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Determining the Presence of BPA in Vaping Mouthpieces

Jessica Cook-Snelgrove
Ouachita Baptist University

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SENIOR THESIS APPROVAL

This Honors thesis entitled

“Determining the Presence of BPA in Vaping Mouthpieces”

written by

Jessica Cook-Snelgrove

and submitted in partial fulfillment of
the requirements for completion of
the Carl Goodson Honors Program
meets the criteria for acceptance
and has been approved by the undersigned readers.

Dr. Sara Hubbard, thesis director

Dr. Timothy Knight, second reader

Dr. Elizabeth Kelly, third reader

Dr. Barbara Pemberton, Honors Program director

April 23, 2019

DETERMINING THE PRESENCE OF BISPHENOL-A IN VAPING MOUTHPIECES

BY JESSICA COOK-SNELGROVE

Bisphenol-A in Vaping Mouthpieces

Introduction:

As more and more teenagers and young adults begin to vape, whether it be to quit smoking or to look cool, there is a potential for them to be exposed to bisphenol-A (BPA) through the plastic mouthpieces on the vapes. BPA is a molecule that is added in many plastics to make the plastic harder (Houlihan, Lunder, & Jacob, 2008). Unfortunately, BPA is structurally similar to estrogen as can be seen in Figure 1. The closeness of the structures can cause issues in humans such as cardiovascular disease, diabetes, and reproductive problems (Houlihan, Lunder, & Jacob, 2008). In recent years, BPA has been removed from many items such as water bottles and Tupperware, but vaping mouthpieces remain unregulated, therefore, there is a possibility that BPA could be used in making them.

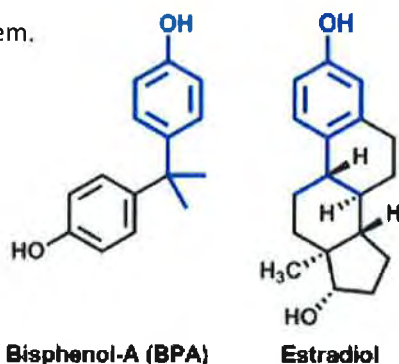


Figure 1.

People who vape come in contact with their mouthpieces multiple times throughout the day, risking exposure to any BPA that may be in the plastic. To examine the levels of BPA in vaping mouthpieces, several mouthpieces were tested for BPA content. There were two different test groups. The first was ceramic mouthpieces, which do not contain BPA. The second consisted of plastic mouthpieces, that could possibly contain BPA. In order to determine the amount of BPA that was released from each mouthpiece, the mouthpieces were submerged in 50/50 methanol-water. Samples were taken at 0, 5, 10, 15, 20, 30, 45, 60, 120, 180, and 1440 minutes. The samples were tested using a FS5 Spectrofluorometer from Edinburgh Instruments to measure the amount of fluorescence given off by the sample, which was then used to determine the concentration of BPA in the methanol-water based off of calculations from a calibration curve.

Background:

I began investigating vaping during my directed study the spring of my sophomore year. At that time, vaping was still very new, and I really did not even know it existed. Although learning about the electronic cigarettes was not my initial intent of the study, I became intrigued by the new favorite pastime of people my age.

My interest in vaping actually started with curiosity about the effects of smoking on children, which I first learned about at the Spotlight on Arkadelphia, a festival put on by local businesses, in the fall of 2016. There was a table covered in neon orange items that said "Project Prevent" in bold blue letters. The woman working the table, Mrs. Theresa Davis, called me over to win a backpack full of goodies if I signed up to help prevent adolescent smoking. As a college student who enjoys free things, and as an aspiring doctor, I quickly filled out the application. A couple of Tuesdays later, we had our first meeting with a representative from Children's Hospital named Feather Linn and began our campaign against smoking.

During the meetings, we learned about the new laws that have been passed to try and alleviate the temptation children have to smoke cigarettes. Whenever I went to gas stations, I looked to see if the cigarettes and lighters were displayed near the candy and gum and if so, I tried to compare the packaging of the tobacco products to that of the packaging of candy and gum. I saw many similarities, so I began to wonder what other techniques big tobacco companies use to appeal to children.

Dr. Kelly and I began our study with a brief history of the Minimum Age of Legal Access (MLA) in the United States to better understand the historical context of our research. As early as the 1600's, people were aware that tobacco products were addictive (Apollonio & Glantz, 2016). Because of this realization, restrictions on the sale of tobacco were implemented to keep children from becoming addicted. An article published by the Los Angeles Times in 1908 described tobacco smoking as "very popular... it is not uncommon, it is said, to see a child not more than five years old smoking contentedly." (Apollonio & Glantz, 2016).

In California at that time, it was not uncommon for those under the MLA of 16 to be sent by their employers to get cigarettes. However, those selling the products didn't know if the children were getting cigarettes as an errand or for personal pleasure. To keep that from happening, the laws about the sale of tobacco increased the MLA to 18 and forbade any sale of tobacco products to minors (Apollonio & Glantz, 2016).

For the next few decades, the states and tobacco companies fought against one another. Many times, tobacco was completely prohibited regardless of age, but the entrance of the United States into World War I quickly put an end to the efforts to keep tobacco at bay because tobacco was in the soldier's daily rations. By the 1950's, children were the main targets of the tobacco advertisements (Apollonio & Glantz, 2016). I vaguely remember eating candy cigarettes as a kid, and this sweet snack was introduced to consumers in this era. Tobacco companies such as RJ Reynolds and Philip Morris had schools come take tours of the factories, and Lorillard's Spring even claimed that teens and young adults are the "new market" (Apollonio & Glantz, 2016). By this time, efforts to reinstate strict sales policies for tobacco began to take flight once more. After years of lobbying and proposing solutions, in 1991 Philip Morris proposed that the company should encourage an MLA of 18 years old. This interesting turn of events led to the Synar Amendment in 1992 that set the MLA at 18 (Apollonio & Glantz, 2016).

Fifteen years later, a small town in Massachusetts raised its MLA to 21 years old. The rate of smoking in that area was cut in half, so many more cities followed suit. Of course, tobacco companies strongly protested. The battle back and forth of the MLA shows how important tobacco companies find young adults. To increase the change of return customers, it is important that people get addicted while they are still young and impressionable (Apollonio & Glantz, 2016).

Next in my study, I looked at the Family Smoking Prevention and Tobacco Control Act that was passed in 2009. This act prohibits tobacco companies from several advertising methods that appeal towards

children as well as requiring most of the packaging of tobacco products to be covered with warnings about the content's addictiveness and cancerous tendencies.

To begin, the new law only allows for advertisements with black text on white background and spoken words if the advertisement is anywhere besides an adult-only facility. Also, there can be no brand sponsorship of sports or outdoor advertisements within a 1,000-foot radius of a school (Law, 2009).

The packaging of cigarettes was also restricted by this new law to ensure that those who buy the product know the health risks. At least 50% of the front of the box and all of the back of the box must contain warnings about nicotine being an addictive substance and that tobacco causes cancer. Graphic images may also be used as warnings for the harmful effects of the product. In addition, no tobacco product can claim to be FDA approved or claim to be light, mild, or low-tar (Law, 2009).

Flavored cigarettes were also prohibited by the act in 2009. Because many of the flavors were made with the same chemicals found in children's candy, the flavored tobacco products were considered "kid-friendly" (Bach, 2016). Not only do they contain similar flavors, but similar packaging and display as in Figure 2. To reduce the appeal to children, the only flavored cigarette still allowed is menthol. However, tobacco companies found a loophole. By changing the dimensions of their products, cigarettes can be manipulated to meet the criteria of cigars or cigarillos, which are not monitored under the FDA like cigarettes. The power to monitor these other tobacco products lies in the hands of the state and local governments. Until laws are put in place by smaller governments, all other flavored tobacco products can continue except cigarettes (Bach, 2016).



Figure 2: Examples of similar packaging and display.

Because the new law cracked down so hard on advertisements, I wondered if there were any methods that tobacco companies were using to try to sidestep the restrictions. I thought that looking in magazines that have a higher adult readership may contain advertisements. Children may look at magazines left throughout the home, so it is conceivable that tobacco companies may utilize the idea that the parents will take the magazines home and children will read them too.

To get an idea of what magazine advertisements are out in circulation, I flipped through many magazines looking for ads for smoking or vaping. I went to the library and flipped through all of the magazines there, and I couldn't find any. Even bigger magazines like *People* and *Sports Illustrated* did not have any. I went later to Books a Million to sift through all the magazines they had, with the help of a friend, and we came up empty yet again. I tried one more time to find advertisements in Walmart's selection, but again found none. This random and limited search seemed to indicate that the new law truly reduced

magazine advertisements, but I figured that there had to be another form of advertisement that tobacco companies were using to reach their clients.

While I was doing the research on cigarette advertising, I also read the book *How to Get People to Do Stuff* by Susan M. Weinschenck, Ph.D. Dr. Kelly and I chose this book because it discusses the seven drives that can be used to motivate people to agree to do something new, buy a product, support an action, and even to change old habits or form new ones. I wouldn't think a person would begin smoking just because it seemed like the healthy choice, so there would have to be a reason for the person to begin a detrimental habit like smoking. After reading the book, I isolated two drives that I felt would appeal most towards children or teens that tobacco companies could utilize to gather new clients.

First, the need to belong. As humans, we were created to be social beings who want to be connected to each other. One way to draw someone into a group is to use a noun to identify an idea or action rather than a verb. This leads one to social validation because others are doing it (Weinschenk, 2013). By being "a smoker" or "a vaper," a child or teen will immediately feel a part of a larger group. Images with laughing, happy people will also draw in teens because they will want to experience that joy of being together. If tobacco or vape advertisements show the community a group of smokers or vapers have, then the company catches the eye of a lonely child or teen. The idea of belonging then adds to the power of stories. If a kid wants to be "a jock" he can change his story by changing what he wears and how he walks. Eventually everyone else will see him as a jock. Likewise, if vaping or smoking is perceived as cool, a kid might change their story by deciding that they are also cool so they should vape or smoke. This change in the story allows for a kid to become someone else, and, by becoming someone else, become part of a group. If the words on an advertisement for smoking or vaping match the story of a "cool person," then a child or teen may be much more enticed to start using that product. For example, the advertisement below in Figure 3 shows a pretty woman, a group of people releasing lanterns, and another woman dressed up for a masquerade party. This



Figure 3.

could seem to a teenager as a girl they would like to either become or date, and a group of people that are a lot of fun to be around. Also, words like moments, imagination, and discovery sound rather intriguing, fun, and hipster. To join this crowd of awesome people, buy Camel cigarettes.

Because smoking cigarettes has shifted from being viewed as cool to being viewed as unhealthy, many have turned to vaping. It is supposedly healthier because it doesn't have many of the carcinogens or additives that cigarettes have. However, because the juices are not regulated by the FDA,

no one is certain if the vapor is truly only vegetable glycerin, propylene glycol, and nicotine. Many vape advertisements offer vaping as a healthy way to quit smoking. Unfortunately, it is believed that the transition from vaping to smoking can also occur. Kids start vaping because it looks "cool" and then make the switch to cigarettes when they get older.

I had yet to see many advertisements in magazines, so I decided to change tactics for a couple of weeks. I visited several convenient stores around the area to see where I could purchase a vape pen if I wanted to do so. We went to several gas stations and Walgreens. In both Walgreens and the Phillips gas station, we found vape pens with a multitude of flavored juices. We noticed that many of the flavors were that of ice cream, such as mint, vanilla, and chocolate. The vape displays were usually in close proximity to the "power walls," which are the displays where the boxes of cigarettes are neatly arranged, typically close to the cash register (Law, 2009).

These power walls can be monitored by local or state government under the 2009 Family Smoking Prevention and Tobacco Control Act. Many times, all one can see upon entering a gas station is one of these walls, and children are no exception. Even though it isn't an advertisement in technical terms, it is definitely a way to promote the sale of tobacco products, so perhaps the state should consider prohibiting power walls at the entrance of a store.

Because the vape displays were sort of in the shadow of the cigarette displays, I thought it would be helpful to learn more about a shop that focused more on vape products. I interviewed a man I know who worked at the Vape Shop in my hometown of Camden, Arkansas. I asked him how the store advertised its products, and he said that the store has ads in the local papers and magazines as well as on the radio. I also asked him what his average client age range was. He gave me the range of 20-45. When asked if they send email advertisements to any of their clients, I was told all they did was a text club. I was also curious if the store itself contained ads for the different brands. It does, with posters of the different vaping flavors hung around the store. They carry 101 juices, and the top flavor is called "Monroe," which is fruity. This store has only been open since 2015, so they may not have fully developed their advertisement strategies yet.

The next investigation I performed was a survey for a few people I know who vape. The results I received were almost contradictory. First, a couple of my participants said they started vaping to quit smoking. Another told me that he started using electronic cigarettes because a friend's mom had one and he was curious about what it tasted like. He was 16, and it was coffee flavored. Then, when he was in college, he started to smoke. The former contrasts the latter. It appears that there is a two-way street between vaping and smoking. Two of my participants did say that vaping was helping them quit smoking, but still isn't making it easy to stop.

After my interview with the participant who started using e-cigarettes at the age of 16, I was curious to see how much of an impact vaping has on the youth in the United States. It appears that youth are more strongly affected by marketing ploys because youth look at media to know how to dress and how they compare to their peers (Krugman, 2016). There are three areas where it seems adolescents are most responsive to marketing: cognitive, emotional, conative (Krugman, 2016). The cognitive area would relate to a teen's learning or attentiveness, emotional to the need to belong or the want of something, and conative to an action. Vaping advertisements can touch on all of these responses with a single image and a few words.



Figure 4.

The Blu advertisement to the left is an example. An almost naked woman definitely catches the attention, and the word “slim” describes what girls are expected to look like, reflecting that in the product. “Ready to go” is vague as to where, but for a teen, that may sound like freedom to go anywhere. The advertisement does its job well, because it sticks out on the page and in the mind of the viewer.

In a study done in 2016 on the correlation between e-cigarette advertisement exposure and the use of e-cigarettes in adolescents, it was found that the more exposed to the advertisements, the greater

the likelihood of vaping (Dai & Hao, 2016). According to this study, there are less advertisements for vaping in magazines and newspapers than on the internet, in stores, and on TV. Many of the students interviewed reported that they didn't use e-cigs to stop smoking, but rather because they taste good. Several also reported that they wanted to just try it out and to hang out with friends (Dai & Hao, 2016). Shortly after this study was conducted, the FDA expanded the act from 2009 to include all tobacco products, thereby prohibiting the sale of e-cigarettes to anyone under the age of 18 (Dai & Hao, 2016). The act also regulated the advertisements for e-cigarettes. If one visits a website to try to find vaping supplies one is asked if one is over 18, from my personal experience in my attempts to conduct research.

Another interesting outcome of this study was the impact that e-cigarette users in a home have on teens. If another member of the household used e-cigarettes, then it was more likely that a youth in that same home would begin to use e-cigarettes (Dai & Hao, 2016). From the testimony of my interviewees, I think that teenagers are curious. Having been a teen myself, rather recently, I can attest to the fact that I've spent a semester researching vape products to satisfy my curiosity. If someone else in the house is using a

cool little device that is not supposed to be harmful to one's health, why not try it? Especially if the user is an adult. Teenagers simply want to be seen as mature, so mimicking an adult would fulfil the image the teen is yearning for.

Having not personally seen an advertisement on television for e-cigarettes, I was surprised that so many youth reported being exposed to advertisements by watching TV (Dai & Hao, 2016). I decided to read one more study to see what kind of channels or regions had the highest rate of advertisement showings. Channels like Comedy Central and AMC had the highest amount of advertisements aired (Duke, 2014). Because this study was conducted prior to the Family Tobacco Act Amendment in 2016, I don't think that the data is representative of today's youth. However, I did think it was odd that I couldn't remember seeing these ads in my youth.

I learned a lot that semester about how smoking and vaping ads affects kids, teens, and even young adults like myself. I hoped to take this knowledge back to Project Prevent to incorporate my new understanding of vaping and its appeal to youth into our battle to counteract the effect of the advertisements and promote overall public health, beginning with the younger generation. When it came time to choose my thesis topic, I decided I would continue with vaping in young adults. The initial plan was to look at the effects of a metabolite of nicotine, called cotinine, on the environment. However, after receiving a SURF grant to do the research, the cotinine proved impossible to attain. Instead, I chose to examine a different aspect of the effects of vaping on people that did not involve the vape juice.

Because Dr. Hubbard was already doing research on BPA in plastics that kids are often exposed to, I thought it would be interesting to look at BPA in something that young adults are often exposed to. Because teens are still developing, there is potential for BPA to harm them. BPA has been limited in many plastic products such as water bottles and baby toys, but many things are not monitored for BPA. According to an article published on WebMD, 90% of people have BPA in their systems due to containers that food comes in, water bottles, and even dust in the air (Brennan, 2017). Another issue mentioned in the article is the lack of

definite answers on what BPA does to humans and how much is toxic. The levels currently allowed by the FDA are supposed to be safe, but more research is underway to be sure this is true (Brennan, 2017). At this time, the toxicity data suggest the dose-effect level of BPA to be 5 mg/kg bw/day; oral exposure (Aungst, 2014). Because so little is known about vaping and the mouthpieces are unmonitored, I chose to look at BPA in vaping mouthpieces to see if teens who vape should be concerned about their exposure to BPA.

Since conducting my research, there have been new developments in regulation of vaping. In Arkansas, there is a proposition for new taxes on vaping products in an attempt to reduce teen vaping. However, there is a strong opposition from the vaping community. One of the handouts says that “SB571 is proposing taxing ADULT VAPERS 67% because teens keep ILLEGALLY vaping.” The handout continues to explain how there are thousands of jobs provided in Arkansas because of the vape stores in the state. It also points out that tens of thousands of Arkansans have switched from smoking to vaping, improving the lifestyles of those people. The handout also provides statistics about the number of Americans who will die this year from smoking. It further provides a study from the UK College of Physicians that has shown vaping to be 95% safer than smoking, and in addition smoking rates are at their lowest disproving any gate-way effect (although that is not consistent with previous findings in this paper). The paper also mentions a study that examined the lung tissue of vapers who have been vaping for 3.5 years, and there was no long-term damage to the tissue.

The handout argues that it doesn't make sense to tax the people who are legally vaping and trying to quit smoking when underage teens are illegally vaping. Moreover, the handout pleads for people to support enforcement actions against underage vapers and those who provide to them rather than taxing a small percentage of the population who are attempting to improve their lives. Whoever created this handout emphasized that vaping is for smokers who do not want to smoke, and that minors should not be vaping.

Clearly, vaping is a very debated topic, and research on vaping is very important because it affects so many people. For this reason, I decided to research a novel aspect of vaping, BPA in the mouthpieces.

Because BPA is a fluorescent compound, using fluorescence spectroscopy is an ideal way to investigate the presence of BPA because it is specific and sensitive to the compound, even at low concentrations. After being excited at 280 nm and absorbing energy, BPA emits light at 310 nm in the form of fluorescence as it returns to ground state, as shown in figures 5 and 6.

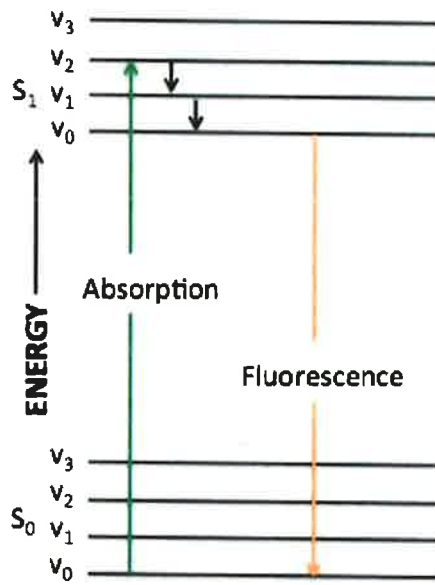


Figure 5.

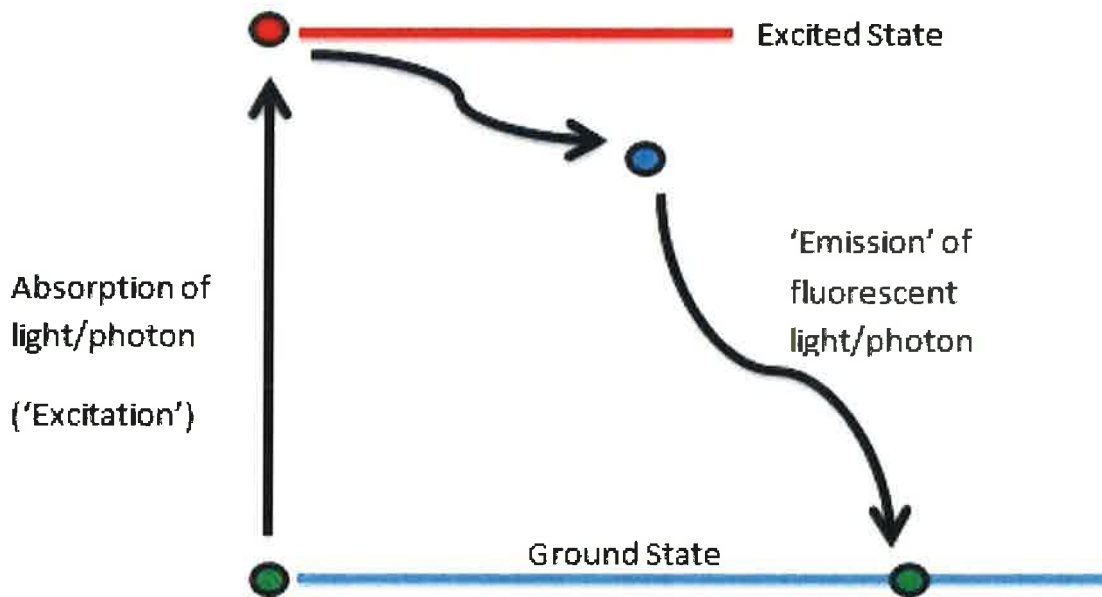


Figure 6.

Materials:

- Vape Mouthpieces (2 identical ceramic and 5 different plastic)
- HPLC Grade Methanol
- HPLC Grade Water
- 22 10-mL Volumetric Flasks
- FS5 Spectrofluorometer
- Pasteur Pipettes
- 10% Nitric Acid
- KimWipes
- BPA
- 25-mL Volumetric Flasks
- Medium Gloves
- Goggles
- Labelling Materials
- Parafilm

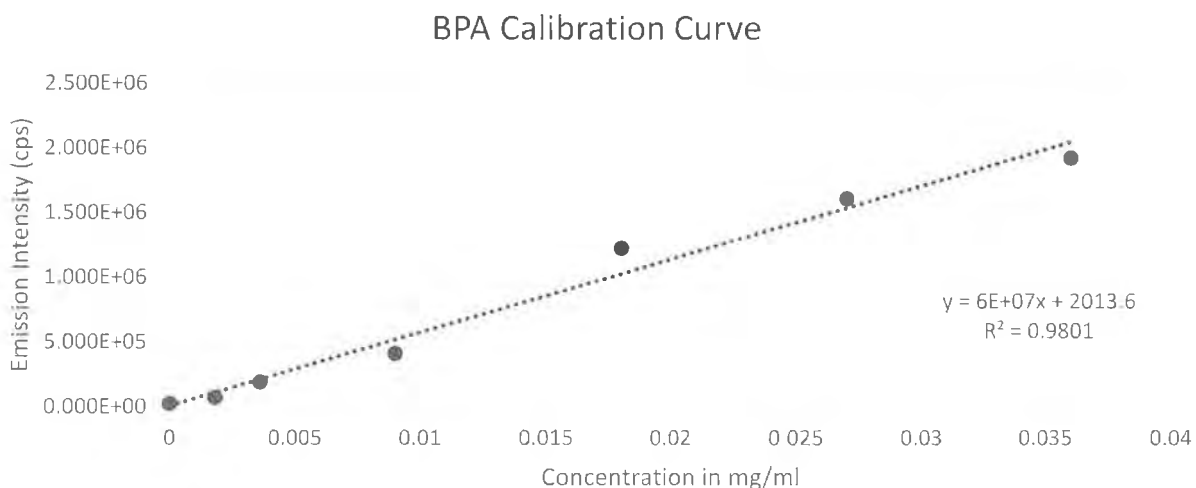
Methods:

Before the experiment could take place, all of the glassware that would be used was cleaned using 10% HNO₃. After the acid was in the glassware for 20 minutes, the glassware was rinsed 3 times with reverse osmosis water. Then, the glassware was allowed to dry overnight in a drawer or in the 80° C oven if the glass was needed that day. Once the glassware was clean, dry, and cool the experiments were able to be performed. After using glassware, the same protocol was followed before using the glassware again. The cleaning process allows for more accurate results by eliminating some possible sources of contamination in the samples. When working in the lab or instrument room, gloves and goggles were worn at all times.

To begin, a calibration curve was obtained using a prepared BPA dilution series. The standard BPA solution was made by measuring 0.0045g BPA into a 100 mL volumetric flask and then filling to the line with 50/50 methanol water. The methanol water solution was prepared by measuring 100 mL each of HPLC grade methanol and HPLC grade water and combining them in a 200 mL volumetric flask. Due to the hydrogen bonding of the methanol to the water, the volume is less than 200 mL, which is why each must be measured separately rather than filling to the line in a volumetric flask. If one was measured to 100 mL and then the other was filled to the line, the solution would not be 50/50. Methanol water was made as needed

throughout the experiment. Using the equation $M_1V_1=M_2V_2$, dilutions were made so that the calibration curve contained 0.0 mg/mL, 0.0018 mg/mL, 0.0036 mg/mL, 0.009 mg/mL, 0.018 mg/mL, 0.027 mg/mL, and 0.036 mg/mL BPA in the methanol water solution. After preparing the dilutions, each concentration was analyzed in the spectrofluorometer in triplicate, and the averages of the emission wavelength (Em), emission intensity (Em I), excitation wavelength (Ex), and excitation intensity (Ex I) are shown in the following chart. The BPA Calibration Curve chart was used to calculate the concentration of BPA in samples later by relating the emission intensity to the concentration of BPA in the methanol water.

Concentration (mg/mL)	Em (nm)	Em I (cps)	Ex (nm)	Ex I (cps)
0	306	1.840E+04	278.6667	1.016E+04
0.0018	305	6.416E+04	278.333	3.575E+04
0.0036	304.6667	1.867E+05	277	1.010E+05
0.009	303	4.060E+05	275.6667	2.336E+05
0.018	303.3333	1.223E+06	275.6667	7.005E+05
0.027	303.6667	1.606E+06	274.3333	9.243E+05
0.036	304	1.918E+06	272.3333	1.124E+06



Following the creation of the calibration curve, the mouthpieces were tested. The first day, a ceramic mouthpiece and a small marbled teal plastic mouthpiece were each submerged in beakers containing 150 mL of 50/50 methanol water. Because two mouthpieces were being tested at the same time, all glassware that contained samples from the ceramic mouthpiece had labels with “C (minutes)” and the glassware with the

plastic mouthpiece samples had labels with "P (minutes)." Once the mouthpieces were submerged in the methanol water, 10 mL samples were taken using a pastuer pipette as 0, 5, 10, 15, 20, 30, 45, 60, 120, 180, and 1440 minutes. As soon as a sample was placed in a 10 mL volumetric flask, the flask was sealed using parafilm then placed in a drawer to keep the samples from degrading due to light exposure. After the 24-hour samples were taken, the samples were run through the SF5 Spectrofluorometer by Edinburgh Instruments. In between each sample, two rinses were done. Then, each sample was run in triplicate (unless there wasn't enough of the sample left for the third due to spilling), and measurements were taken for the emission wavelength and intensity as well as the excitation wavelength and intensity. The results were recorded in a table in the lab notebook (see Results). Unfortunately, the plastic mouthpiece that was tested on this day may have inaccurate results due to an error. The lamp in the instrument did not ignite and the experimenter, being unfamiliar with the instrument, did not restart the spectrofluorometer in order to fix the issue. There was an identical teal plastic mouthpiece that was tested later to replace the results seen in this trial.

The next two mouthpieces were both plastic, so they were labeled based on color. The first was clear, so it was "C (minutes)" and the second was a translucent yellow, so it was "Y (minutes)." On the third day of testing, another ceramic mouthpiece was tested, labeled again with "C (minutes)" and a marbled teal plastic mouthpiece was tested in order to correct the error from the first day of the experiment. The samples taken from the plastic mouthpiece were labeled "P (minutes)." On the fourth day, only one mouthpiece was tested. It was a large plastic teal mouthpiece. The next mouthpiece that was tested was a white plastic mouthpiece. When it was placed in the methanol water, this mouthpiece floated, roughly halfway submerged. The samples were still taken, but the results were expected to be lower since the whole mouthpiece was not in the solvent to release BPA. The protocol remained the same with all of the mouthpieces, and the results are summarized in the Results section of this paper.

During the course of the experiment, two ceramic mouthpieces and five plastic mouthpieces were analyzed. The five plastic mouthpieces do not include the first plastic mouthpiece due to the error that

occurred. To analyze the data, an average and standard deviation were found for each of the triplicate samples from each of the mouthpieces for the emission wavelength, emission intensity, excitation wavelength, and excitation intensity. The emission intensity was the parameter used to evaluate the increase in BPA released into the methanol water, and the averages are summarized in a table and as a graph for each mouthpiece in the following section, including a linear regression and an R^2 value.

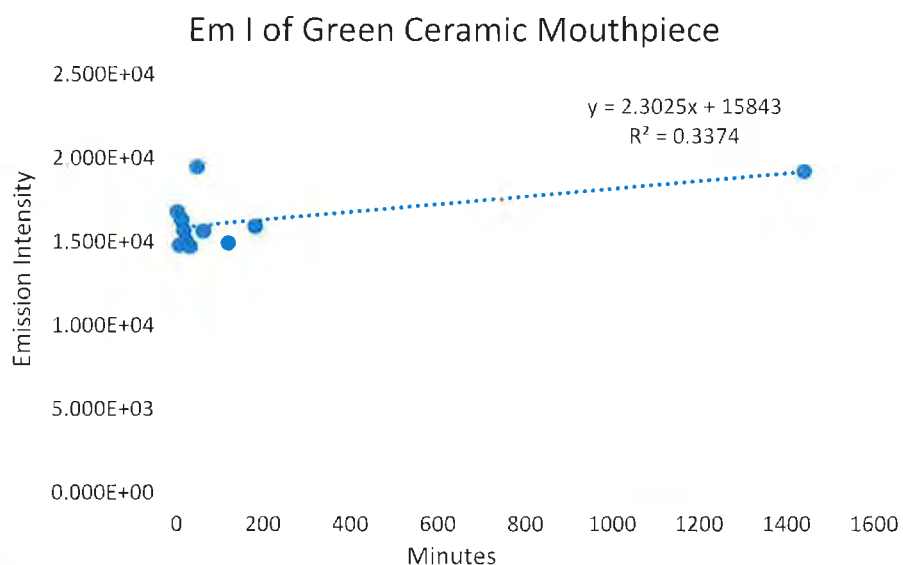
Results and Discussion:

Tests from Day 1 (10-10-18): Ceramic Mouthpiece (Control) and Plastic Marbled Teal mouthpiece

(data not evaluated due to instrument malfunction)

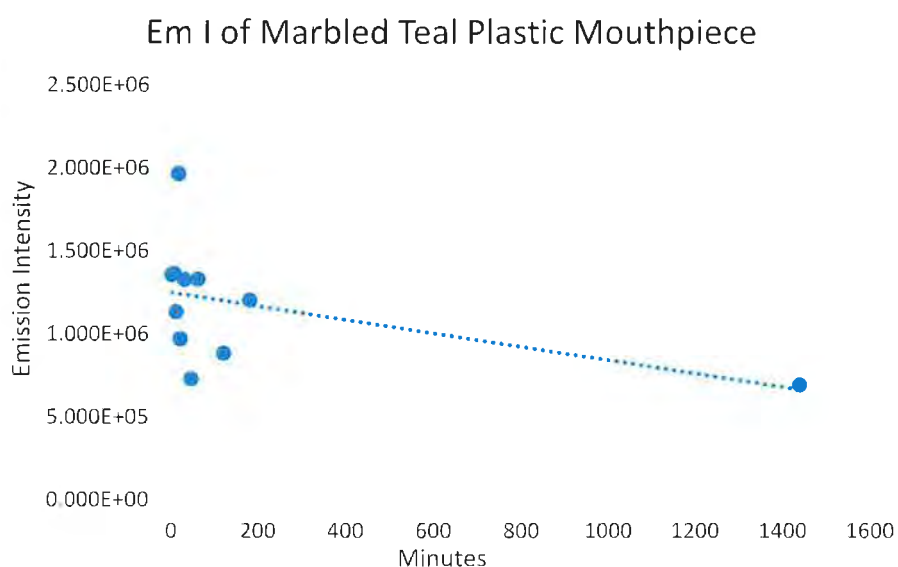
In the Ceramic Mouthpiece, there is little variation in Emission Intensity at the given time intervals.

Em I	Minutes
1.680E+04	0
1.480E+04	5
1.635E+04	10
1.570E+04	15
1.508E+04	20
1.473E+04	30
1.950E+04	45
1.566E+04	60
1.491E+04	120
1.594E+04	180
1.924E+04	1440



The readings for the Marbled Teal Plastic Mouthpiece were inconsistent due to the researcher being unaware that the instrument was not properly calibrated. Therefore, the data was recorded but is not valid for interpretation.

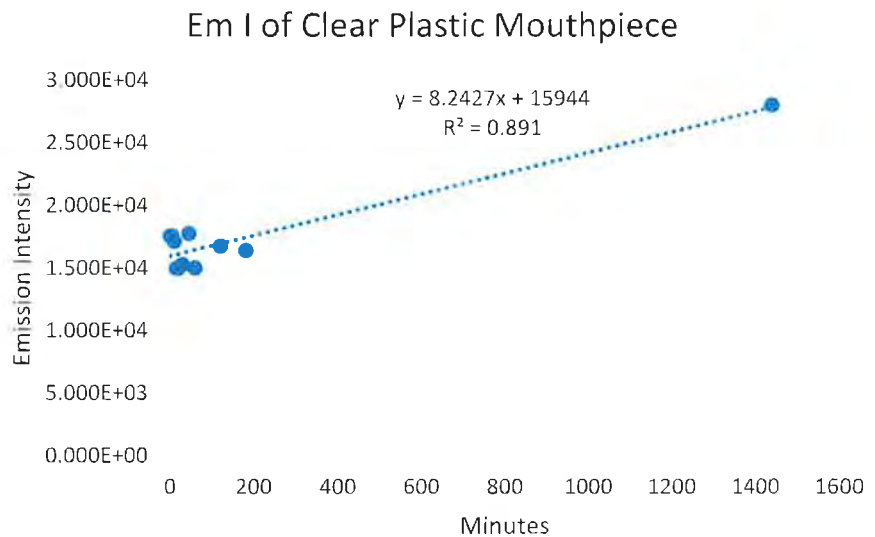
Em I	Minutes
1.680E+04	0
1.480E+04	5
1.635E+04	10
1.570E+04	15
1.508E+04	20
1.473E+04	30
1.950E+04	45
1.566E+04	60
1.491E+04	120
1.594E+04	180
1.924E+04	1440



Tests from Day 2 (10-15-18): Clear Plastic Mouthpiece and Translucent Yellow Plastic Mouthpiece

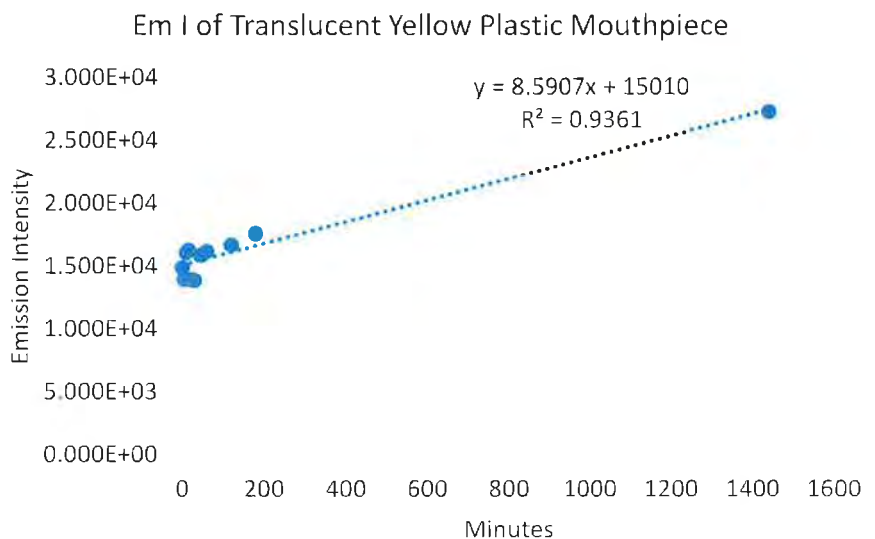
For the Clear Plastic Mouthpiece, the measurements stayed at a mostly consistent reading until the measurement of the 24 hr. sample, which showed a higher intensity, suggesting that more BPA had been released during the longer period of time.

Em I	Minutes
1.755E+04	0
1.752E+04	5
1.713E+04	10
1.497E+04	15
1.497E+04	20
1.524E+04	30
1.775E+04	45
1.499E+04	60
1.674E+04	120
1.638E+04	180
2.801E+04	1440



The Translucent Yellow Plastic Mouthpiece had a clearer trend of increasing emission intensity, although it too was roughly the same early in the experiment with a noticeable difference between the 3 hr. and 24 hr. tests.

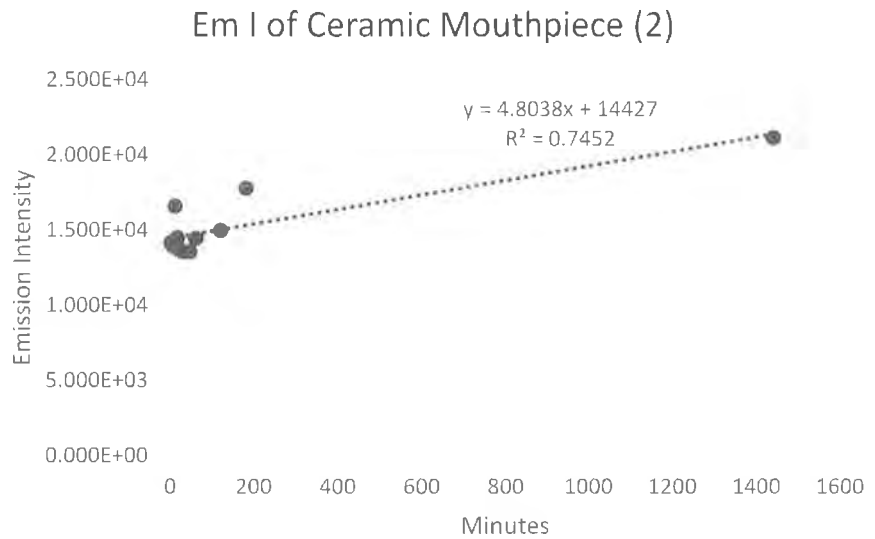
Em I	Minutes
1.481E+04	0
1.386E+04	5
1.600E+04	10
1.619E+04	15
1.384E+04	20
1.379E+04	30
1.577E+04	45
1.609E+04	60
1.660E+04	120
1.748E+04	180
2.722E+04	1440



Test from Day 3 (10-17-18): Ceramic Mouthpiece 2 (Control)

To be sure that the experiment was valid, a second Ceramic Mouthpiece identical to the first was tested. The results from this control were slightly less consistent, but still showed the same general trend of the emission intensity not increasing very much after 24 hrs.

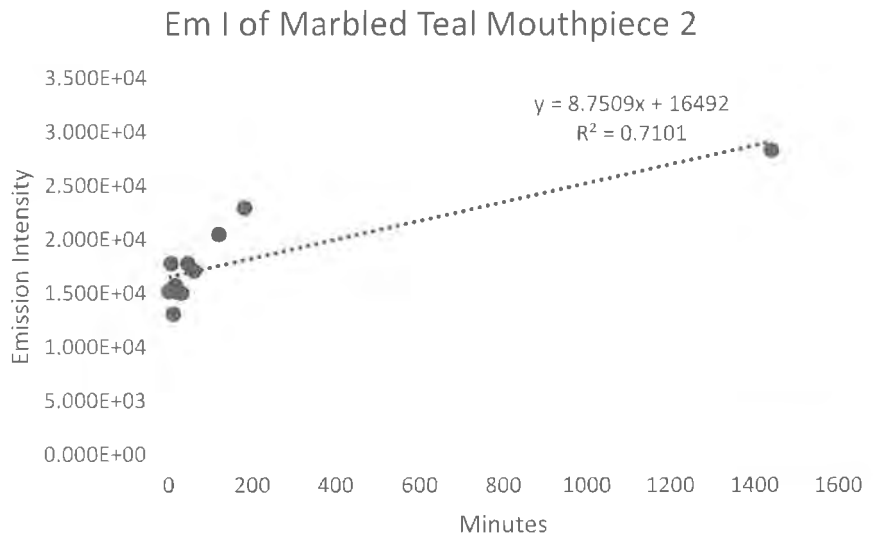
Em I	Minutes
1.412E+04	0
1.390E+04	5
1.656E+04	10
1.444E+04	15
1.367E+04	20
1.351E+04	30
1.351E+04	45
1.441E+04	60
1.495E+04	120
1.776E+04	180
2.111E+04	1440



Tests from Day 4 (10-22-18): Marbled Teal Plastic Mouthpiece 2 (identical to the first plastic sample) and Large Marbled Teal Mouthpiece, and White Plastic Mouthpiece

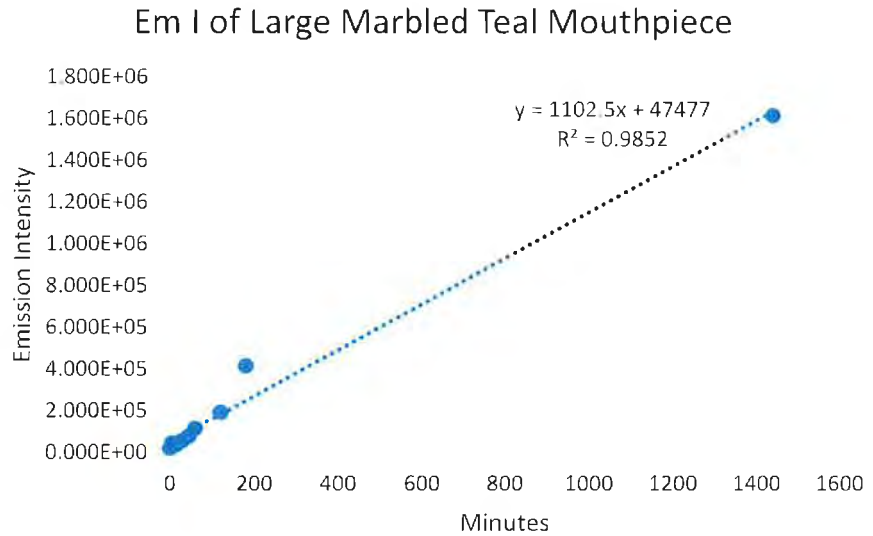
To correct the issue from the first Marbled Teal Plastic Mouthpiece, a duplicate mouthpiece was tested. This mouthpiece showed a steady increase in the emission intensity as time passed, suggesting that BPA was being released into the methanol water.

Em I	Minutes
1.516E+04	0
1.775E+04	5
1.301E+04	10
1.571E+04	15
1.508E+04	20
1.501E+04	30
1.776E+04	45
1.707E+04	60
2.047E+04	120
2.293E+04	180
2.830E+04	1440



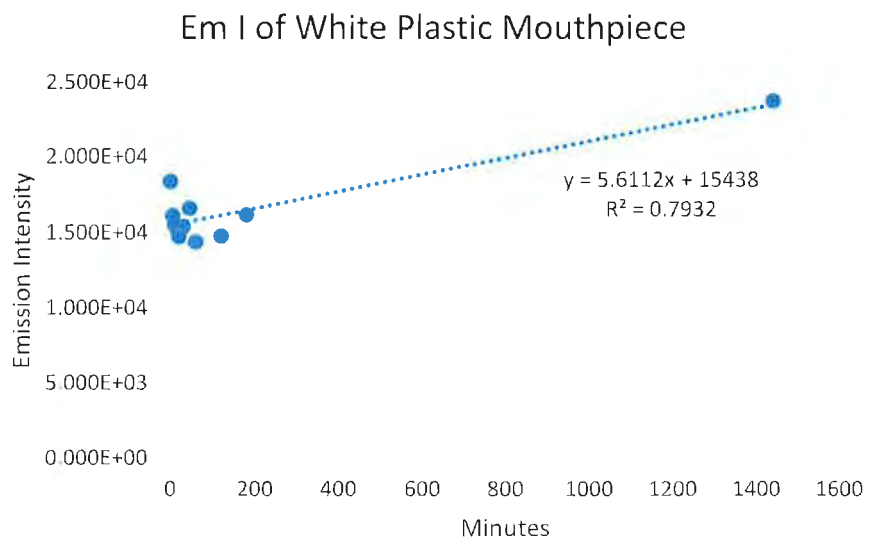
The Large Marbled Teal Mouthpiece was almost double the size of the other mouthpieces, with a much larger surface area to be submerged in the methanol water. The measurements of emission intensity reflected the large size, by showing a much higher intensity than seen in any other mouthpiece.

Em I	Minutes
1.880E+04	0
4.303E+04	5
3.495E+04	10
3.388E+04	15
4.487E+04	20
5.591E+04	30
7.808E+04	45
1.148E+05	60
1.912E+05	120
4.140E+05	180
1.615E+06	1440



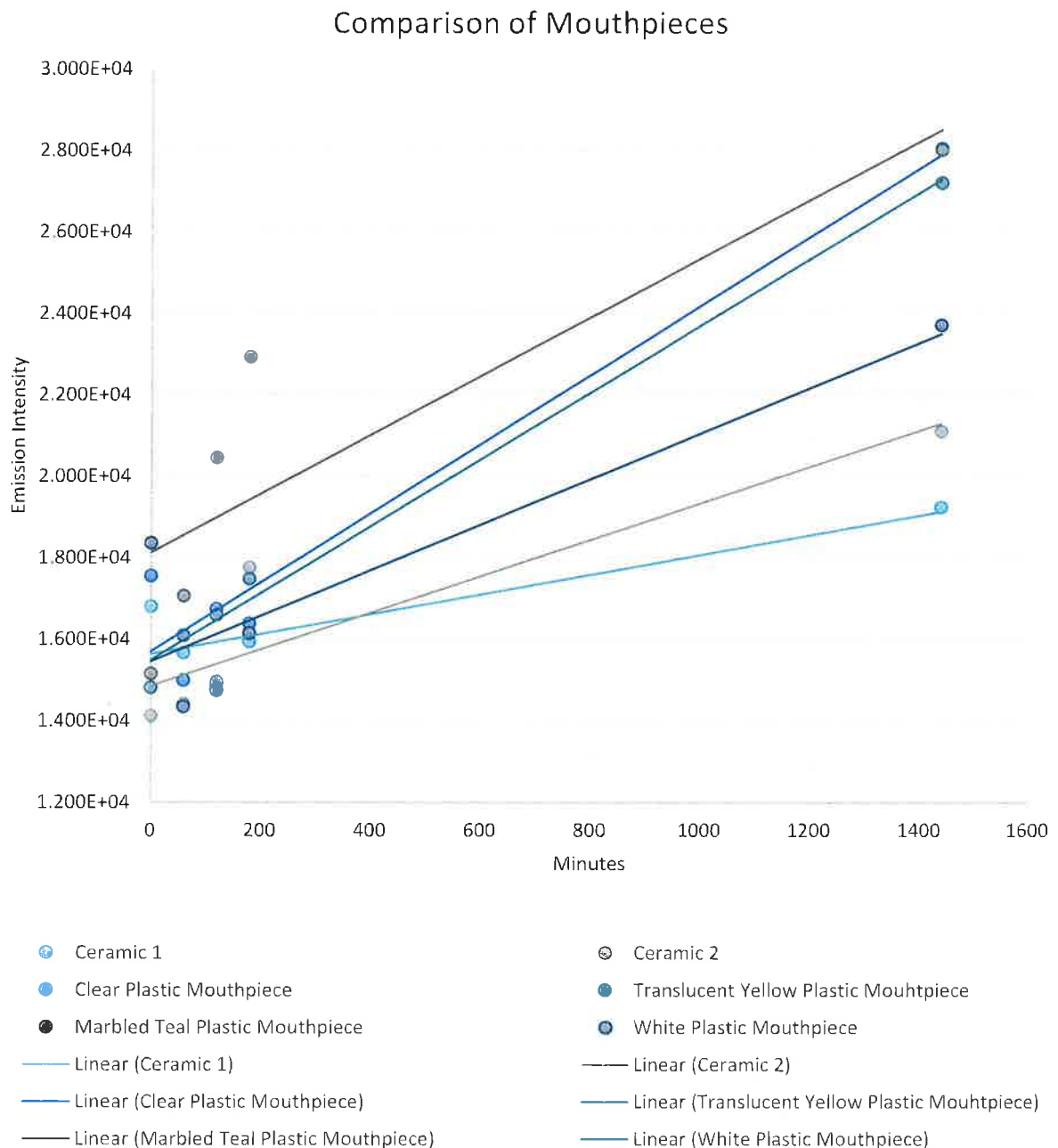
The last sample was a White Plastic Mouthpiece. Unfortunately, this mouthpiece was made from a lighter plastic, which had a lighter density than the methanol water solution that caused it to float. Because the White Plastic Mouthpiece was not fully submerged as the other mouthpieces were, the results from this sample are not comparable to the other mouthpieces, but even though it was not completely under the solution, there is still a slight increase in emission intensity.

Em I	Minutes
1.836E+04	0
1.608E+04	5
1.542E+04	10
1.519E+04	15
1.467E+04	20
1.536E+04	30
1.658E+04	45
1.434E+04	60
1.475E+04	120
1.615E+04	180
2.372E+04	1440



Comparison of Mouthpieces:

To compare emission intensity of all the mouthpieces with valid data, the average intensities at 0, 60, 120, 180, and 1440 minutes are show in the graph below. The hour increments were chosen to make the graph more concise to maximize the visualization of the differences between mouthpieces. The Large Marbled Teal Plastic Mouthpiece is excluded from this chart because the intensity was so much greater that it made it difficult to view the other points on the graph.



Conclusions:

Using the calibration curve, the limits of detection and quantification were determined. Quantitative

Figures of Merit:

- Limit of Detection: 1.44×10^{-4} mg/mL
- Limit of Quantification: 4.79×10^{-4} mg/mL
- Equation: $(Em / 6e7) - 2013.6 = \text{Conc.}$

When the emission intensities are plugged into the equations, negative values are returned for all of the concentrations for all of the mouthpieces, meaning that the concentrations are well below the limit of detection. Unfortunately, this means that the results of the experiment cannot be used to draw any conclusions. There is an increased intensity with the large marbled teal mouthpiece when compared to the other plastic mouthpieces, suggesting that a bigger mouthpiece may have more BPA in it. It does appear that there is an increase in emission intensity for the plastic mouthpieces compared to the ceramic mouthpieces, but again these are also below the limit of detection so no conclusions can be drawn at this time.

Future Experiments:

In the future, the mouthpieces should be tested while the methanol water is being heated to mimic the conditions of the hot vapor passing through the mouthpiece. Because BPA release is affected by heat, it is plausible that more BPA will be found in the samples taken when the hot methanol water than the room-temperature methanol water. In addition, all of the trials in this study need to be repeated many times to ensure the accuracy of the data. Perhaps results would be conclusive if it were possible to increase the ratio of mouthpiece to methanol water to improve the concentration in the samples. This could be possible by using less methanol water and using three identical mouth pieces or potentially using standard addition of a BPA solution to the methanol water to bring the concentration to a detectable level.

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