin sealants. Three indentations were performed under a load of 200 g for 15 s. A specimen was selected from each group for qualitative scanning electron microscope assessment.

### **Results**

The median (inter-quartile range) microhardness for the saliva group was 59.6 (1.2) VHN; for the orange juice group 56.0 (1.1) VHN; for the Coca-Cola group 51.1 (0.9) VHN; for the Gatorade group 52.7 (0.9) VHN; for the milk group 61.4(1.8) VHN; and for the group of specimens that not immersed in any solution 63.9 (3.1) VHN. Groups that showed statistically significant differences were: Coca-Cola-saliva, Coca-Cola-

milk, Coca-Cola-not immersed, Gatorade-milk, Gatorade-not immersed, and orange juice-not immersed. Differences were not statistically significant for the other comparison groups.

### **Conclusion**

Within the limitations of this study and based on the results, the microhardness of UltraSeal XT<sup>®</sup> hydro decreased more after exposure to beverages with the lowest pH level. Coca-Cola, Gatorade, and orange juice had the greatest effect on the microhardness.

## **Introduction:**

### **Dental Caries**

Dental caries is the most common chronic disease of childhood.<sup>1</sup> It occurs when the balance between demineralization and remineralization is disrupted. This begins when the bacterial biofilm is exposed to fermentable carbohydrates over time, leading to an ecological shift in the composition and activity of the tooth.<sup>2</sup> Even though for many children tooth decay has been declining in permanent teeth since the 1960s, recent findings show that between 1988 and 2004, there was an increase from 24% to 28% in the prevalence of dental decay in the primary teeth of preschool children.<sup>3</sup> Data from that the National Health and Nutrition Examination Survey (NHANES) in the US show the prevalence of dental decay between 2011 and 2012 in primary and permanent teeth of children of different age groups. The prevalence of caries was almost 37% in deciduous teeth for a child aged 2-8 years, and 14% of this age group had untreated tooth decay. The prevalence of caries was almost 21% in the permanent teeth of children aged 6-11 years, and 6% of this age group had untreated tooth decay.<sup>4</sup> The consequences of failing to treat tooth decay may include toothache and infection, which can lead to eating, speaking, and learning problems.<sup>5</sup> Pit and fissure caries account for approximately 90% of all caries that affecting the permanent posterior teeth,<sup>6</sup> and pit and fissure sealants can help to prevent a significant amount of this decay.<sup>7</sup>

## **Pit and Fissure Sealants**

Pit and fissure sealants are materials that micromechanically bond to the occlusal pits and fissures of teeth, thus helping to preventing nutrients from reaching cariogenic bacteria present in the fissures.<sup>8</sup> Buonocore performed one of the first studies on sealants and reported that caries can be prevented by sealing pits and fissures.<sup>9</sup> A study by Burt recommended that sealants be placed as soon as possible after the eruption of molars to prevent occlusal caries.<sup>10</sup>

In 2008, Ahovuo-Saloranta et al. conducted a systematic review and found that resin-based sealants in children and adolescents reduced the incidence of caries by up to 86% after 12 months, and 57% at 48-54 months.<sup>11</sup> Another systematic review in 2013 evaluated caries in sealed and non-sealed teeth and found that sealants reduce the development of occlusal caries in permanent molars by up to 48 months.<sup>12</sup> A study by Griffin et al. assessed how effective dental sealant is in stopping dental decay progression. They reported that applying sealant on non-cavitated dental decay in permanent teeth is effective in stopping dental decay progression.<sup>13</sup> A comparison between the use of sealants and a six-monthly application of fluoride varnish to prevent occlusal caries revealed that the sealants and topical fluoride application offer better protection than topical fluoride alone.<sup>14</sup> However, the effectiveness the resin-based sealant had on reducing caries in permanent first molars depended on the retention in pits and fissures.<sup>15</sup> Furthermore, the success of sealants was related to moisture control, which is almost impossible to control in partially erupted molars or uncooperative patients. As such, it is one of the main problems that practitioners encounter, especially when using

resin-based sealants. To overcome these issues, glass ionomer was introduced, although this has a lower retention rate than resin-based sealants.<sup>16</sup> However, the American Dental Association (ADA) continues to recommend the use of resin-based sealants as the preferred treatment mode.<sup>17</sup>

Many studies have examined the failure of pit and fissure sealants in the cases of permanent molars that need to be replaced or restored. A study in Finland, Sweden, and Greece that compared sealed and non-sealed teeth and the frequency at which each group required replacing with a restoration, found after 7-10+ years follow-up that restorations were required in 60-80% of the cases that had not been sealed versus 30-40% of those that had.<sup>18</sup> Simecek et al. reported a 12.2% failure rate of sealed teeth at the 35-month follow-up stage. The percentage of teeth requiring a restoration was 7.5%, and the percentage of teeth requiring a sealant replacement was 4.7%.<sup>19</sup> Studies also examined failure rates amongst different materials used as sealants. A study comparing the success rate of Fuji VII and resin sealants in low-risk patients found that the percentage of successful sealants using Fuji VII was 31.2%. In contrast, the percentage of successful resin sealants was 77.1%.<sup>20</sup> Several additional factors are typically associated with the failure of sealants. One follow-up study of between 2-6.7 years found that the failure of sealants is associated with a history of caries. Also, this study compared sealants placed by residents and dental students and whether there was any difference in clinical outcome. The authors concluded that no difference was found between the two groups regarding successful outcomes. This might be because patients of the dental students group were older and had less history of caries.<sup>21</sup> Another significant factor contributing

to the success of sealants is the follow-up care. The success of sealants with periodic recalls and maintenance is 80% to 90% after ten years .<sup>22</sup>

### **Prevalence of pit and fissure sealants**

Data from the 2011-2012 (NHANES) highlighted the prevalence of sealant-use in primary and permanent teeth of people of different age groups. Approximately one-half of children aged between 9 and 11 had at least one sealant in place on permanent teeth. The prevalence of sealant for children aged 6-8 years was 31%; and 43% of adolescents aged 12-19 had at least one permanent tooth with a dental sealant.<sup>4</sup>

### **Classification for pit and fissure sealant materials**

Anusavice and colleagues proposed four classifications of sealant materials. The first is bisphenol A-glycidyl methacrylate or urethane dimethacrylate resin-based sealant. This type of sealant comes as a filled or opaque material or as an unfilled or transparent material. The second type is glass ionomer cement (GIC) sealants which consists of an aqueous-based polyacrylic acid solution and a fluoroaluminosilicate glass powder. The advantage of using GIC as a sealant is that it offers fluoride-release properties. The third type of material is known as compomers or polyacid modified-resin sealants. These offer a combination of the fluoride properties found in GI sealants and the advantages of the resin materials found in tradition resin-based sealants. The last type is a combination of GI sealants and resin components and is commonly known as resin-modified GI sealants. In addition to offering less sensitivity to water than traditional GI sealants, this type of sealant offers the fluoride-release properties of GI sealants.<sup>23</sup>

## **UltraSeal XT<sup>®</sup> hydro**

UltraSeal XT<sup>®</sup> hydro is a resin-based pit and fissure sealant developed by Ultradent products, USA. It is a light-cured, hydrophilic, and self-adhesive sealant.<sup>24</sup> A radiopaque material, it contains methacrylate-based, thixo-tropic diurethane dimethacrylate, triethylene glycol dimethacrylate, and methacrylic acid.<sup>25</sup> Additionally, inorganic filler particles comprises 53wt% of the sealant. Previous research has reported that UltraSeal XT<sup>®</sup> hydro eliminates moisture-related failure because it chases the moisture into pits and fissures.<sup>24</sup>

## **Saliva**

Saliva is a fluid that surrounds oral and soft tissues and consists of individual liquids and components derived from several sources. The major and minor salivary glands produce the bulk of saliva, with nonglandular sources, such as crevicular fluid, oral microorganisms, and diet-related components, making a minor contribution.<sup>26</sup> There are many factors that affect the role of saliva, such as the flow rate and the pH level. When the subject swallows, the flow of saliva eliminates food debris and bacteria from the mouth. After food consumption, the clearance of sugars and acid increases if the saliva flow rate increases.<sup>27</sup> Three major systems in mouth saliva that play a role in the buffer capacity are the phosphate, the bicarbonate, and the protein buffer systems.<sup>28</sup> The drop of plaque pH creates an environment that facilitates the growth of acidophilic microorganisms such as *Streptococcus Mutans* and the *Lactobacilli*. This produces the ideal conditions for caries to develop and, with a further fall in pH, creates areas of demineralization of the dental enamel.<sup>29</sup> These factors can affect the performance of

restorative materials by softening them, increasing surface roughness, enhancing tooth erosion, and making them more susceptible to wear.<sup>30</sup>

The critical pH value of saliva is 5.5. When the pH level drops below 5.5, the process of caries development commences due to the loss of minerals from the tooth structure (demineralization). When the pH value increases, redistribution of mineral occurs (re-mineralization).<sup>31,32</sup> Diet also plays a role in the caries process and can either reduce the pH level further (acidic diet), or help to increase it.<sup>33</sup>

### **Sugar-sweetened beverages**

There was an increase in the prevalence of snackers among children aged between 2-12 years. The prevalence was 74% in 1977-1978 and rose to 98% in 2003-2006.<sup>34</sup> In the US, more than 90% of the population consumes discretionary foods and almost 50% drink sugar-sweetened beverages (SSB).<sup>35,36</sup> Higher consumption of SSB is not only related to dental problems but is also linked to health issues, such as weight gain and type 2 diabetes.<sup>37,38</sup> Between 2009 and 2010, almost half of US adults consumed one or more SSB a day.<sup>39</sup> In addition, between 2011 and 2014, at least one sugar-sweetened beverage a day was consumed by 62.9% of the young people. SSBs contributed 4.1% of the total daily calories consumed by boys aged between 2 and 5, 6.6% consumed by boys aged between 6 and 11, and 9.3% consumed by boys aged between 12 and 19.<sup>40</sup>

## **Acidic Dissolution**

The behavior of dental material can be directly impacted by the environmental conditions in the oral cavity, such as temperature and acidity. As such, it is important that dental material exhibits minimal changes in response to variations in conditions.<sup>41</sup> The extent to which restorative material is resistant to dissolution is one of the most significant properties and plays a fundamental role in the durability of a restoration.<sup>42</sup> Acidic dissolution of the tooth surface or the dental materials that result from chemical processes as opposed to the presence of acids of bacteria plaque origin is known as dental erosion.<sup>43</sup> Erosion of intrinsic origin, such as gastric acid exposure, occurs due to vomiting, regurgitation, or GERD.<sup>44</sup> It can also be caused by extrinsic sources, such as acidic foods and beverages with low pH, which may induce pronounced intermittent or continuous erosive wear of dental materials.<sup>45</sup> For these materials to be clinically successful, they need to offer a high degree of resistance to wear.<sup>46,47</sup>

## **Surface Topography**

Any irregularity of the surface texture that develops during the manufacturing process or results from the material condition is known as surface roughness,<sup>48</sup> and many methods can be used to measure it. These include scanning electron microscopy (SEM), contact stylus tracing, non-contact laser stylus metrology, atomic force microscopy (AFM), and 3D optical profilometry.<sup>49</sup> High-resolution images of the surface of a sample are produced by scanning electron microscope. These black and white images have a sharp focus over a great depth of field with a three-dimensional appearance.<sup>50</sup> The most common parameter used in dentistry to measure surface roughness is roughness average

(Ra).<sup>51,52</sup> It is defined as the arithmetic average of all deviations of the profile of the roughness from a line in the center.<sup>51,53</sup>

### **Aging Protocol**

Different aging protocols have been employed in previous studies. A report assessed the effect of the temperature of beverages on the microhardness (MH) of composite resin. The researchers immersed specimens in test solutions for 15 min three times a day (morning, afternoon, and night) for 30 days.<sup>54</sup> Another study assessed the effect of alcoholic beverages on the MH of three different composite resins. The authors immersed specimens for 15 mins in different liquids, three times a day over 30 days.<sup>30</sup> A further study evaluated the effect of lactic and acetic acid on the MH of different composite materials. Samples were stored in an incubator at 37C for seven days.<sup>55</sup> A further study assessed hardness changes after bulk-fill and conventional resin-composite materials were stored in ethanol. Aging protocol was used to assess hardness changes after immersing the materials for 7, 30, and 90 days.<sup>56</sup> A report examined the efficacy of staining solutions on nanofilled resin composite. Authors stored specimens for 24 h in coffee, red wine, or distilled water.<sup>57</sup>

### **Microhardness**

The environment of the mouth and masticatory force puts dental materials under mechanical stresses such as flexion and compression. To ensure long-term clinical success, the materials need to meet a broad range of specifications.<sup>58</sup> It is important to assess surface hardness because this provides an indication of a material's strength and its

ability to withstand abrasion from opposing dental structures or any chemical soft drink.<sup>59</sup> Hardness tests use a sharp point or abrasive particle to produce an indentation on the surface of the material and assess numerous properties such as, compressive strength and ductility. Several types of surface hardness tests are in common use including Brinell, Vickers, and Knoop, and the majority assess the extent to which a material can resist penetration by diamond point or steel ball under specific load. This test uses a square-based pyramid and is calculated by dividing the load by the projected area of the indentations.<sup>61</sup> Dental materials, such as resin sealants, are subject to abrasion from opposing teeth and materials, or may exhibit chemical softening due to exposure to the beverages children often drink. This might influence the clinical durability of sealants.<sup>59,</sup>

62

Many studies have examined the MH for different resin composite materials. A report assessed the effect of beverage temperature on the MH of composite resin. Each sample was immersed in coffee, cola, or artificial saliva at different temperature degrees of each beverage. The researchers reported higher MH values in the artificial saliva group than in the other groups. There was also a significant difference in the MH after immersing composite resins in coffee and cola.<sup>54</sup> Another study was conducted to determine the filler content and the MH of various flowable resin composites. The authors concluded that the MH of the flowable resin restoration is affected by the filler content.<sup>58</sup> A further study assessed the effect of alcoholic beverages on the MH of three different composite resins. The results revealed that the low pH alcoholic beverages had more effect on the MH values of the composite than alternative alcoholic beverages.<sup>30</sup> A further study assessed MH of different composite materials after immersing them in either lactic or

acetic acid. This study reported a decrease in the MH in all the materials and lactic acid had the greatest effect.<sup>55</sup> One study investigated the effect of staining solutions on a nanofilled resin composite that had previously been in contact with bleaching agents. A total of 135 specimens were fabricated with a nanofilled resin and then allocated into three groups regarding bleaching materials. Specimens were then immersed into one of the following liquids: coffee, red wine, or distilled water. The results revealed that after contacting the composite materials to hydrogen peroxide, they exhibited lower MH values. Also, MH values of specimens immersed in red wine and coffee were lower than those immersed in distilled water.<sup>57</sup> A study was conducted to assess hardness changes after bulk-fill and conventional resin-composite materials were stored in ethanol. After storing the samples in 75% ethanol/water solution, the MH was re-measured. All the materials exhibited a significant reduction in the MH, ranging from 14.5% to 7.2%, after 24 h of storage in 75% ethanol/water.<sup>56</sup> A report was conducted to examine the effect of four mouth rinses on the MH of esthetic restorative material. Forty specimens of resin composite were divided into four groups and ten specimens were immersed in 20 ml of different mouth rinses. The pH level of each mouth rinse was recorded. The results showed that the MH values for the specimens that immersed in the mouth rinse with the lowest pH level were significantly decreased.<sup>62</sup>

A study assessed the surface hardness of the three different composite restorations and whether if they were affected by the pH of the topical fluoride. Composite materials were placed in either 1.23% acidulated phosphate fluoride gel (APF), sodium fluoride mouth rinse, 0.9 neutral fluoride, or distilled water. The pH was 3.9 for the APF gel, 6.5 for sodium fluoride mouth rinse, and 7 for neutral fluoride. The results revealed that MH

decreased significantly for samples that were placed in the APF gel more than for the other two groups.<sup>63</sup> A report assessed whether the acidic pH will decrease the MH of three-tricalcium silicate materials. Samples were divided to four groups. The first three were immersed at three different pH levels of butyric acid (5.4, 6.4 and 7.4). The fourth group was immersed in distilled water. Findings revealed that the MH was higher for the group was immersed in butyric acid at high pH level compared to the group immersed in butyric acid at low pH level. Also, the MH of the group immersed in distilled water was significantly higher than the other groups.<sup>64</sup> A study investigated surface MH of various tooth-colored restoratives and the effects of dietary acids on them. The authors immersed Vitremer, Fuji IX GP, Dyract AP, and Prodigy in lactic, citric, orthophosphoric, acetic acids, or bidistilled water. Results showed that the MH of Vitremer and Fuji IX GP was decreased after immersion in acetic or citric acid. On the other hand, the MH of the same materials was increased after immersion in lactic and orthophosphoric acids. The MH of Prodigy and Dyract AP was decreased by all acidic solutions.<sup>65</sup> Another study, the authors examined the effect of pH on the MH of resin composite, compomer, and giomer. Specimens were stored in 0.3% citric acid at pH 2.5, 3,4,5,6, and 7. Results showed that the effect of pH on the MH of resin fillings was material dependent. Citric acid at low pH affected the composite materials less than they did the compomer and giomer.<sup>66</sup>

In the United States, almost 40% of children aged 6-11 have at least one sealant in a permanent tooth, but the success rate is only 57% after 48-54 months.<sup>4, 11</sup> Consequently, further research is needed to refine the current advice given to parents about sealant durability. Although many studies have examined composite resin sealants, few have investigated the effect of low pH beverages on the MH of resin sealants.<sup>58</sup> For this

reason, the principal purpose of this study is to assess the MH of one such sealant—UltraSeal XT<sup>®</sup> hydro—after exposing it to beverages with different pH levels.

## **HYPOTHESIS**

After exposure to different beverages, the MH of the UltraSeal XT<sup>®</sup> hydro will decrease more with the beverage with the lowest pH level. Coca-Cola, Gatorade, and orange juice are beverages that will have the greatest effect on the MH. Artificial saliva, milk, and non-immersed samples will have the least effect on the MH.

## **SIGNIFICANCE OF THE STUDY**

Results of this study will provide the clinician with understanding of the performance of resin sealants in terms of MH, following a drop in the saliva pH level, which often occurs in patients with chronic beverage intake.

## **MATERIALS AND METHODS**

### **Specimens Preparation**

In this study, we used UltraSeal XT<sup>®</sup> hydro, a resin-based sealant. A total of sixty samples were prepared of the resin sealants. First, resin sealants were applied to a plastic mold (20 mm in diameter and 3 mm in thickness) and a glass slide was placed over it (Figure 6). This was used to ensure no voids and that specimen had a uniform thickness. All specimens were Cured for 20 s using a light emitting diode (LED) curing light (1,100 mW/cm<sup>2</sup>, Bluephase, Ivoclar Vivadent, Schaan, Lichtenstein) (Figure 4).

### **Specimens Cycling**

Specimens were divided into six groups by beverage type. Specimens were then immersed in milk, Coca-Cola, orange juice, Gatorade or, as control groups, in artificial saliva and samples without immersion for 14 days at room temperature. Specimens were kept in the beverage for the period of the study, except when solutions were changed. A pH meter (Orion Star A214, Thermo Scientific, USA) was used to measure the pH value of each solution (Figure 2). Specimens were immersed in 90 mL of tested solutions. The pH of each solution is shown in Table 1.

### **Microhardness Assessment**

An indenter (Buehler, Lake Bluff, IL, USA) was used to perform the MH analysis on the surface of the resin sealants (Figure 3). Three indentations were performed under a load of 200 g for 15 s for each specimen after immersing the specimens in the tested solutions. The Vickers MH value was obtained using the following formula:  $VHN = (1854.4 \sqrt{3 W}) / d^2$  (where VHN is the Vickers hardness number expressed in  $kg/mm^2$ , W is the weight in g, and d is the length of the diagonal in mm). The three readings of the indentations were converted using the conversion table for Knoop and Vickers hardness numbers (Figure 9). The three readings for each specimen were then averaged prior to the statistical analysis.

### **Electron Microscope Assessment**

A specimen was selected from each group for qualitative scanning electron microscope assessment. Isomet 1000, Buehler Ltd was used to section specimens to a size of one-half

inch (Figure 1). Specimens were then completely dried and gold coated with a sputter coater. Analysis was performed using Amary 3300 FE SEM with x10,000 magnification.

### **Power Calculation**

A pilot study (n = 3 per group) was conducted to obtain anticipated values for a power calculation. Based on the results of the pilot study, an effect size of  $\Delta^2 = 9.9$  was assumed in the power calculation. Using the software package nQuery Advisor (Version 7.0), it was determined that a sample size of n = 10 per group was adequate to achieve a power greater than 99% in the main study, alongside a Type I error rate of  $\alpha = 0.05$ .

### **Data Analysis**

Means, medians, standard deviations (SD), and inter-quartile ranges (IQR) were computed. As the data were not normally distributed, statistical significance was determined using the Kruskal-Wallis test (P-value < .05). Post-hoc tests were conducted using Dunn's test with Bonferroni correction (P-value < .003). SPSS version 24 was used in the analysis.

## **RESULTS**

For each group the microhardness results (mean, median, IQR, and SD) are presented in Table 2. The median (IQR) MH for the saliva group was 59.6(1.2) VHN; for the orange juice group 56.0 (1.1) VHN; for the Coca-Cola group 51.1 (0.9) VHN; for the Gatorade group 52.7 (0.9) VHN; for the milk group 61.4(1.8) VHN; and for the group of specimens not immersed in any solution 63.9 (3.1) VHN. A boxplot for each group is shown in Figure 17.

The result of the Kruskal-Wallis test was statistically significant (P-value < .001). Groups that showed statistically significant differences in post-hoc tests were: Coca-Cola-saliva (P-value < .001), Coca-Cola-milk (P-value < .001), Coca-Cola-not immersed (P-value < .001) (P-value < .001), Gatorade-milk (P-value < .001), Gatorade-not immersed (P-value < .001), and orange juice-not immersed (P-value = .001). The results were not statistically significant for the other groups.

SEM images are shown in Figures 11-16. The images were taken at the end of the experiment at x10,000 magnification. We observed differences in the surface topography between the specimens immersed in low pH beverages and those immersed in high pH beverages or those not immersed at all. The surface topography of the Coca-Cola and Gatorade groups were more pitted and eroded than the specimens immersed in milk and those not immersed in any beverage.

## **DISCUSSION**

Approximately, 50% of children aged 9-11 had at least one sealant on their permanent teeth.<sup>4</sup> Resin-based and glass ionomer cement are the two main types of sealants.<sup>63</sup> The ADA continues to recommend the use of resin-based sealants as the preferred treatment mode.<sup>14</sup> The behavior of dental materials can be directly impacted by the environmental conditions of the oral cavity such as temperature and acidity. As such, it is important that dental materials exhibit minimal changes in response to variations in conditions.<sup>41</sup> Wear might happen to dental materials if they are exposed to beverages with a low pH.<sup>43</sup> To be clinically accepted, dental materials need to offer a high degree of resistance to wear.<sup>46, 47</sup> Many studies have assessed the effect of different solutions with different pH levels on

the MH of resin composites.<sup>54, 55,56,57</sup> Our search of the literature yielded only a few studies that assessed the MH of sealants. In this in vitro study, we assessed the effect of different beverages on the MH of UltraSeal XT<sup>®</sup> hydro resin based sealant.

The results of the study support the hypothesis and show that, after exposure to different beverages, the MH of the UltraSeal XT<sup>®</sup> hydro will decrease more with the beverage with the lowest pH level. Therefore, Coca-Cola, Gatorade, and orange juice were beverages with the greatest effect on the MH. Artificial saliva, milk, and samples not immersed in any solution were groups that had the least effect on the MH.

Results show that the lowest median MH number was for specimens immersed in Coca-Cola 51.1 VHN, followed by specimens immersed in Gatorade group 52.7 VHN, and orange juice group 56.0 VHN. In these groups, the pH levels of the solutions were 2.4, 2.9, and 3.9 respectively. This is consistent with the findings in the other studies.<sup>30, 54</sup> One study assessed the effect of alcoholic beverages on the MH of three different composite resins, and reported that the composite specimens with the lowest MH values were exposed to the alcoholic beverage with the lowest pH level.<sup>30</sup> Another report evaluated the effect of beverage temperature on the MH of composite resin. The beverages were coffee, cola, and artificial saliva. The pH level of coffee and cola were lower than that of artificial saliva. The findings showed that the MH values of the samples that were immersed in coffee and cola were lower than those immersed in artificial saliva.<sup>54</sup>

In this research, the highest median MH result was for specimens not immersed in any beverage 63.9 VHN, followed by specimens immersed in the milk group 61.4 VHN, and the saliva group 59.6 VHN. For the later two groups, the pH levels of the solutions were 6.9, and 5.8 respectively. The artificial saliva group exhibited a lower MH result than the milk group. This could be related to the lower pH of this group compared to the milk group. The MH of the artificial saliva group was lower than the MH of the milk group because the pH of the artificial saliva was less than the milk. Again, findings in other studies support previous results.<sup>57, 63</sup> Previous research investigated the effect of staining solutions on nanofilled resin composites that had previously been in contact with bleaching agents. Specimens were immersed in coffee, red wine, or distilled water after exposure to different bleaching materials. Regardless of the bleaching agent to which the specimen was exposed, the results reveal that specimens that were immersed in red wine and coffee exhibited lower MH values than those immersed in distilled water.<sup>57</sup> Another report evaluated the effect of four mouth rinses on the MH of esthetic restorative materials. Ten specimens of resin composite were immersed in 20 ml of different mouth rinses, and the findings show that MH values of specimens immersed in the mouth rinse with the highest pH level were not significantly decreased.<sup>62</sup>

In this study, SEM was used as a qualitative assessment of the effect of the pH on the MH of UltraSeal XT<sup>®</sup> hydro. Images were taken with x10,000 magnification after 14 days of immersing the sealant materials in different beverages. Coca-Cola and Gatorade specimens, more than the specimen not immersed in any beverage, were eroded and pitted because of scraping the filler particles.<sup>55</sup> This might be one of the causes behind decreasing the MH. Additionally, it was reported that inorganic filler particles

compromise 53wt% of the sealant.<sup>24</sup> On the other hand, the specimen immersed in milk was less pitted than those immersed in Coca-Cola or Gatorade.

Different aging protocols have been employed in previous studies,<sup>54, 55,56,57</sup> and these protocols depend on how long the researchers immersed the samples in solutions and if the immersion time was continuous or interrupted. Some studies immersed samples for days, others for several months.<sup>57, 54</sup> In some cases, the samples were immersed in solution for the period of the study. Others researchers preferred to immerse samples several times a days but only for several minutes at a time.<sup>57, 54,55</sup> One study investigated the effect of staining solutions on nanofilled resin composite. Specimens were stored in coffee, red wine, and distilled water for one day.<sup>57</sup> In another study, researchers immersed specimens for three times a day in test solutions for 30 days.<sup>54</sup> Researchers in a further study stored the samples for seven days in an incubator.<sup>55</sup> In this study, specimens were immersed in beverages for 14 days and kept in the beverage for the period of the study, except when solutions were changed. Results of this study indicated that the period of immersion was sufficient to show a significant change in the MH of the sealant materials.

The pH of the beverage might not be the only factor that decreased the MH of the specimens. Coca-Cola contains has a phosphoric acid while orange juice and Gatorade contain citric acid. One or the other, or both, might influence the specimen's MH. A second possible factor is whether the beverage is carbonated. In this study, all beverages except Coca-Cola were uncarbonated. However, milk contains calcium, phosphate,

casein, and lipids that have anti-cariogenic properties.<sup>68</sup> These, therefore, and the aforementioned factors, need to be evaluated in future research.

One of the limitations of the present study is that the aging protocol did not mimic the oral cavity environment. Specimens were kept in the solutions for the whole period of the study. Nevertheless, in the oral cavity, teeth are not exposed to beverages for 24 hours. In addition, 62.9% of the youth population consumes at least one sugar-sweetened beverage a day and clinicians cannot predict how long it takes a child to finish a beverage.<sup>40</sup> Another limitation is that buffering agents will help to increase the pH level of the saliva after exposing the oral cavity to acidic solution. This was difficult to replicate in an in vitro study. Additionally, there are other parameters that might affect the durability of the sealant material such as wear.<sup>44</sup>

Diet analysis is an important first step to minimize the possibility of sealant failure. Dentists should discuss how many snacks the child has each day and how many ounces of juice he or she drinks in any twenty-four hours.<sup>69</sup> The American Academy of Pediatric Dentistry recommends no more than 6 ounces of soft drinks per day for children one through six years of age.<sup>70</sup> Dentists need to educate parents, guardians and caregivers about the risk sugar poses to dental sealants and that there may be other factors, such as the beverage's pH, that affect the sealant's durability. Furthermore, to investigate if sealants need to be replaced and to protect against permanent restoration, a recall visit may be necessary depending on a caries risk assessment.

In this study, UltraSeal XT<sup>®</sup> hydro was assessed to provide the clinician with an idea of the mechanical performance of resin sealants, and to observe what might happen to

patients with chronic beverage intake after dropping the pH level of saliva for a long period. In further research, the time of exposing the oral cavity to a daily beverage needs to be determined. This will help researchers to mimic the environment in the oral cavity, which will be reflected in the accuracy of the results. Also, other parameters, such as wear resistance and surface roughness, should be examined beside MH to give clinicians a better overview of the durability and resistance to wear of UltraSeal XT<sup>®</sup> hydro. On the other hand, pit and fissure sealant is one of the most effective preventive measures in modern dentistry. Dental materials exposed to different environments in the oral cavity might increase the chance of children having dental caries, so dentists should follow up continuously to help decrease the failure rate of dental sealants.<sup>22</sup>

## **CONCLUSION**

Based on the methodology employed and within the limitations of the study, it was concluded that the MH of UltraSeal XT<sup>®</sup> hydro would be affected by low pH beverages after 14 days of immersion. Coca-Cola, Gatorade, and orange juice decreased the MH values more than milk, artificial saliva, and specimens immersed in no solution.

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## **APPENDICES**

Appendix A: Tables

Appendix B: Figures

## APPENDIX A: TABLE

Table 1 pH level of the tested solutions

Beverage	pH level
Artificial saliva	5.8
Orange juice	3.9
Coca-Cola	2.4
Gatorade	2.9
Milk	6.9
Not immersed	

Table 2 Microhardness (VHN) results (n=10 per group)

Beverage	Mean	SD	Median	IQR
Artificial saliva	60.0	0.8	59.6	1.2
Orange juice	56.2	0.7	56.0	1.1
Coca-Cola	51.3	0.5	51.1	0.9
Gatorade	52.4	0.6	52.7	0.9
Milk	61.8	1.2	61.4	1.8
Not immersed	63.2	1.7	63.9	3.1

## Appendix B: Figures



*Figure 1: Isomet 1000 (Buehler Ltd).*



*Figure 2: pH meter. Orion Star A214 (Thermo Scientific, USA).*



Figure 3: An indenter. Buehler (Lake Bluff, IL, USA).



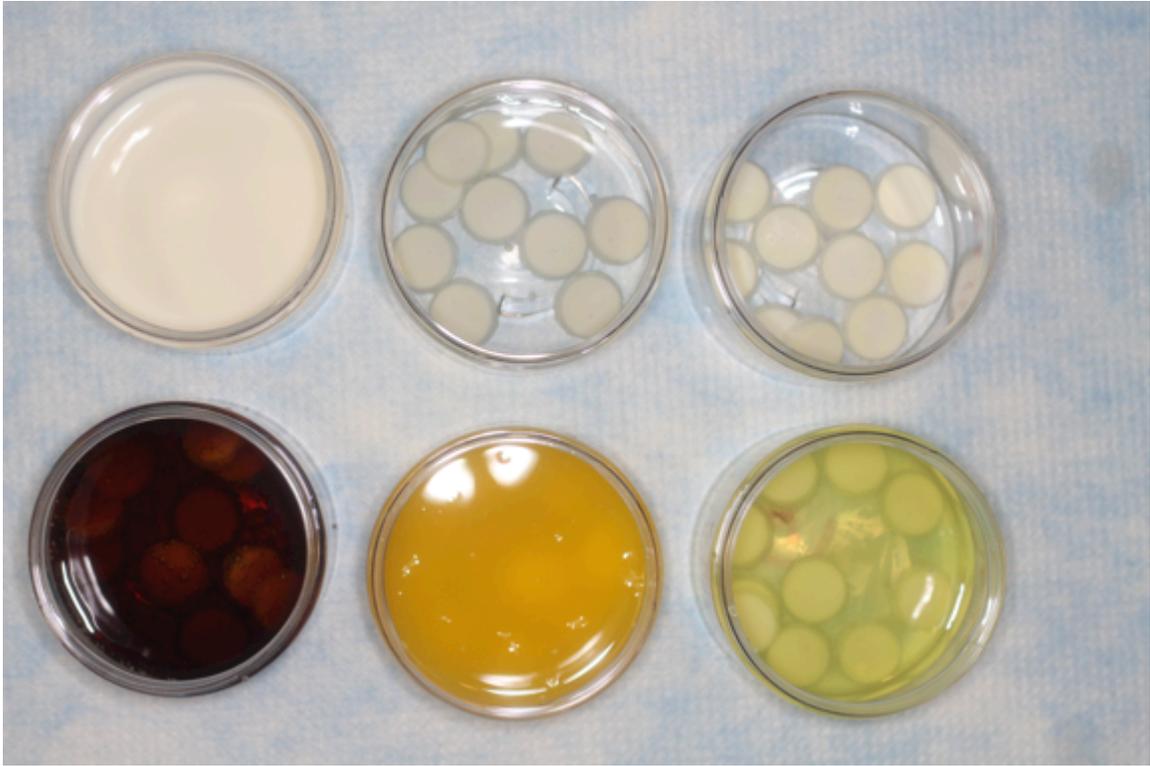
*Figure 4 Curing light. Bluephase (Ivoclar Vivadent, Schaan, Lichtenstein).*



Figure 5: UltraSeal XT<sup>®</sup> hydro.



Figure 6: A specimen of UltraSeal XT<sup>®</sup> hydro.



*Figure 7: Specimens of UltraSeal XT<sup>®</sup> hydro in six different immersed test solutions.*



Figure 8: A specimen of UltraSeal XT<sup>®</sup> hydro after sectioning.

Load 200 gf (0.2 kg f)

Diagonal ( $\mu\text{m}$ )	Vickers Hardness Number									
	0.0	0.1	0.2	0.3	0.4	0.5	0.6	0.7	0.8	0.9
59	107	106	106	105	105	105	104	104	104	103
60	103	103	102	102	102	101	101	101	100	100
61	99.7	99.3	99.0	98.7	98.4	98.0	97.7	97.4	97.1	96.8
62	96.5	96.2	95.8	95.5	95.2	94.9	94.6	94.3	94.0	93.7
63	93.4	93.1	92.8	92.5	92.2	92.0	91.7	91.4	91.1	90.8
64	90.5	90.2	90.0	89.7	89.4	89.1	88.9	88.6	88.3	88.0
65	87.8	87.5	87.2	87.0	86.7	86.4	86.2	85.9	85.6	85.4
66	85.1	84.9	84.6	84.4	84.1	83.8	83.6	83.3	83.1	82.8
67	82.6	82.4	82.1	81.9	81.6	81.4	81.1	80.9	80.7	80.4
68	80.2	80.0	79.7	79.5	79.3	79.0	78.8	78.6	78.3	78.1
69	77.9	77.7	77.4	77.2	77.0	76.8	76.5	76.3	76.1	75.9
70	75.7	75.5	75.2	75.0	74.8	74.6	74.4	74.2	74.0	73.8
71	73.6	73.4	73.1	72.9	72.7	72.5	72.3	72.1	71.9	71.7
72	71.5	71.3	71.1	70.9	70.7	70.5	70.4	70.2	70.0	69.8
73	69.6	69.4	69.2	69.0	68.8	68.6	68.5	68.3	68.1	67.9
74	67.7	67.5	67.3	67.2	67.0	66.8	66.6	66.5	66.3	66.1
75	65.9	65.7	65.6	65.4	65.2	65.0	64.9	64.7	64.5	64.4
76	64.2	64.0	63.9	63.7	63.5	63.4	63.2	63.0	62.9	62.7
77	62.5	62.4	62.2	62.1	61.9	61.7	61.6	61.4	61.3	61.1
78	60.9	60.8	60.6	60.5	60.3	60.2	60.0	59.9	59.7	59.6
79	59.4	59.3	59.1	59.0	58.8	58.7	58.5	58.4	58.2	58.1
80	57.9	57.8	57.6	57.5	57.4	57.2	57.1	56.9	56.8	56.7
81	56.5	56.4	56.2	56.1	56.0	55.8	55.7	55.6	55.4	55.3
82	55.1	55.0	54.9	54.7	54.6	54.5	54.3	54.2	54.1	54.0
83	53.8	53.7	53.6	53.4	53.3	53.2	53.1	52.9	52.8	52.7
84	52.6	52.4	52.3	52.2	52.1	51.9	51.8	51.7	51.6	51.4
85	51.3	51.2	51.1	51.0	50.8	50.7	50.6	50.5	50.4	50.3
86	50.1	50.0	49.9	49.8	49.7	49.6	49.4	49.3	49.2	49.1
87	49.0	48.9	48.8	48.7	48.5	48.4	48.3	48.2	48.1	48.0
88	47.9	47.8	47.7	47.6	47.4	47.3	47.2	47.1	47.0	46.9
89	46.8	46.7	46.6	46.5	46.4	46.3	46.2	46.1	46.0	45.9
90	45.8	45.7	45.6	45.5	45.4	45.3	45.2	45.1	45.0	44.9
91	44.8	44.7	44.6	44.5	44.4	44.3	44.2	44.1	44.0	43.9
92	43.8	43.7	43.6	43.5	43.4	43.3	43.2	43.1	43.1	43.0
93	42.9	42.8	42.7	42.6	42.5	42.4	42.3	42.2	42.1	42.1
94	42.0	41.9	41.8	41.7	41.6	41.5	41.4	41.3	41.3	41.2
95	41.1	41.0	40.9	40.8	40.7	40.7	40.6	40.5	40.4	40.3
96	40.2	40.2	40.1	40.0	39.9	39.8	39.7	39.7	39.6	39.5
97	39.4	39.3	39.2	39.2	39.1	39.0	38.9	38.8	38.8	38.7
98	38.6	38.5	38.5	38.4	38.3	38.2	38.1	38.1	38.0	37.9
99	37.8	37.8	37.7	37.6	37.5	37.5	37.4	37.3	37.2	37.2
100	37.1	37.0	36.9	36.9	36.8	36.7	36.6	36.6	36.5	36.4
101	36.3	36.3	36.2	36.1	36.1	36.0	35.9	35.9	35.8	35.7
102	35.6	35.6	35.5	35.4	35.4	35.3	35.2	35.2	35.1	35.0
103	35.0	34.9	34.8	34.7	34.7	34.6	34.5	34.5	34.4	34.3

Figure 9: Conversion table for Vickers hardness numbers.

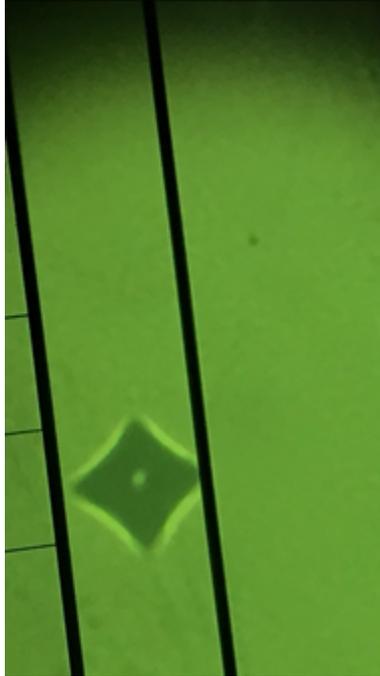


Figure 10: A representative microscopic view of Vickers microhardness test. The image was obtained after indentation of the MH test of an Ultraseal XT hydro specimen.

Not immersed

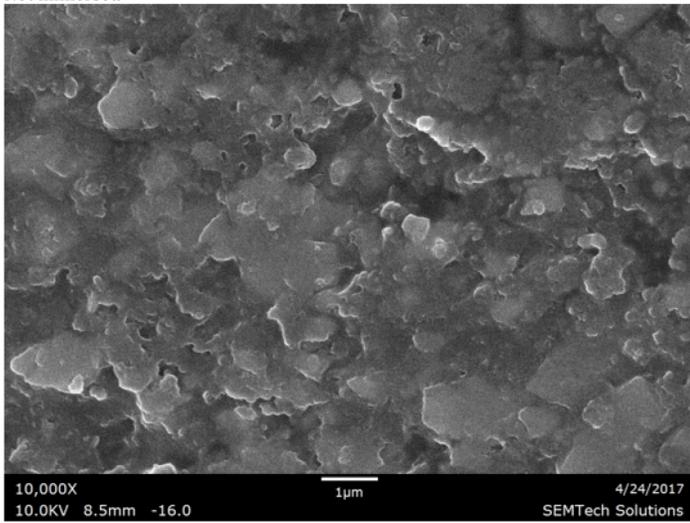


Figure 11: SEM image of a representative not immersed specimen.

Milk

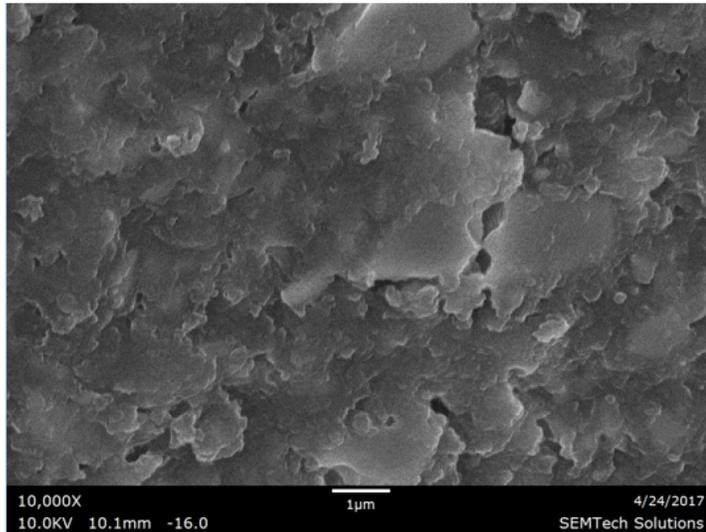


Figure 12: SEM image of a representative specimen, which has been immersed in milk.

Orange Juice

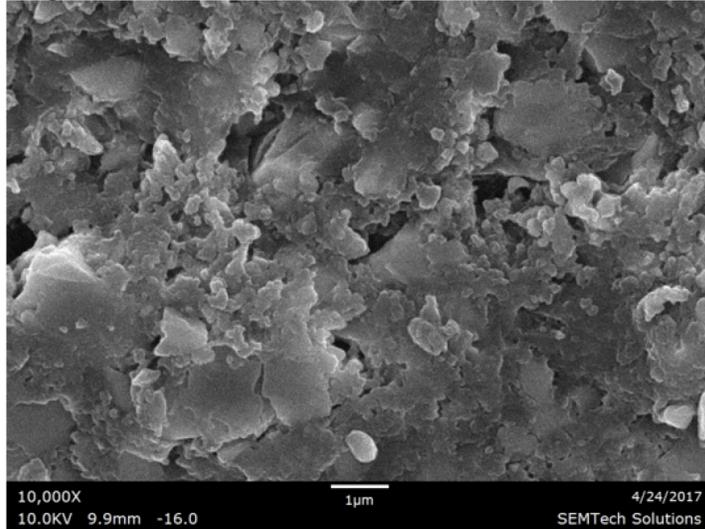


Figure 13: SEM image of a representative specimen, which has been immersed in orange juice.

Artificial saliva

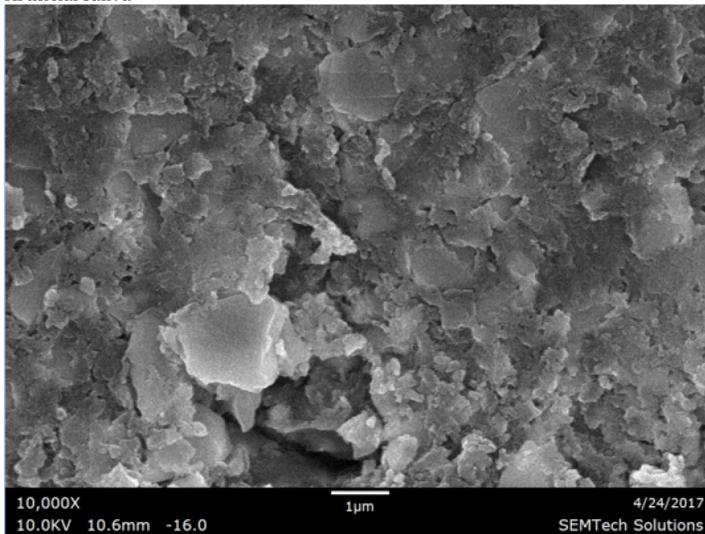


Figure 14: SEM image of a representative specimen, which has been immersed in artificial saliva.

Gatorade

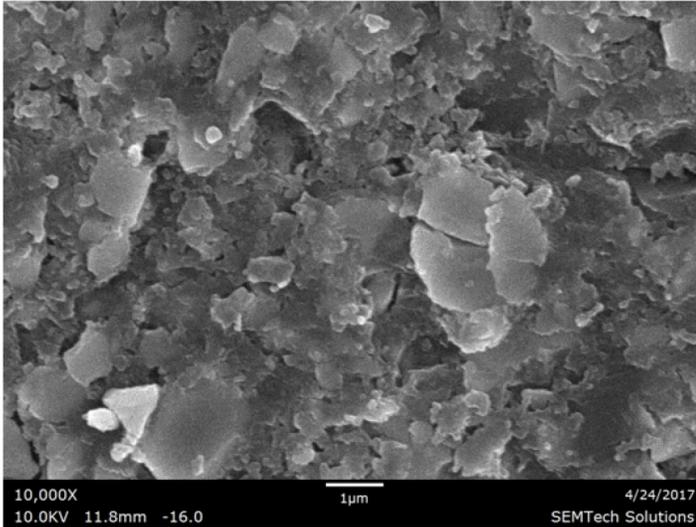


Figure 15: SEM image of a representative specimen, which has been immersed in Gatorade.

Coca-Cola

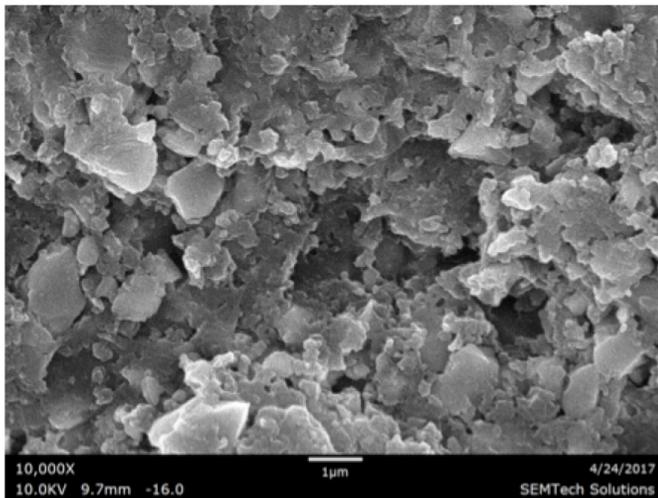


Figure 16: SEM image of a representative specimen, which has been immersed in Coca-Cola.

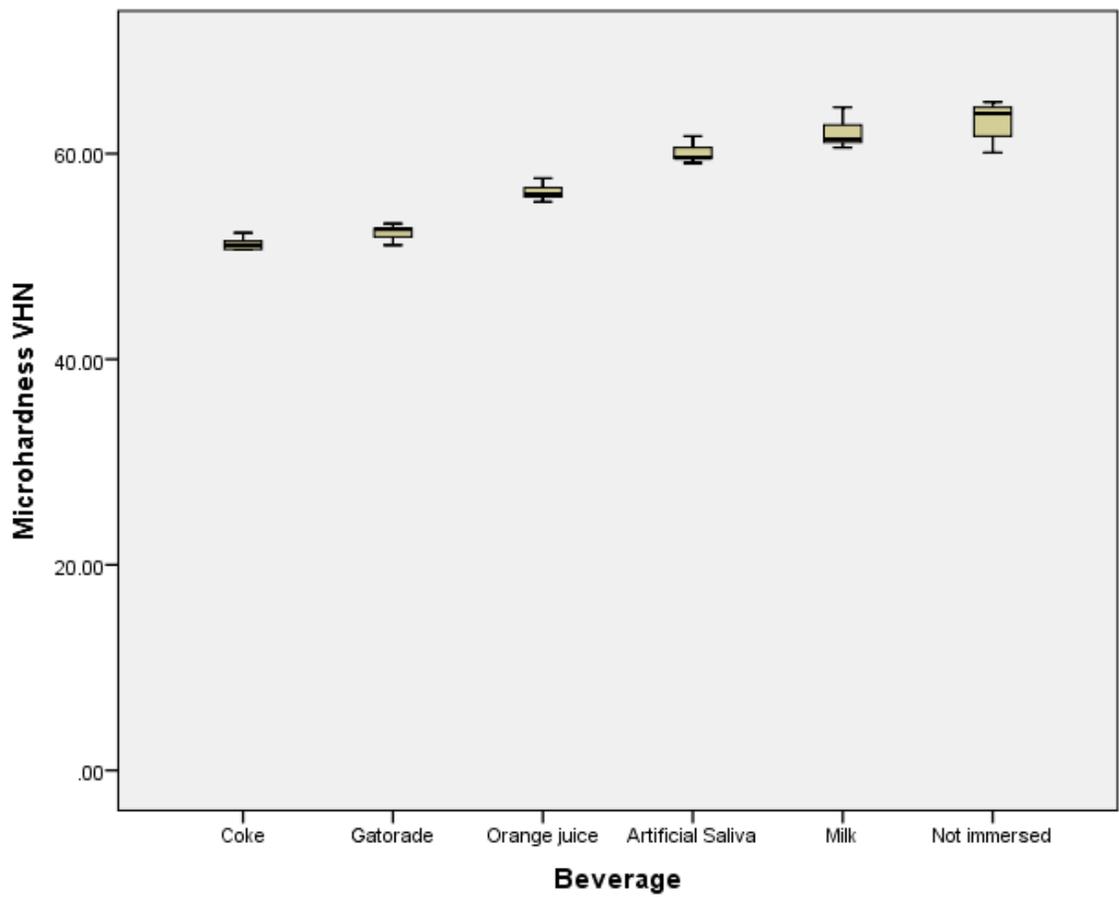


Figure 17: Microhardness boxplots.