

## **Evaluating the Effects of using Plastic, Silicone, and Glass on Time of Sous Vide Cooking of Gravy**

**May-Lee Guan<sup>1</sup>, Helen Heacock<sup>2</sup>, Lorraine McIntyre<sup>3</sup>**

1 Lead Author, B. Tech Student, School of Health Sciences, British Columbia Institute of Technology, 3700 Willingdon Ave, Burnaby, BC

2 Supervisor, School of Health Sciences, British Columbia Institute of Technology, Burnaby, BC

3 Contributor, BC Centre for Disease Control, 655 West 12th Ave, Vancouver, BC

### **Abstract**

**Background:** Sous vide cooking is popular method of cooking involving a water bath with immersion circulator or a steam convection oven. This process is also known as low-temperature long-time method (LTLT), where food is held at a lower temperature over extended time for cooking (1). Plastic is the more commonly used medium with sous vide cooking. Glass and silicone are an environmentally alternative to plastic, since they are reusable mediums. These alternative mediums have not been studied and may affect the time to reach pasteurization. The purpose of this experiment is to determine how plastic, silicone, and glass influence time in sous vide cooking of gravy to 56.5°C.

**Methods:** Four of each medium: plastic, silicone, and glass containing 500 mL of gravy with SmartButton data loggers at 4°C were introduced into a 56.5 °C water bath for 150 minutes. The data loggers recorded the temperature at one minute intervals. The data was used to run a One-Way Analysis of Variance (ANOVA) to analyze if there were any statistically significant differences between the three mediums and time, and a Scheffe's test to compare the mean time of each of the mediums.

**Results:** There was a difference in time of sous vide cooking for gravy at 56.5°C between the mediums: plastic, glass, silicone. The p-value was 0.00, therefore rejecting Ho and accepting there is a difference in time of sous vide cooking gravy at 56.5°C between the different mediums: plastic, glass, silicone. Comparing the mediums among each other, it showed that there was a difference between glass and silicone, glass and plastic and no significant difference between plastic and silicone medium.

**Conclusion:** The results indicate that there is a statistically significant difference in time that it took for gravy to reach 56.5°C between the mediums: plastic, silicone, and glass. The mean time for each medium to reach 56.5°C differed; 65 minutes for plastic, 68 minutes for silicone, and

129 minutes for glass. This shows that a more environmentally friendly alternative to plastic sous vide pouches can be used. Silicone pouches show to be the best alternative, least compromising of the come-up and pasteurization time. If sous vide users opt to use glass, the come-up time almost doubles in time.

**Keywords:** Sous vide, gravy, silicone, glass, plastic, SmartButtom, public health

## **Introduction**

Sous vide cooking has been a popular practice that has been growing in the culinary industry in recent years. Sous vide is a method of cooking where food is commonly placed in a plastic pouch, vacuumed sealed, and cooked in a water bath or steam oven. This style of cooking is not just limited to placing food in a plastic medium, but other options include glass or silicone. Glass jars are commonly used with recipes that require liquid or high cooking temperatures. Glass is known to be a good insulator of heat (2). Moreover, silicone is an alternative to plastic. Both glass and silicone attribute to a more environmentally sound method by reducing the carbon footprint and saving space in landfills, whereas plastic cannot be reused. Using mediums such as glass and silicone will relieve some of the anxiety about food safety of cooking in plastics, which is a main concern of sous vide cooking. A concerning issue in this type of cooking style is that the low temperatures may not be sufficient to kill microorganisms.

With sous vide cooking, juices are produced from the foods because no evaporation is

occurring within then enclosed medium. These sous vide juices are commonly made into a sauce (3). The type of food that will be examined in this study is gravy. Studying these different mediums of plastic, glass, and silicone, will help determine how they influence time and whether additional information in guidelines for sous vide cooking is required.

## **Literature Review**

### ***What is sous vide cooking***

The French term “sous vide” means “under vacuum” and describes a method of cooking where food is vacuum sealed and placed in a water bath or steam oven under controlled temperature and time (4). This process is also known as low-temperature long-time method (LTLT), where food is held at a lower temperature over extended time for cooking (1). Cooking and pasteurization occurs simultaneously to destroy pathogens and produce a product with a longer shelf life (5). The equipment required for sous vide cooking typically comprises of a thermometer, sous vide tape, vacuum sealer, water oven or thermal circulator with a

container, and mediums: plastic pouches, glass jars, or silicone bags (6).

### ***Procedures of sous vide cooking***

Sous vide cooking typically can be used for: cook-hold, cook-serve, cook-chill, or cook-freeze (7). The sous vide cooking process consists of four steps: prep, package, cook, finish. The initial step is prepping the food the same way as traditional cooking methods with seasoning. The next step involves packaging the food into the medium. The most common mediums used are food graded plastic pouches. The air is removed most commonly with the use of a vacuum sealer. Removal of air helps with two aspects, it prevents the plastic from floating in the water bath and provide an even distribution of heat for the food item (8). The next step is selecting the appropriate time and temperature for the food. The temperature of the water bath is set and is dependent on the type of food. The British Columbia Centres of Disease Control (BCCDC) recommends sous vide pasteurization is achieved when the internal food core temperature is held long enough to achieve a 6.5 log bacterial reduction (1). Another factor to beware of for sous vide cooking is that overcrowding can affect the distribution of temperature, which result in requiring longer cooking times (9).

The last step of the sous vide process can consist of any of the following, portioning,

repacking, and preserving cooked food to use later. If the food is refrigerated, it is recommended to be labelled with the with date, time, discard date and identity (1).

### ***The advantages of sous vide cooking***

Sous vide cooking differs from traditional cooking by using vacuum sealing and precision control over temperature. These provide advantages over traditional cooking methods. Sous vide is especially useful for cooking meats and seafood, for which the window of proper doneness is small, this helps provide consistent results. Whereas, traditional cooking with a range, oven, or grill uses high and fluctuating temperatures, there is little margin for error. Sous vide cooking preserves moisture, flavor, color of food and by breaking down the proteins – it results in food that is more tender (1).

Vacuum sealing has many benefits, which include reduction of lipid oxidation for an extended shelf life and prevention of the loss of volatile flavors because of vacuum packaging (10). It also reduces aerobic bacteria growth. Sous vide cooking also enables the retention of nutritional values in addition to enhancing flavors and providing hygienically safe foods (11).



**Figure 1. Sous Vide Temperature of Safety Zone**

### ***Risk and concerns of sous vide***

Traditional cooking requires most products to reach an internal temperature of 75°C for 15 seconds to be considered safe for consumption (12). With sous vide cooking the use of LTLT, is used to achieve pasteurization. The recommended temperature for sous vide cooking is to be a minimum of 55°C, except for poultry products which require a minimum of 60°C. (1). The danger zone is the temperature between 60°C and 4°C is where pathogens can grow in potentially hazardous foods (PHFs), but with sous vide cooking the danger zone is in a decreased range since it uses LTLT. Shown in figure 1, the danger zone for sous vide cooking is between 10-55°C (1). The time and temperature control is crucial in the process of sous vide pasteurization, if not controlled properly it can support the growth of pathogens and lead to a foodborne illness. Most data on microbial growth in food is based on

temperatures below 40°C, with studies focusing on how bacteria grow at ambient temperatures, for example during storage (13). Lack of information in the range of about 40 to 60°C makes it very difficult for cooks and environmental health officers (EHOs) to calculate the lethality of such low temperature heat treatments and judge the risk of foods containing pathogens.

When cooking with sous vide, it creates an anaerobic environment which then provides an opportunity for pathogens such as *Clostridium botulinum*, *Listeria monocytogenes*, and *Bacillus cereus*. There have been cases of vacuum-packaged foods causing outbreaks of botulism in the past (14). To control for botulism, it is recommended that vacuum-packaged foods should be stored below 3.3°C (1). Even after sous vide pasteurization, if foods are refrigerated, holding temperature are still required to be less than 3.3°C, as spores of C.

botulinum could still be present and viable (1).

Another concern with sous vide cooking is that there is no high temperature kill step for bacteria. Some bacteria are also able to form spores that are very resistant to heat and chemicals. Pasteurizing does not reduce pathogenic spores to safe levels and pathogenic spores can outgrow and multiply to dangerous levels leading to cause foodborne illnesses (FBIs) (7). Bacteria that can form spores include *Bacillus* and *Clostridium* (15)

In 2014, there were two outbreaks caused by sous vide cooking recorded by Vancouver Coastal Health Authority of BC. The cause of these two outbreaks were linked to improper preparation of sous vide style shell eggs and duck breast resulting in *Salmonella enteritidis* (16). Recommendations that were made to chefs who use sous vide method of cooking, was to use approved safety plans and EHO's should beware of how to evaluate these premises and assess the risk of the type of protein used (16).

#### ***Mediums: Plastic, Glass, Silicone***

Plastic is the most common method used currently for sous vide cooking. It can withstand a variety of temperatures and, when used in conjunction with a vacuum sealer, it offers better surface contact and prevent bags from floating (17). They are an inexpensive

option to use for sous vide cooking. Using glass and silicone are an environmentally friendly alternative to plastic. Glass and silicone are durable and reusable substitute. Glass and silicone are known to be a good insulator of heat, but not a good conductor of heat (18). This may result in longer cooking times to be able to reach desired temperatures.

#### ***Pathogens associated with Gravy***

Gravy is a sauce made from cooked meat juices combined with other ingredients. Some common ingredients include wine, broth, cream, herbs and thickened with flour or cornstarch. (19) *Clostridium perfringens* is a common pathogen associated with gravy. *Clostridium perfringens* is usually an outcome of food that has been temperature abused. If food is kept in the danger zone between 4°C and 60°C, it is likely to grow *Clostridium perfringens* bacteria. (20) There was a reported outbreak in Greece linked to under heated gravy causing 800 to 1000 people to be ill. Laboratory test confirmed the pathogen *Clostridium perfringens*. (21) Other pathogens associated with gravy include *Campylobacter*, *Salmonella*, and *Bacillus cereus* (22)

#### ***Regulations and Guidelines***

Currently there are no regulations regarding sous vide cooking that establishments are required to abide to. There are guidelines developed by BCCDC regarding the use of

sous vide cooking in restaurant establishments (1). These guidelines are provided as a resource to chefs and home cooks that utilize sous vide as a cooking method and are not enforceable. However, according to the Public Health Act's Section 15, a person must not willingly cause a health hazard and as per Public Health Act: Food Premises Regulation Sect 14(1) operators must ensure that food on the premise is prepared in a safe manner to eat (23). Therefore, chefs and operators are responsible for using safe sous vide practices in establishments.

### ***Purpose of the Study***

The purpose of this study is to assess the impact of using different mediums of plastic, glass and silicone in a sous vide water bath. This will be determined by examining the time it takes to reach 56.5°C with sous vide cooking of gravy using three mediums: plastic, glass, and silicone.

### **Methods**

#### ***Sous vide – Immersion Circulator***



**Figure 2. 18 Liter Polycarbonate Container with Custom Cut lid (24)**

The brand of immersion circulator used for sous vide cooking was the Anova Precision Cooker. The study procedure involved the immersion circulator being clamped to a sous vide container filled with 16 L of water and set to 56.5°C, according to the manufacturer's manual. A thermocouple probe was submerged into the water to ensure that the Anova Precision Cooker heated the water bath to 56.5°C to ensure precision.

### ***ACR SmartButtons and Trend Reader***



**Figure 3. ACR Smart Buttons, Accessories, and TrendReader Software (25)**

ACR SmartButtons data loggers were used to examine the temperature of gravy sauce within each medium. Trend Reader software was installed onto a personal laptop according to the manual. The SmartButtons were programmed using the software. For calibration, SmartButtons were immersed into ice bath and into a hot water bath measured at 56.5°C with a calibrated thermocouple. The data readings of the thermocouple and SmartButtons were compared to ensure that temperatures were within  $\pm 1^\circ\text{C}$  of each other. Each SmartButton was programmed 15 minutes

before the start of the experiment and the data collection was set to 1 minute intervals

### ***Thermocouple***



**Figure 4. Digi-Sense Thermocouple (26)**

The thermocouple thermometer used to calibrate the immersion circulator and ACR SmartButtons was the brand Digi-Sense DualogR. This thermometer was calibrated through ice bath and hot water bath to ensure measurements were accurate within  $\pm 1^{\circ}\text{C}$  increments.



**Figure 5. Mediums: FoodSaver Polyethylene Plastic Sous Vide Bags, 500 mL Bernardin Glass Jars and Two-Piece Snap Back Lids, and Kepeak Reusable Silicone Food Storage Bags (27)( 28)(29)**

### ***Mediums***

The following mediums that were analyzed: plastic, silicone, and glass. The plastic sous vide bags from the brand FoodSaver are constructed of polyethylene (29). They are suitable for a wide range of temperatures and, when used in conjunction with a vacuum sealer, offer the best performance (8). Silicone bags provide a variety of uses,

including as an alternative to plastic sous vide bags (30). With the use of food grade silicone, there are no harmful byproducts or chemicals (30). The glass medium that was used was the 500 mL Bernardin jars and two-piece snap lids (27). Silicone and glass were tested because they provide an environmentally friendly alternative to polyethylene bags for sous vide cooking.



**Figure 6. FoodSaver FM5400 2-in-1 (31)**

### ***Preparation of the Gravy***

A large stainless pot was used to hold six liters of water. It was brought to a boil and the Trio gravy mix was added to the boiling water. The heat was reduced to low and the sauce was beaten with a whisk to break up the clumps. The sauce was allowed to thicken approximately twenty minutes before it was removed from heat. This procedure of preparing the gravy was repeated five times. The sauce was transferred into the 500 mL Bernardin mason jar to the lower neck through a funnel. This represented the total volume of gravy that went into the three mediums. The 500 mL Bernardin jar with the gravy was transferred to eighteen silicone bags and eighteen plastic sous vide bags.

Eighteen 500 mL Bernandin mason jars were filled. There were eighteen of each medium. All mediums were stored in the refrigerator with a temperature of 4°C, until the gravy within each medium reached 4°C measured with a thermometer.

### **Experimental Method**



**Figure 7. Kepeak Reusable Silicone Food Storage Bags with arrow showing the 250 mL line where the SmartButtons were entered**

All mediums were removed from the 4°C fridge. A programmed SmartButton was inserted approximately at the 250-mL line marked on the outside of the bag into the middle of mixture using forceps (Figure 4). The SmartButtons were inserted into the plastic medium by using forceps and the placement of the devices were determined by what the experimenter deemed to be the middle of the mixture. The plastic sous vide bags were sealed using the FoodSaver FM5400 2-in-1 food preservation system (31). The vacuum sealing of the gravy prevents air bubbles from developing during the heating process, which would cause the package to float. The SmartButtons for the glass medium were required to be suspended

in the middle of the glass medium to acquire an accurate reading. This was done with cheese cloth wrapped around the SmartButton, suspended by string. The string was attached to the lid of the mason jar with a glue gun. All mediums were transferred into the cooler to ensure that they were at the same temperature before the start of the experiment. Once the sous vide reached 56.5°C, four of each medium was added to the hot water bath. The mediums were arranged randomly in the hot water bath spaced out from each medium and submerged (Figure 8). An important note is not to block the immersion circulator, this may reduce its effectiveness to disperse the heat in the water bath. After 150 minutes, all the mediums were removed. This procedure was repeated three more times. A total of eighteen runs were conducted with each medium, with two of those runs being conducted in the pilot study.



**Figure 8. Arrangement of mediums in hot water bath in first run**

An alternative method to this experiment would be to use a sous vide convection steam

oven instead of a water bath, but due to the financial restraints of this project and resources this option was not available. The sous vide water bath method was selected for methods as BCCDC supplied the equipment, helping to reduce financial limitations.

### ***Reliability and Validity of Measures***

The methodology of this project follows the BCCDC Guidelines for Sous Vide Cooking and protocols used by previous BCIT students who have completed sous vide projects (1)(8)(32)(9). These protocols from the guidelines have been developed in collaboration with chefs working in the industry and researchers at BCCDC. By following these procedures, it increases validity. To increase the reliability of the experiment, multiple runs of each medium were done by the same researcher using the same protocols used. Lastly to further enhance reliability and validity of measures, a pilot test was conducted. Even though the sous vide set up is the same as previous experiments, gravy and alternative mediums to plastic sous vide bags have not been studied (8)(32)(9). To increase the reliability and validity to the alternative mediums and gravy, instructions of how to sous vide liquids and the use of silicone and sous vide

cooking were studied and followed (33)(34)(35).

### ***Inclusion and Exclusions***

The inclusion criteria for the mediums used include: FoodSaver polyethylene plastic bags, 500 mL Bernandin glass jars and two-piece snap lids, and Kepeak Reusable Silicone Food Storage Bags. The gravy mix brand was Trio Brown Gravy Mix, 777 g purchased from Costco, no other brand of gravy maybe used for this experiment. The Dritz 606 cheesecloth - 36" X 6 yd and Norpro cotton twine was used to suspend the SmartButtons in the glass medium. The ACR SmartButtons and TrendReader Software were used to record the temperature of the gravy. The equipment set up for this study includes the immersion circulator; Anova Culinary Bluetooth Sous Vide Cooker 800 Watts in the color black, 18 L Polycarbonate container with custom cut lid, and the FoodSaver FM5400 2 in 1 vacuum sealer.

### ***Data Collection***

SmartButtons were removed from each sample after the sous vide process and data was backed up on the TrendReader software. The times and temperatures of each medium was recorded to determine how long it took to reach 56.5°C and used in both the descriptive and inferential statistical analysis.

### ***Statistics***

In this study, numeric continuous data of time (min) and temperature (°C) were collected for each sample. The data was extracted from the ACR smart buttons through the software and onto an Excel spreadsheet. The data was grouped by each medium into their own Excel spread sheets. Each run was analyzed to determine at how long it took to reach 56.5°C for each medium. That provided data points for each medium. The minimum time for each run for each medium was transferred

from Excel to NCSS 12 Data Analysis software (36). The statistical packages used for analyzing the data was the NCSS 12 Data Analysis software (36). The Descriptive statistics is presented in the Table 1. below for each of the mediums over the eighteen runs from data collected. The statistical test used was a One-Way Analysis of Variance (ANOVA) to analyze the if there are any statistically significant differences between the three mediums. Scheffe's test was ran to compare the means of each of the mediums.

**Table 1. Descriptive statistics for time to reach 56.5°C in sous vide cooking of gravy in the three mediums.**

	<b>Plastic</b>	<b>Silicone</b>	<b>Glass</b>
<b>Mean</b>	65	68	129
<b>Median</b>	69	67	127
<b>Min Temp</b>	33	36	106
<b>Standard Deviation</b>	17.7	18.7	13.0
<b>Count</b>	18	18	18

**Table 2. The null and alternate hypotheses of the experiment**

<b>Ho:</b> There is no difference in time of sous vide cooking for gravy at 56.5°C between the mediums: plastic, glass, silicone.	<b>Ha:</b> There is a difference in time of sous vide cooking for gravy at 56.5°C between the mediums: plastic, glass, silicone.
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### ***Interpretation of the Data***

The normality and equal variance is not rejected. Therefore, the test looked at is the Analysis of Variance Table and F-Test. The p-value was 0.00, therefore rejecting Ho and accepting there is a difference in time of sous vide cooking gravy at 56.5°C between the different mediums: plastic, glass, silicone. The power at  $\alpha=0.05$  is 1.0, showing no beta-errors and the results are true. Looking at the Scheffe's Multiple-Comparison Test, it showed that there was a difference between glass and silicone, and glass and plastic.

There was no significance between plastic and silicone medium.

### **Discussion**

Based on the results of this study, there is a difference in time of sous vide cooking of gravy to reach 56.5°C between the mediums: plastic, silicone, and glass. The mean time for each medium to reach 56.5°C differed; 65 minutes for plastic, 68 minutes for silicone, and 129 minutes for glass. From the Scheffe's Multiple Comparison Test, there is

no difference between plastic and silicone, but there is a difference with both plastic and silicone compared to glass. With the statistics showing the results of this experiment valid, there were some methodological limitations that are later addressed in this paper.

With plastic and silicone medium, the shape of the pouches is flexible allowing a larger surface area for heat to be distributed. The glass medium is solid structure making it harder for the heat to penetrate the internal mixture of the gravy. Glass is considered an insulator rather than a conductor, it doesn't convey heat very quickly (33). This helps explain why the mean time of the glass medium is almost double that of plastic and silicone. There is no previous research studying the different mediums in sous vide cooking, the results. Methods such as silicone pouches and glass jars are a new concept to the sous vide community and therefore their heat transfer and cooking capabilities need to be explored.

### ***Placement of Smart Buttons***

The smart buttons for plastic and silicone were both inserted into the middle of the mediums with nothing holding them in place. For the glass medium, the smart button was held in place in the centre of the glass jar. This may have affected accuracy of results for plastic and silicone mediums. With nothing holding the smart buttons in place, it may have been exposed to areas that reached 56.5°C first rather than if it was centralized.

### ***Water Bath Depression***

When initially introducing four of each medium into the hot water bath, it caused a drop-in temperature of 4°C-7°C, requiring 18 to 23 minutes to recover back up to 56.5°C. This confirms previous studies that have shown fluctuation in temperatures when refrigerated foods were introduced to the hot water bath, requiring an extension in time for food to be thermalized (37)(8)(38)(9). The factors that contribute to the hot water bath depression and recovery time include: the amount of refrigerated product added, volume of water in the container, overcrowded conditions, and the type of sous vide equipment (38) (37) (9). The time required for the water bath to be thermalized, extended the time of each medium to reach 56.5°C. Those that choose to use alternative mediums and do not account for this factor may not reach pasteurization, these foods are still considered raw and pose a potential hazard (1). Therefore, it is important for sous vide users to ensure proper come up time and pasteurization and factor in the appropriate time needed for the type of medium used.

### **Limitations**

#### ***Sample Size***

Due to time constraints, only eighteen samples of each medium were cooked using the sous vide method. It would ideal for each medium to have  $\geq$  thirty samples to achieve a more confident statistical analysis.

### ***Placement of Smart Buttons***

The placement of Smart Buttons for glass was done with the usage of cheese cloth and string. The cheese cloth was found to be flimsy, even though it was still able to hold up the smart button. After removal of smart buttons from glass samples, the cheese cloth was found at a lower position than at the start of the sample. A more suitable substitute is to use high temperature polyester mesh (39). It would provide a more rigid structure to work with possibly increasing precision and accuracy of results with glass mediums.

### ***Experiment Setting***

The experimental runs were carried out throughout the day over a two-day period in a household kitchen. Factors that were not controlled for were relative humidity and ambient temperature, which could have affected results. A domestic refrigerator was used to chill all samples overnight, which may have caused uneven cooling to 4°C before the start of each experiment. When each medium was removed from the refrigerator and put into the hot water bath, the ambient temperatures varied both days and could have allowed samples to increase in temperature before being placed in the hot water bath.

### ***Viscosity of Gravy***

Viscosity is a strong function temperature (40). The viscosity of each gravy batch was not measured. Each batch was determined by

following instructions of the Trio gravy package (41). Due to a lack of equipment resulting from budget restraints, each batch of gravy may have had a different viscosity. Gravy with higher viscosity was expected to require more time to reach a temperature of 56.5°C and lower viscosity was expected to require less time to reach a temperature of 56.5°C. A solution to this limitation is using a viscometer, which measures the viscosities of liquids (42). Each sample of gravy would be the same viscosity increasing the precision and accuracy of the experiment.

### **Knowledge Translation**

The results obtained from this study can be incorporated into BCCDC's Sous Vide Guidelines to be viewed by the public. EHOs' and operators' can also use the results of this study to enhance their knowledge on sous vide practices through the guidelines. It can expand on how alternative mediums: plastic, silicone, and glass affect sous vide cooking times. With this knowledge, EHOs can also apply it to inspection of restaurants that use alternative mediums and educate operators. Although gravy was used in this study, the results are not limited to sous vide gravy. Food products all have different penetration rates due to varying food consistencies. The results of this study can be applicable in any sous vide processes. If food products do not reach pasteurization in sous vide cooking, this may present a risk to consumers causing foodborne illnesses.

Understanding the effects of using each medium in extension in time, ensures food safety with sous vide cooking. The following results of this experiment can be presented to CIPHI members, BCIT's Environmental Health Program, BCCDC Environmental Health Services and establishments that utilize sous vide cooking.

### **Future Research**

Some potential areas of research that can be further analyzed are as follows:

- Analyzing if using a sous vide convection steam oven instead of an immersion circulator show similar results.
- Surveying how many sous vide users choose to use alternative mediums other than sous vide plastic pouches.
- Analyzing how different viscosities of gravy or other liquid products (eg. hollandaise sauce) affect the time of sous vide cooking.
- Performing the same experiment with a different food item.

### **Conclusions**

According to the results of this study, there is a statistically significant difference in time that it took for gravy to reach 56.5°C between the mediums: plastic, silicone, and glass. This

shows that a more environmentally friendly alternative to plastic sous vide pouches can be used. Silicone pouches show to be the best alternative, least compromising of the come-up and pasteurization time. If sous vide users opt to use glass, the come-up time almost doubles in time. EHOs should become familiarized with different sous vide mediums if they come across it during restaurant inspections and emphasize the importance of allowing enough time for pasteurization to restaurant operators. Inadequate time to allow for pasteurization of food products may pose a risk to public health due to survival of any pathogen.

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### **Competing Interests**

The authors declare that there are no competing interests.

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