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EFFECT OF VARIOUS CONFECTIONS ON ORAL PH LEVELS

IOANA CIOCAN
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Reviewed and approved* by the following:

James Karlinsey
Assistant Professor of Chemistry
Thesis Supervisor

Sandy Feinstein
Associate Professor of English
Honors Adviser

* Signatures are on file in the Schreyer Honors College.

ABSTRACT

The potential daily use of various oral confections may have an important impact on salivary pH. The regulation of salivary pH is imperative to maintain a healthy bacterial environment in the oral cavity. For those who may have difficulties with the secretion of saliva, artificial saliva has become commonly prescribed. The use of oral confections has also been shown to increase saliva secretion. Many confections of varying ingredients and flavors are readily available to the public. This study used a variety of confections tested in artificial saliva to monitor the effect the confections had on the pH level of the artificial saliva. The confections containing citric acid and malic acid were identified to have the strongest effect on the pH levels of the artificial saliva, making it more acidic.

TABLE OF CONTENTS

List of Figures.....	iii
List of Tables	iv
Acknowledgments.....	v
Chapter 1 Introduction.....	1
Background on Saliva	1
Background on Artificial Saliva.....	4
Background on Confections	5
Confections and Artificial Saliva in Research.....	6
Chapter 2 Methodology	7
Development of Acid and Base Standard Solutions	7
Development of Artificial Saliva	8
pH Studies for the Integration of Confections into Deionized Water	9
pH Studies for the Integration of Confections into Artificial Saliva.....	10
Chapter 3 Results	12
pH Standards	12
pH Studies for the Integration of Confections into Deionized Water	14
pH Studies for the Integration of Confections into Artificial Saliva.....	17
Chapter 4 Discussion.....	23
Conclusions	27
Bibliography	29

LIST OF FIGURES

Figure 1. Major Salivary Protein Families	3
Figure 2. Various Forms of the Samples.....	11
Figure 3. pH Solutions Tested with Thymol Blue	13
Figure 4. Vernier Probe pH Verification.....	13
Figure 5. Neutral Effects on the pH Level of Deionized Water	14
Figure 6. Slightly Acidic Effects on the pH Level of Deionized Water	15
Figure 7. Acidic Effects on the pH Level of Deionized Water	16
Figure 8. Acidic Effects on pH Levels of Deionized Water Caused by Candy Confections.....	17
Figure 9. Representative Samples Analyzed Under Normal Conditions in Various Forms	19
Figure 10. Representative Samples Analyzed at 37°C	21
Figure 11. Representative Samples Analyzed at Various Volumes	22
Figure 12. Replenished Comparison.....	22
Figure 13. Chemical Structure of Citric Acid and Malic Acid	25

LIST OF TABLES

Table 1. Preparation of pH-Standard Solutions	8
Table 2. The composition of artificial saliva according to Macknight-Hane and Whitford	9
Table 3. The list of confections tested and their respective masses	10
Table 4. Representative samples, their categories, and respective masses	18
Table 5. Confections and Their Respective Ingredients	24

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Chapter 1

Introduction

Background on Saliva

A. Chemical Composition of Saliva

Studies show that there has been an interest in the role of saliva in the health of the oral cavity for over 100 years.^{1,2,3} Extensive experiments were conducted in 1927 to investigate the chemical composition of saliva and how its components affect the role of saliva in oral diseases.⁴ Human salivary glands secrete 1000-1500 ml per day of saliva composed of approximately 99% water which contains a variety of electrolytes (sodium, potassium, calcium, chloride, magnesium, bicarbonate, phosphate) and proteins. The latter are represented by enzymes, of which amylase is the most abundant, immunoglobulins and other antimicrobial factors, mucins, traces of albumin and some polypeptides and oligopeptides.^{6,7} Nearly 26% of the salivary proteins are composed of mucins, which are glycoproteins that have a high molecular mass.⁶ Glucose and nitrogenous products such as urea and ammonia are also present.^{7,8} The interactions between all of these components are responsible for the diverse functions of saliva.

B. Functions of Saliva

The conformation of a molecule is imperative to its biological function. Saliva's multi-functionality is due to the unique amino acid composition of each enzyme that creates its distinctive shape (Figure 1).⁶ The interactions between molecules may cause

them to be involved in more than one of the functionalities of a substance. The variety of enzymes, amylases, cystatins, histatins, mucins, and peroxidases provide saliva with its anti-bacterial nature. Cystatins and mucins serve as anti-viral agents, while histatins are anti-fungal agents.^{9,7} Amylases, cystatins, mucins, proline-rich proteins, and statherins together are responsible for the tissue coating of saliva.⁶ The lubrication and viscoelasticity of saliva is due to mucins and statherins.^{6,9} Mineralization is produced from the interaction of cystatins, histatins, proline-rich proteins, and statherins. Amylases, mucins, and lipase are responsible for aiding in digestion.² Finally, carbonic anhydrases and histatins are the buffering agents in saliva.² Figure 1 provides more detail about the chemical structure of each salivary protein family and gives insight into the functional role of each family.

Saliva is considered to be amphifunctional, meaning a molecule may have protective properties, such as the ones listed above, as well as detrimental properties.² The extent of the damaging effects that a molecule can have may depend on the location of the molecule. This is clearly seen in amylases. They facilitate the clearance of viridans streptococci in solution, but when adsorbed to tooth surface, can promote adherence of bacteria and may stimulate the digestion of starch to dietary maltose, which can ultimately result in the production of acid.² Statherin and acidic proline-rich proteins are responsible for mineralization at the enamel surface through the formation of primary and secondary calcium phosphate salts.² However, when adsorbed to the enamel surface, they promote attachment of cariogenic, or cavity-causing, microorganisms.² Due to the possible formation of harmful substances, it is important that saliva be well monitored and maintained. A simple way to monitor the acidity of saliva is through pH testing.

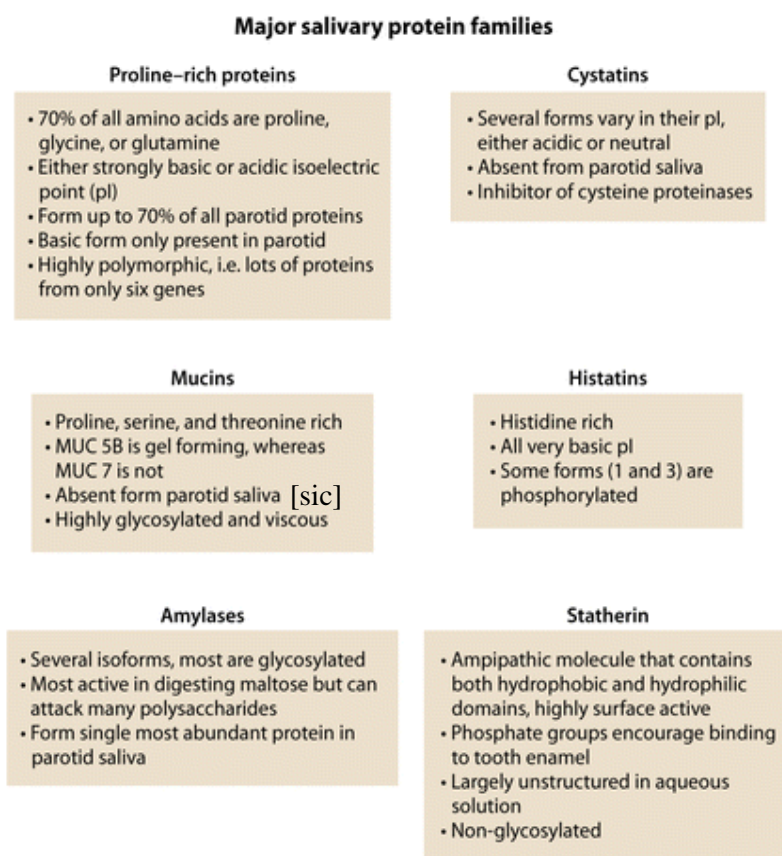


Figure 1: Major salivary protein families. Each of the salivary proteins have different functions due to the unique amino acid composition that they contain. Amylase is the only salivary protein that contains a typical amino acid composition.⁶

C. Monitoring Salivary pH

Saliva has a varying pH, but normally is found between 6 and 7.² The variability is due to the amount that is secreted at any given time.² For example, when smelling food there is an increased secretion of saliva which causes the pH to be higher. Salivary pH can be monitored in several different ways. A commonly used method is the use of a pH meter. A pH meter is an electronic device that measures the acidity of a liquid. Typically, a pH meter uses a glass electrode that is connected to an electronic meter that will display the pH reading when submersed into a solution.¹⁰ Another method is through the addition of small amounts of halochromic chemical compounds to a solution so that the acidity of

the solution can be visually determined.¹⁰ This is commonly referred to as a pH indicator. It chemically detects for hydronium ions or hydrogen ions.¹⁰ An important advantage of using a pH indicator is that it can then be readily adapted for portable analysis, as evidenced by the widespread use of litmus paper and alkacid paper.

Background on Artificial Saliva

D. Development of Artificial Saliva

Due to the ample information readily available on the molecules involved in the creation and secretion of saliva^{6,9,11}, the development of artificial saliva to aid in the treatment of dysfunctional saliva secretion or enhancement of salivary function has been on the rise.¹² Artificial salivas contain a mixture of buffering agents, cellulose derivatives, and flavoring agents.³ Unlike natural saliva, artificially created saliva typically does not contain the digestive and antibacterial enzymes and other proteins or minerals present in real saliva.³ This is why it is important to try to include as many of the enzymes, minerals, and buffering agents that are found in natural saliva when creating artificial saliva in a lab setting to be used for testing. It is imperative that the composition be as close as possible to natural saliva when testing in a lab to ensure that the results obtained would be close to the results that would be obtained if natural saliva was to be used. If the composition is not close to the natural saliva, the results obtained from the experiments could not be used to draw any conclusions regarding natural saliva.

E. Medical Use of Artificial Saliva

It is common for a person who may be having problems with dry mouth to be prescribed an artificial saliva.³ Artificial saliva can be prescribed to individuals who may be experiencing a decreased salivary flow or dysfunction.³ It can help increase the flow and ultimately cure the dry mouth condition.³ Artificial saliva can also be prescribed to individuals who have normal salivary flow, but may be prone to having problems with plaque mediated diseases, mucous membrane conditions, or occlusal dysfunction.³ Artificial saliva can be prescribed in the form of a spray, gel, or rinse.³ Here, unique artificial saliva may be created for an individual that will contain more of the protein or enzyme to specifically aid in the treatment of the condition. For example, individuals with occlusal dysfunction may be prescribed an artificial saliva that contains more statherin to aid with lubrication.³

Background on Confections

F. Confections Currently Available on the Market with Oral Implications

There are many different brands and flavors of oral confections that are readily available to the public that offer promises of teeth cleaning and whitening, fresher breath, and long lasting flavor. Wrigley for example, which sells its products in more than 180 countries¹³, offers the popular Altoids, Life Savers, Orbit White, Extra, Eclipse, and 5® gum and mint brands and claims that chewing their sugar-free gum or consuming their mints increases the production of saliva, which can help neutralize plaque acid.¹³

Hershey offers more than their famous chocolate candies, they also offer a wide variety of mints including the popular Ice Breakers brand that promises to refresh the mouth. Breath Savers, also a Hershey product, markets their mints with the ability to neutralize bad breath.¹⁴ In this research, the word confection will refer to a confection that promises to freshen breath, while candy confection will refer to a candy that does not promote fresh breath.

Confections and Artificial Saliva in Research

The oral cavity contains one of the most complex bacterial floras found in the human body.¹⁵ It has been shown that changes in the flora can be related to a change in the pH level of the oral cavity that can lead to specific diseases.¹⁵ Normally, the pH of the oral cavity is between 6 and 7.⁸ If the pH level rises above or falls below those numbers, the prevalence of the flora can be altered.⁸ A high pH may kill some of the necessary bacteria that help protect against the development of caries, while a low pH may allow for the accumulation of bacteria that can be harmful to tooth enamel, gums, and the oral cavity as a whole. There has been a global increase in the prevalence of caries that has been partially linked to an individual's diet.¹⁶ Oral confections, which seem to have become integrated into the human diet as an in-between-meals activity for the mouth, have been marketed as promoting a healthy oral environment, but under several different circumstances their implications for oral pH have yet to be fully determined. Artificial saliva may be used in the lab to help identify these effects due to its ability to be easily manipulated into circumstances that the oral cavity may often encounter. It is important for the population to be cognizant of the impact that oral confections may have on their

oral health. This research project is committed to understanding the effects that various oral confections may have on oral pH under a variety of circumstances that may be encountered daily.

Chapter 2

Methodology

Development of Acid and Base Standard Solutions

Component solutions used in the preparation of pH-standard solutions were prepared in one liter quantities as follows: 14.9 g of KCl was dissolved in 500 mL of distilled water and then diluted to 1.0 liter to create 0.20M potassium chloride, KCl.

Additional solutions were created using similar techniques:

- 0.20M hydrochloric acid, HCl – 16.7 mL of concentrated (12M) HCl in 1.0 liter of water.
- 0.10M potassium hydrogen phthalate, $\text{KHC}_8\text{H}_4\text{O}_4$ – 20.4 g of $\text{KHC}_8\text{H}_4\text{O}_4$ in 1.0 liter of water.
- 0.20M sodium hydroxide, NaOH – 8.0 g of NaOH in 1.0 liter of water.
- 0.10M potassium dihydrogen phosphate, KH_2PO_4 – 13.6 g of KH_2PO_4 in 1.0 liter of water.
- 0.025M borax, $\text{Na}_2\text{B}_4\text{O}_7 \cdot 10\text{H}_2\text{O}$ – 9.5 g of $\text{Na}_2\text{B}_4\text{O}_7 \cdot 10\text{H}_2\text{O}$ in 1.0 liter of water.
- 0.050M sodium bicarbonate, NaHCO_3 – 4.2 g of NaHCO_3 in 1.0 liter of water.

Then, 500 mL of stock solution of each pH-standard was prepared to test the Vernier pH probe for validity before submersion into deionized water and artificial saliva solutions as

described in Table 1. To verify the pH of the standardized solutions, the indicator Thymol Blue was added to each standard.

<i>pH</i>	<i>Components</i>
1	125 mL of 0.20M KCl and 335 mL of 0.20M HCl
2	125 mL of 0.20M KCl and 33 mL of 0.20M HCl
3	250 mL of 0.10M KHC ₈ H ₄ O ₄ and 56 mL of 0.20M HCl
4	250 mL of 0.10M KHC ₈ H ₄ O ₄ and 0.25 mL of 0.20M HCl
5	250 mL of 0.10M KHC ₈ H ₄ O ₄ and 56 mL of 0.20M NaOH
6	250 mL of 0.10M KH ₂ PO ₄ and 14 mL of 0.20M NaOH
7	250 mL of 0.10M KH ₂ PO ₄ and 73 mL of 0.20M NaOH
8	250 mL of 0.10M KH ₂ PO ₄ and 115 mL of 0.20M NaOH
9	250 mL of 0.10M KH ₂ PO ₄ and 12 mL of 0.20M HCl
10	250 mL of 0.025M Na ₂ B ₄ O ₇ and 27 mL of 0.20M NaOH
11	250 mL of 0.050M NaHCO ₃ and 57 mL of 0.20M NaOH
12	125 mL of 0.20M KCl and 30 mL of 0.20M NaOH
13	125 mL of 0.20M KCl and 330 mL of 0.20M NaOH

Table 1: Preparation of pH-Standard Solutions¹⁷

Development of Artificial Saliva

The artificial saliva was prepared using the formula and instructions offered by Karlinsey, Mackey, and Schwandt.¹³ The chemicals used were obtained from Fisher Scientific and are listed in Table 2. In 2 liters of deionized water, 0.709 grams of calcium nitrate was added. After complete dissolution, 0.247 grams of potassium phosphate monobasic was added. After complete dissolution, 19.4 grams potassium chloride was added. After complete dissolution, 8.56 grams of cacodylic was added slowly and then allowed to mix for a minimum of 15 minutes. Finally, sodium hydroxide was added to adjust the pH to 7.0.

Materials	Quantity
Deionized Water	2.00 Liters
Calcium Nitrate	0.709 grams
Potassium Phosphate Monobasic	0.247 grams
Potassium Chloride	19.4 grams
Cacodylic Acid	8.56 grams
Sodium Hydroxide	As needed to adjust pH

Table 2: The composition of artificial saliva according to Karlinsey, Mackey, and Schwandt (2012) formula.¹⁸

pH Studies for the Integration of Confections into Deionized Water

All confections, listed below in Table 3, were first tested for pH effects in deionized water under a chosen set of normal conditions. The normal conditions consisted of the sample being placed in 15 mL of deionized water after 20 seconds of pH data collection by Vernier Logger Pro Software. The sample was analyzed at 20° C, constantly stirred, and analyzed until it was completely dissolved.

Brand	Flavor	Average Mass (grams)
Altoids	Cinnamon	0.744 ± 0.07
	Wintergreen	0.734 ± 0.06
Ice Breakers Sours	Strawberry Mix	0.804 ± 0.03
	Green Apple Mix	0.785 ± 0.004
Ice Breakers	Wintergreen	0.801 ± 0.01
	Cinnamon	0.794 ± 0.003
Mentos	Peppermint	0.736 ± 0.02
	Wintergreen	0.745 ± 0.01
Tic Tac	Orange	0.496 ± 0.01
	Wintergreen	0.484 ± 0.008
	Strawberry Fields	0.493 ± 0.01
Life Savers	Cherry	3.758 ± 0.09
	Wint-O-Green	3.656 ± 0.1
Sugar Free Life Savers	Cherry	3.711 ± 0.03
Warheads	Blue Raspberry	12.623 ± 0.2
Breath Savers	Wintergreen	1.799 ± 0.05
	Peppermint	1.823 ± 0.02

Table 3: The list of confections tested and their respective masses.

pH Studies for the Integration of Confections into Artificial Saliva

Four samples, Ice Breakers Strawberry Mix, Altoids Wintergreen, Breath Savers Wintergreen, and Warheads, were chosen as representative samples of an acidic effect on pH, slightly acidic effect, no effect, and an acidic effect caused by a candy confection respectively to be tested in artificial saliva. These samples were then tested under various conditions explained below using the Vernier Logger Pro Software to monitor pH.

- Normal Conditions
 - Each sample was tested four times under the previously set normal conditions from the deionized water experiment. One sample was kept whole, one was chopped, one was crushed into a fine powder, and the last sample used was crushed and the mass used was doubled to mimic the effect of consuming two confections at once.
- Temperature
 - Each sample was tested three times while incubated at 37 degrees Celsius to mimic body temperature. One sample was kept whole, one was chopped, and one was crushed into a fine powder as shown in Figure 2. The pH level was tested every 30 seconds.



Figure 2: Various forms of the Samples. Example of a sample kept whole, chopped, and crushed.

- Volume
 - Each sample was tested three times using the set normal conditions. The sample was kept whole and tested in 5 mL, 10 mL, and 15 mL of artificial saliva.

- Replenishment
 - o Each sample was tested once. The sample was kept whole and placed in 15 mL of artificial saliva. Every 10 seconds, approximately 1 mL of the solution was removed and replaced with approximately 1 mL of artificial saliva to mimic swallowing.

Chapter 3

Results

pH Standards

The addition of Thymol Blue to each solution, shown in Figure 3, indicated that the pH standards measured correct values on the pH scale. The top row in Figure 3 shows pH standards 1 to 7 from left to right, and the bottom row shows pH standards 8 to 13 from left to right. The pH of 1 correctly showed a pink color, pH of 2 showed an orange color, pH values 3-8 showed a yellow color, pH of 9 showed a teal blue color, and lastly, the basic pH values, 10-13, showed a dark blue color. Figure 4 shows a comparison of the pH standard values to the observed pH values while using a Vernier pH probe. The figure shows that the Vernier pH probe accurately measured the pH levels of the pH standard solutions.

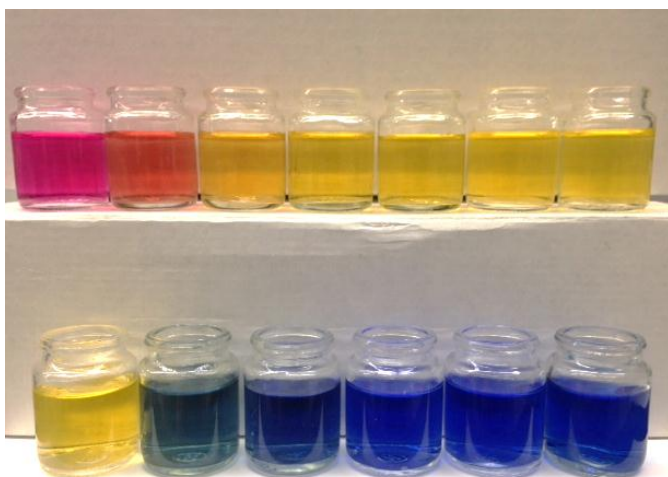


Figure 3: pH Solutions Tested with Thymol Blue. The top row shows the pH values 1 to 7 from left to right, and the bottom row shows the pH values 8-13 from left to right.

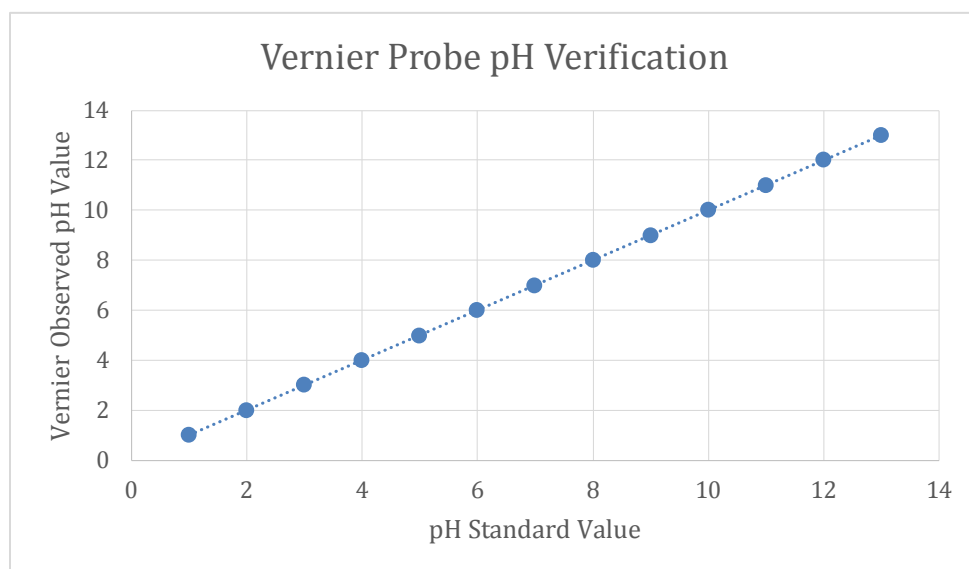


Figure 4: Vernier Probe pH Verification. The Vernier pH probe was able to accurately measure the pH values of the standardized solutions.

pH Studies for the Integration of Confections into Deionized Water

The results of the analysis of the samples listed in Table 3 under normal conditions seemed to separate into four different categories: neutral effects, slightly acidic effects, acidic effects, and acidic effects on the pH level caused by candy confections. The results of the confections that caused a neutral effect on the pH level of the deionized water are shown in Figure 5. Breath Savers Wintergreen, Ice Breakers Cinnamon, Tic Tac Wintergreen, and Ice Breakers Wintergreen all seemed to stabilize within roughly one pH unit of each other. Breath Savers Wintergreen stabilized at a pH of 7.31, Ice Breakers Cinnamon stabilized at a pH of 6.90, Tic Tac Wintergreen stabilized at a pH of 6.21, and Ice Breakers Wintergreen stabilized at a pH of 6.80.

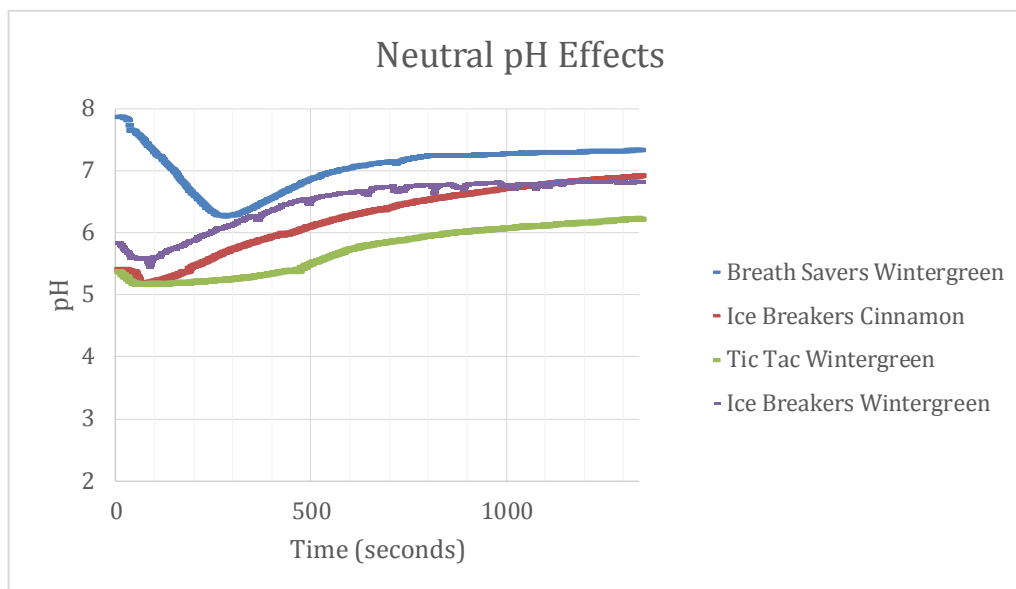


Figure 5: Neutral Effects on the pH Level of Deionized Water. Breath Savers Wintergreen, Ice Breakers Cinnamon, Tic tac Wintergreen, and Ice Breakers Wintergreen all had stabilized pH levels within one pH unit of each other in a fairly neutral range (between pH levels of 6.2 and 7.4).

The confections whose results supported a slightly acidic effect on the pH level of the deionized water were Altoids Wintergreen, Altoids Cinnamon, Life Savers Wint-O-Green. The Altoids Wintergreen stabilized at a pH of 5.94, the Altoids Cinnamon stabilized at a pH of 5.97, and the Life Savers Wint-O-Green stabilized at a pH of 5.81. The results are shown in Figure 6.

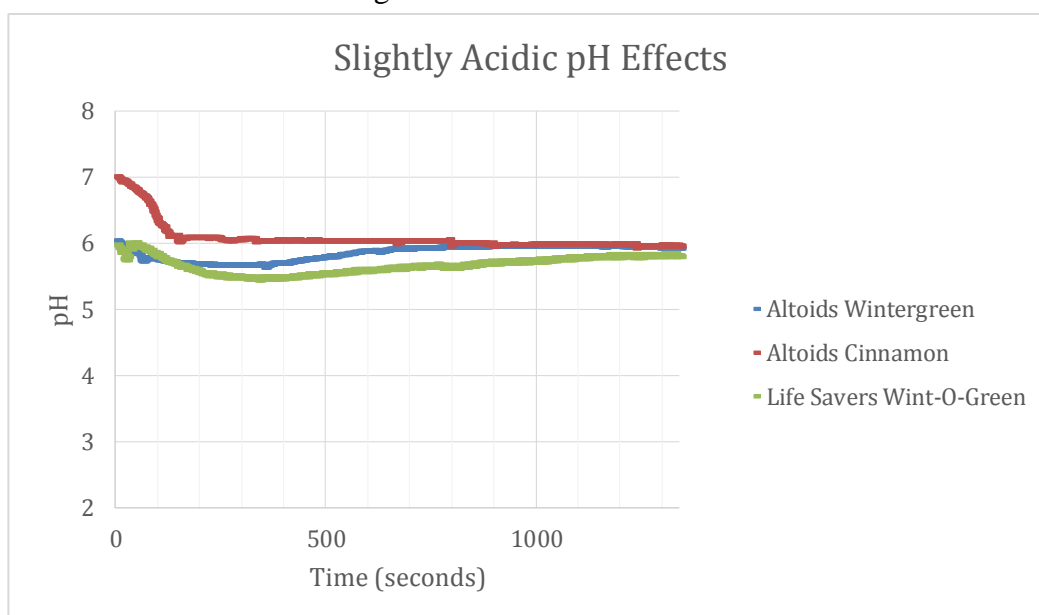


Figure 6: Slightly Acidic Effects on the pH Level of Deionized Water. Altoids Wintergreen, Altoids Cinnamon, and Life Savers Wint-O-Green all had pH levels that stabilized slightly below a pH of 6.

Many of the confections had an acidic effect on the pH level of the deionized water. Ice Breakers Sours Strawberry Mix, Ice Breakers Cool Strawberry, Ice Breakers Sours Green Apple Mix, Tic Tac Strawberry Fields, Tic Tac Orange, Mentos Wintergreen, and Mentos Peppermint all had pH levels that stabilized at an acidic level. The results are shown in Figure 7. Ice Breakers Sours Strawberry Mix stabilized at a pH of 2.74, Ice Breakers Cool Strawberry stabilized at a pH of 3.31, Ice Breakers Sours Green Apple Mix stabilized at a pH of 2.73, Tic Tac Strawberry Fields stabilized at a pH

of 3.23, Tic Tac Orange stabilized at a pH of 3.49, Mentos Wintergreen stabilized at a pH of 3.76, and Mentos Peppermint stabilized at a pH of 3.73.

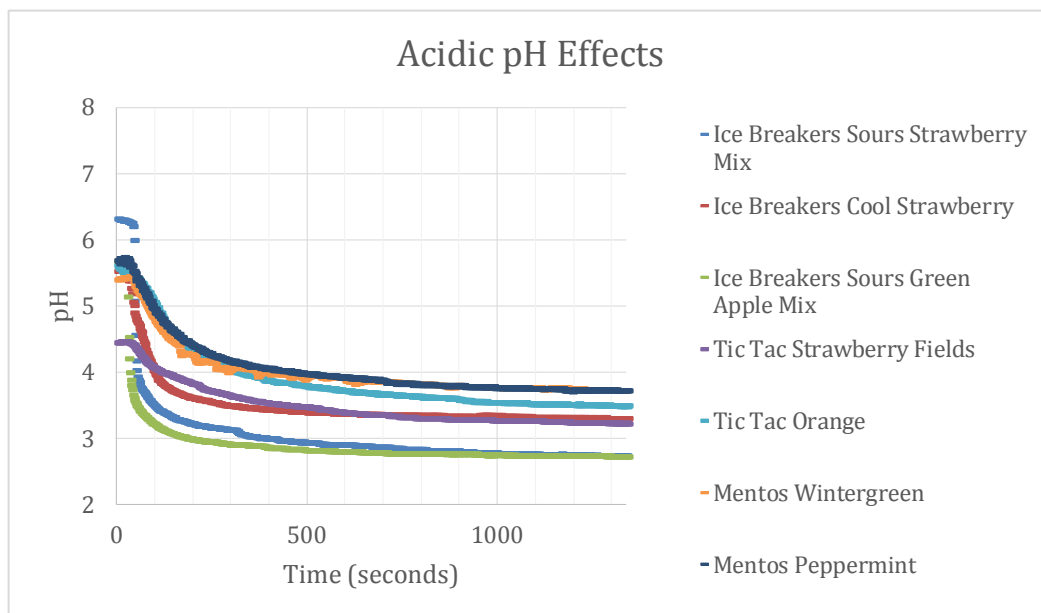


Figure 7: Acidic Effects on the pH Level of Deionized Water. Ice Breakers Sours Strawberry Mix, Ice Breakers Cool Strawberry, Ice Breakers Sours Green Apple Mix, Tic Tac Strawberry Fields, Tic Tac Orange, Mentos Wintergreen, and Mentos Peppermint all stabilized at an acidic pH level between 2.73 and 3.76.

Lastly, Warheads Blue Raspberry, Life Savers Cherry, and Sugar Free Life Savers Cherry were all confections that were not marketed as freshening breath, but just as candy and all had an acidic effect on the pH level of the deionized water. The results are shown in Figure 8. Warheads Blue Raspberry stabilized at a pH of 2.51, Life Savers Cherry stabilized at a pH of 2.84, and Sugar Free Life Savers Cherry Stabilized at a pH of 2.47.

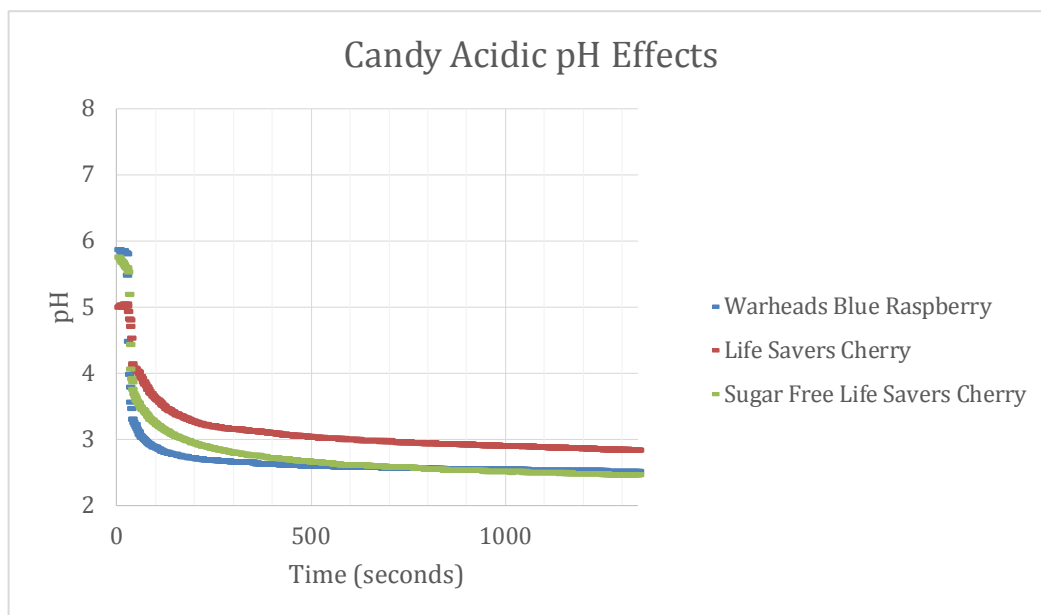


Figure 8: Acidic Effects on pH Levels of Deionized Water Caused by Candy Confections. Warheads Blue Raspberry, Life Savers Cherry, and Sugar Free Life Savers Cherry all stabilized at an acidic pH level between 2.47 and 2.84.

pH Studies for the Integration of Confections into Artificial Saliva

The results from the pH studies for the integration of confections into deionized water were used to decide on four samples that were representative of the categories that presented themselves from those experiments. The four representative samples, their respective categories, and masses are listed in Table 4. The samples, whole, chopped, or crushed to mimic the various forms the confection may take when being eaten, were then analyzed using the Vernier Logger Pro Software to test the pH level at 37 degrees Celsius to mimic body temperature. Next, the samples--whole, chopped, crushed, and crushed again with double the original mass to mimic the consumption of two confections--were analyzed under the normal conditions used in the deionized water experiments. Then, the

samples, kept whole, were analyzed in 5 mL, 10 mL, and 15 mL of artificial saliva. Lastly, the samples, kept whole, were analyzed in 15 mL of artificial saliva with approximately 1 mL of the solution taken out every 10 seconds and replaced with fresh artificial saliva.

Sample	Category	Mass (grams)
Altoids Wintergreen	Slightly Acidic	0.734
Ice Breakers Sours Strawberry Mix	Acidic	0.804
Breath Savers Wintergreen	Neutral	1.799
Warheads Blue Raspberry	Candy Acidic	12.623

Table 4: Representative samples, their categories, and respective masses.

In the normal condition experiments, a clear difference between the deionized water experiments and the artificial saliva experiments could be seen. The results are shown below in Figure 9. Altoids Wintergreen stabilized at a pH of 7.02. This pH is comparatively neutral to the slightly acidic pH of 5.94 that it stabilized to under the same conditions, but in deionized water. The various forms had little to no variation. Breath Savers Wintergreen also stabilized at a neutral pH of 7.02. This was slightly lower than the pH of 7.31 which it stabilized at under the same conditions, but in deionized water. Again, the various forms did not have an effect on the pH levels. Warheads Blue Raspberry previously stabilized at a pH of 2.51 in deionized water, but here it stabilized at a pH of 3.11. The various forms did not have an effect on the pH levels. Lastly, the Ice Breakers Strawberry Sours Mix had different stabilization pH levels for its various forms. The whole sample took longer to effect the pH and stabilized at 4.69, the chopped

stabilized at 4.27, the crushed stabilized at 4.26, and the crushed double volume stabilized at a pH of 3.44. The observed decrease in the latter value is again as expected, due to the increased concentration of components in solution.

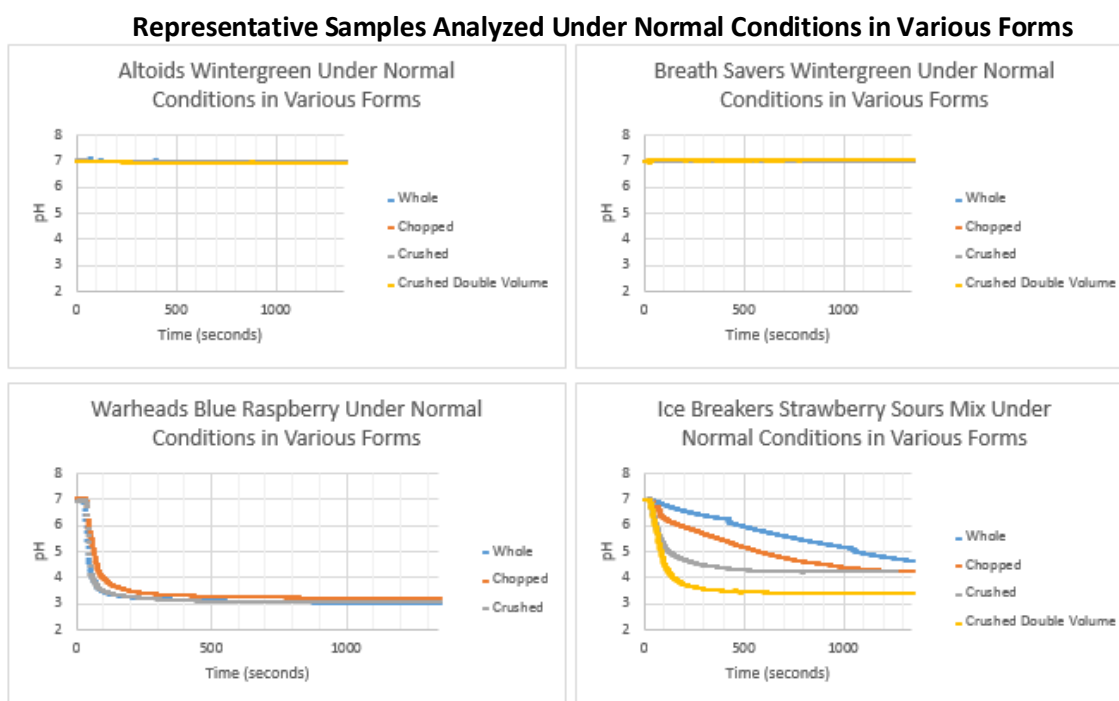


Figure 9: Representative Samples Analyzed Under Normal Conditions in Various Forms. Altoids Wintergreen, Breath Savers Wintergreen, and Warheads Blue Raspberry did not have a distinction among the various forms. A significant difference among the forms of Ice Breakers Sours Strawberry Mix was observed. The Ice Breakers Sours Strawberry Mix whole sample took longer to effect the pH and stabilized at 4.69, the chopped stabilized at 4.27, the crushed stabilized at 4.26, and the crushed double volume stabilized at a pH of 3.44. It is worth noting that both of the crushed samples followed the same trend.

In the incubation experiments, the results are shown in Figure 10. Altoids Wintergreen had an increase in its pH level from the deionized water experiment. It went

from stabilizing at a pH of 5.94 to stabilizing to a pH of 7.01. The various forms that the sample had did not affect the pH level of the sample enough to signify a difference among the forms. The Breath Savers Wintergreen pH was brought down to a more neutral level from its pH in the deionized water experiments that had stabilized at a 7.31, here stabilizing at a 7.02. The various forms showed little variation. The Warheads Blue Raspberry stabilized at a slightly higher pH, 3.05, compared to the deionized water experiments where it stabilized at a pH of 2.47. The whole form of the candy had a slower decrease in pH compared to the chopped and crushed, which was to be expected due to the significant reduction in surface area. The Ice Breakers Sours Strawberry Mix previously stabilized at a pH of 2.74 in the deionized water experiment. Here it stabilized at a higher pH of 3.84. There was a clear distinction among the various forms and the length of time that it took for the pH to drop. The crushed consistently had the lowest pH level.

In the experiments analyzing the pH levels where the sample was tested under various volumes, a clear distinction was observed between the amount of artificial saliva and pH level for Ice Breakers Sours Strawberry Mix. The results are shown in Figure 11. Altoids Wintergreen and Breath Savers Wintergreen stabilized at a neutral pH of 7.02 and 7.03 with very little to no variation between the different volumes of artificial saliva. The Ice Breakers Sours Strawberry Mix had a clear distinction between volume of artificial saliva and pH level. With the artificial saliva at a volume of 5 mL the pH was significantly lower, 3.59, compared to 10 mL and 15 mL where the pH stabilized at 4.53 and 4.68 respectively.

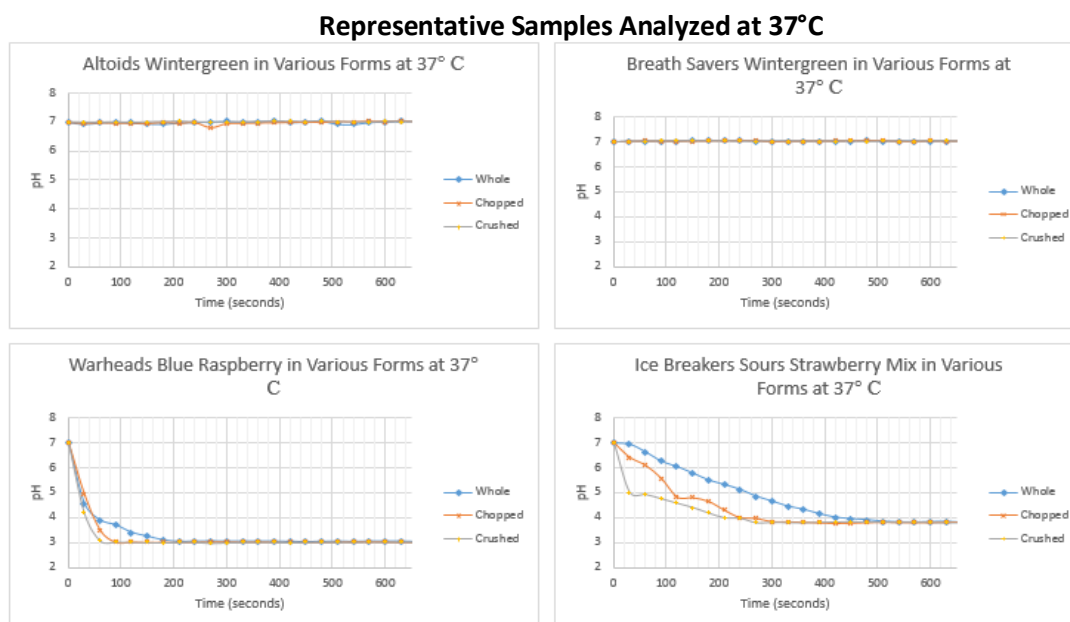


Figure 10: Representative Samples Analyzed at 37°C. Altoids Wintergreen and Breath Savers Wintergreen did not have a distinction between the various forms. It is worth noting that the whole sample of Warheads Blue Raspberry took longer to reach a lower pH than the chopped and crushed. There was also a clear difference in the time it took for the Ice Breakers Sours Strawberry Mix samples to reach a lower pH level among the various forms.

Lastly, during the replenishment experiments a clear distinction between the pH levels of the samples could be made. The results are shown in Figure 12. Altoids Wintergreen and Breath Savers Wintergreen had pH levels that stayed consistent and both stabilized at 7.04. The Ice Breakers Sours Strawberry Mix had a slowly decreasing pH until it stabilized at 6.59. The Warheads Blue Raspberry had the largest drop in pH compared to the other samples, and also stabilized at the lowest pH, 5.64.

Representative Samples Analyzed at Various Volumes

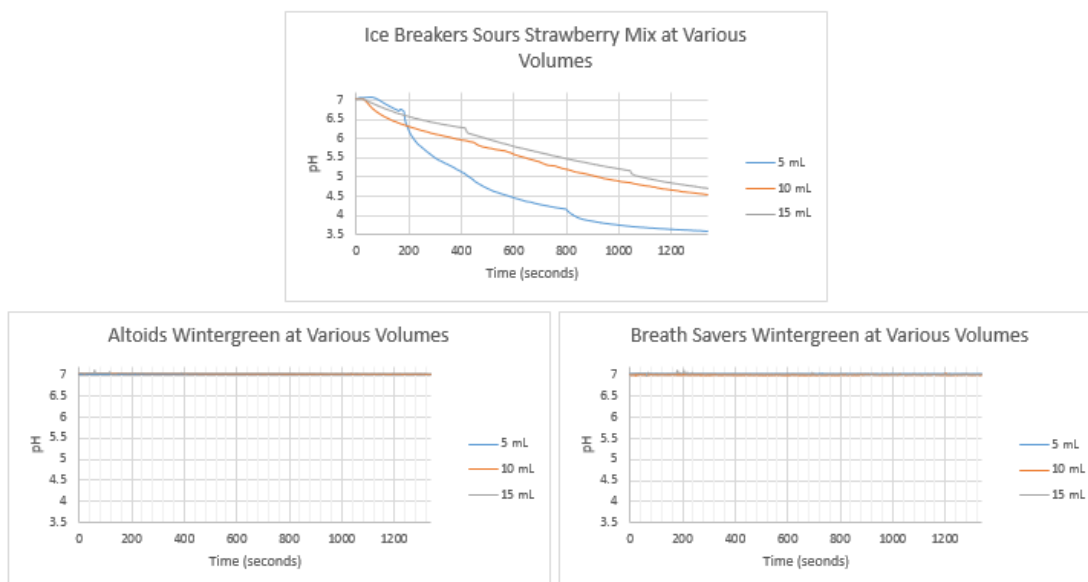


Figure 11: Representative Samples Analyzed at Various Volumes. The representative samples were tested in 5 mL, 10 mL, and 15 mL.

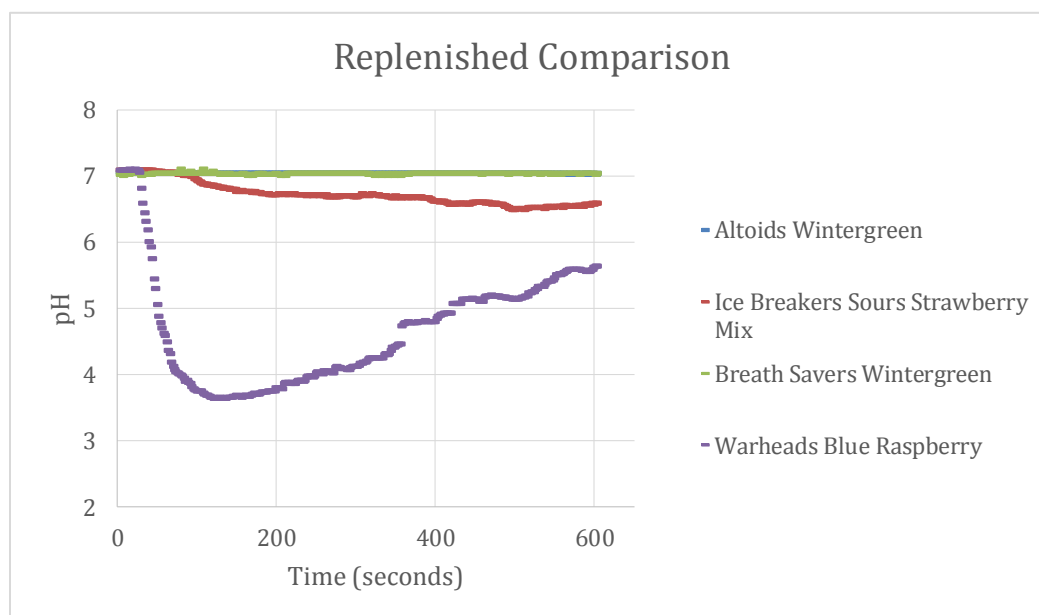


Figure 12: Replenished Comparison. Representative samples monitored while replenishing the artificial saliva solution.

Chapter 4

Discussion

The continuous consumption of various oral confections may have an effect on the pH level of the mouth which could lead to problems with an increase or decrease of bacteria in the oral cavity or erosion of the tooth enamel. Various confections that are readily available at grocery stores were tested in deionized water and artificial saliva to develop an understanding of the relationship between the consumption of the confections and the effects on the pH level directly caused by the confections. The various experiments performed in the deionized water gave a basic understanding of the type of effect that various confections may have on pH levels. From those experiments, four categories of effects were established: neutral, slightly acidic, acidic, and acidic caused by a candy confection. From these four categories, four representative samples were chosen, one from each category, to analyze in artificial saliva to gain an understanding of the potential effects these confections may have on the pH level in the oral cavity. Out of all of the experiments conducted in the artificial saliva, the Ice Breakers Sours Strawberry Mix and Warheads Blue Raspberry had the most consistent and drastic acidic effects on the pH level of the artificial saliva. This may lead to the assumption that the ingredients that may be common to both the Ice Breakers and Warheads confection, but not to the Altoids and Breath Savers confections, may be the cause of the lowering of the pH. Below, Table 5 shows the ingredients of the representative samples tested in artificial saliva. Ice Breakers Sours Strawberry Mix and Warheads Blue Raspberry both contain citric acid and malic acid, which are not present in the other confections.

Confection	Ingredients
Ice Breakers Sours Strawberry Mix	Sorbitol; Malic Acid; Maltodextrin; Contains 2% or Less of Maltitol; Tartaric Acid; Natural and Artificial Flavor; Aspartame; Hydrogenated Palm Oil; Gum Acacia; Magnesium Stearate; Citric Acid; Man Nitol; Soy Lecithin; Artificial Color (Red 40; Yellow 6; Blue 1) Phenylketonurics: Contains Phenylalanine.
Warheads Blue Raspberry	Corn Syrup, Sugar, Microencapsulated Malic Acid (Malic Acid, Hydrogenated Palm Oil), Citric Acid, Gum Acacia, Deproteinized Soybean Oil (Processing Aid), Ascorbic Acid, Artificial Flavors, Carnuba Wax, Corn Starch, Blue 1, Red 40, Yellow 5.
Altoids Wintergreen	Sugar, Gum Arabic, Artificial Flavors, Gelatin, Color (Red 40).
Breath Savers Wintergreen	Sorbitol; Magnesium Stearate, Artificial Flavor, Aspartame, Medium Chain Triglycerides, Zinc Gluconate; Artificial Color (Yellow 5 Lake; Blue 1 Lake); Polysorbate 80.

Table 5: Confections and Their Respective Ingredients.^{13,19}

Citric acid, whose structure is shown in Figure 13, is a weak organic acid that is typically used as a preservative in food products. Malic acid, whose structure is also shown in Figure 13, is a dicarboxylic acid that is naturally made by all living organisms. More importantly, directly relating to the Ice Breakers Sours Strawberry Mix and Warheads Blue Raspberry used in this research project, both citric acid and malic acid can be used to produce a sour or acidic taste in foods. They are common ingredients that can be found in various confections and drinks. The typical pH level of citric acid is 3.2

and malic acid is 3.4. This explains why the pH level of the artificial saliva drastically decreased when a confection with citric acid and malic acid as ingredients was analyzed. Partly due to this drop in pH, citric acid has been linked to the erosion of tooth enamel.²³ The replenishment experiment (Figure 12) demonstrated that frequent swallowing while consuming a confection can drastically lower the effect that it may have on the pH level of the oral cavity, which in turn could protect the tooth enamel from erosion. The Ice Breakers Sours Strawberry Mix stabilized within a pH range of 3.59 and 4.68 under normal conditions in artificial saliva, but when swallowing was mimicked, the pH stabilized at a relatively neutral pH of 6.59. The same effect could be seen with the Warheads Blue Raspberry, which under normal conditions stabilized at a pH of 3.11, but during replenishment, the pH stabilized to a much higher 5.64.

Chemical Structures of Citric Acid and Malic Acid

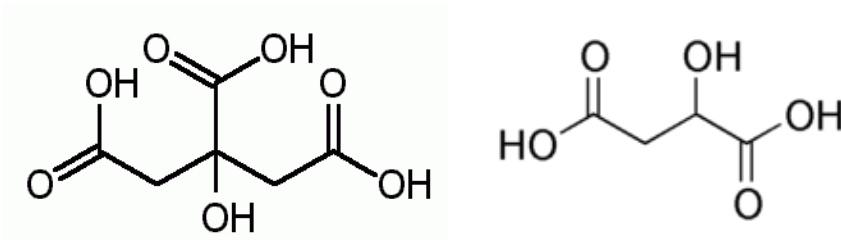


Figure 13: Chemical Structure of Citric Acid and Malic Acid. Citric Acid (left) is a weak organic acid that is commonly used to preserve food and add a sour taste to them. Malic Acid (right) is a dicarboxylic acid that is produced by all living organisms and is responsible for the sour taste in some foods.

Frequent swallowing is one way that can help decrease the effects of these acids on the pH level of the mouth. Another way could be the amount of saliva that is present in the mouth during the consumption of the confection. The various volumes experiments

conducted in the artificial saliva (Figure 11) showed a clear connection between the amount of artificial saliva and the effect of the pH. The Ice Breakers Sours Strawberry Mix at 5 mL had a pH level of 3.59, while at 10 mL the pH was 4.53, and at 15 the pH stabilized at 4.68. These results may indicate that people who have a low salivary flow and consume confections containing citric acid and malic acid can expect a more drastic effect on their pH levels. To help counter the effects, these people could take supplements such as Xylitol Melts that may help increase their salivary flow. Along with the increase in salivary flow, continuous swallowing could help ensure that these acids are not lingering in high concentrations in the oral cavity.

Lastly, to help protect against these effects on the pH level, the form and amount of consumption of the various confections should be taken into account. The artificial saliva studies showed that when a confection was consumed whole without it being chopped or crushed, the pH level was higher and did not have as fast of an effect on the artificial saliva. For example, when the Ice Breakers Sours Strawberry Mix was analyzed under normal conditions (Figure 9) while being kept whole, chopped, and crushed, it was clear that the pH stabilized the quickest and at the lowest level, a pH of 3.44, when it was crushed at twice the volume. The whole sample took 2.2 times longer to stabilize at a low pH of 4.69, the chopped took 2.1 times longer to stabilize to a pH of 4.27, and the crushed took 1.4 times longer to stabilize to a pH of 4.26 than the crushed double volume. These results indicate that the more confections consumed with little to no time in between consumption and whether it is chewed while being consumed, amplifies the acidic effect on the pH level of the artificial saliva. To help decrease these drastic effects,

confections should not be chewed and a sufficient amount of time should pass before another confection is consumed.

Conclusions

The pH level of artificial saliva becomes more acidic with the consumption of confections that contain citric acid and malic acid. If these samples are consumed, people consuming them should have a regular salivary flow, and if they do not have a regular salivary flow, they should take a supplement to help increase their salivary flow. The confections should be consumed whole with frequent swallowing during use, and sufficient time should pass before another confection is consumed.

These experiments can serve as the groundwork for future experiments to further analyze the fullest effects of citric acid and malic acid on the pH level of the oral cavity and tooth enamel erosion. Possible future studies could be conducted on human teeth and may deal with artificial tooth enamel specimens. Sugar free drink mix packets and energy drinks for example, are being increasingly used and could possibly produce the same effects on pH levels that confections do since the ingredients in both are similar. Future studies could be conducted using the analysis of pH levels along with other analysis techniques, including spectral surface analysis and harness testing in the case of enamel, to determine a connection between the health of the oral cavity and citric acid and malic acid consumption. These studies could lead to adjustments to the ingredients in these products so that they are not as corrosive to the oral cavity as a whole.

A new direction for future studies focusing on health and beauty fads may also be considered. Oil Pulling is a health fad that claims the continuous swishing of coconut oil in the mouth for twenty minutes daily may cure a variety of systemic diseases including headache and asthma, prevent tooth decay, cure bleeding gums, and strengthen the teeth and gums.²⁰ Oil Pulling is only one example of many fads with claims of health benefits. However, without proper research these fads may be creating a lot more harm than the benefits that they promise.

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ACADEMIC VITA

Ioana Ciocan
iac5017@psu.edu

Education

Muhlenberg High School August 2005 – May 2009
Albright College August 2009 – May 2010
Penn State University Graduate May 2014
B.S. Science, Life Science Option, Honors in Science
Schreyer Honors Thesis : Effects of various oral confections on salivary pH

Honors and Awards

Albright College- Shirk Scholar August 2009
The Pennsylvania State University- Dean's List Fall 2010 – Fall 2012, Fall 2013
Boscov Award Fall 2011 – Spring 2014

Association Memberships

Honors Club

Undergraduate Research

Biology Undergraduate Research August 2012 – May 2013
Physiological processes of *Halosimplex Carlsbadense*
Chemistry Undergraduate Research May 2013 – May 2014
Honors Thesis
Effects of various oral confections on salivary pH

Presentations

Berks County Undergraduate Research Conference, Reading (April 2014)
Effects of various oral confections on salivary pH

Positions on Campus

Biology 110 Supplemental Instructor August 2011 – May 2012
Chemistry 110 Supplemental Instructor August 2012 – May 2013
Chemistry 112 Supplemental Instructor August 2012 – May 2013
Penn State Berks Thun Library August 2011- Present
Library Aide
Penn State Berks Center for Learning and Teaching August 2012- Present
Intern
Penn State Berks Writing Center August 2012- December 2013
Biology Writing Fellow