The effect of varying temperatures on the denaturation of bromelain as well as the taste of pineapple juice for use in a stable gelatin dessert

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Experiment Objectives:

1. To find the minimum temperature to heat pineapple juice in order to successfully denature bromelain to create a stable gelatin dessert

2. To determine whether there is a significant difference in taste between fresh, fresh heated, and canned pineapple juice

Date of Experiment:
Week 14
4/21/2021

Course Information:
Food Science
Wednesday PM Session.
CULS-2210
College of Food Innovation and Technology
Johnson & Wales University
Professor Seyfarth
Spring 2021

Background Information and Experimental Hypothesis:

Background Information:

Topic Researched:

- What is bromelain?
- How does bromelain affect the stability of gelatin?
- How can bromelain be denatured?
- Describe pasteurization's effect on the taste and nutrition of pineapple juice

Bromelain is a combination of enzymes called proteolytic, or protein digesting enzymes. It is only available in its natural form from pineapples. It is found in both the stem and juice. It breaks down proteins by breaking the internal peptide bonds in the protein. It is readily absorbed by the body and remains fully intact throughout the gastrointestinal system. There are many amazing benefits and healing properties in bromelain. It has anti-inflammatory properties and is often used to treat symptoms of arthritis. It aids in digestion by breaking down proteins to be absorbed by the body. It also is used to treat burns by removing dead and infected tissue and promotes the healing process. It is also believed to help with pain and reduce the risk for cardiovascular disease. (Science Direct)

There are some possible side effects of taking bromelain in large amounts such as nausea, vomiting, diarrhea, and increased menstrual bleeding. It can also interact with different medications such as anticoagulants, amplifies sedatives such as sleep aids and antidepressants, and enhances the absorption of antibiotics. (Pearson 2019)

Gelatin is made from animal collagen, which is the protein that makes up connective tissue, bones, skin, and ligaments. Since Bromelain is a protein-digesting enzyme, it is impossible to create stable gelatin using fresh pineapple. The gelatin would not be able to set and would remain in a liquid state. The bromelain breaks down the gelatin, restricting it from holding a solid, structural form. (Healthline)

Bromelain can be denatured by heat. To be completely denatured, bromelain needs to be heated to 80 degrees celsius, which is 176 degrees Fahrenheit. This is why fresh pineapple juice will only work in gelatin if it is cooked prior to being added. Canned pineapple juice can also be used with gelatin because it has gone through the pasteurization process in order to be canned, which requires significant heat. If fresh pineapple juice is used with gelatin and is not heated at or above 80 degrees celsius (176 degrees Fahrenheit), the final product will not set. Heat is able to deactivate bromelain because it is a proteolytic enzyme. Pineapple juice pasteurization uses a hot-fill process, usually around 92–105 ◦C for 15–30 seconds. Lower temperature treatments were; 75 ◦C for 3 min, 80 ◦C for 2 min, or 85 ◦C for 15 min and stored at surrounding temperatures. These treatments resulted in nonenzymatic browning due to the Maillard reaction and pigment destruction. The quality, taste, and shelf life of untreated juice depend on the raw material and the applied processes. The nonenzymatic browning causes the quality and taste to be determined as too sour, reports of consumers dislike of not enough sugar. This taste happens because of the heat being affected on the pineapple juice, pushing all the sugar mo In pineapple specifically, heat is the only method that can successfully denature bromelain. Bromelain can be neutralized using acid, but the enzyme will still be active and the gelatin will still be able to set.

molecules to be so small, it is not even noticeable on the palette (Leneveu-Jenvrin).

References

Bromelains. Bromelains - an overview | ScienceDirect Topics. (n.d.). https://www.sciencedirect.com/topics/agricultural-and-biological-sciences/bromelains.

Leneveu-Jenvrin, C., Quentin , B., Assemat, S., & Remize, F. (2020). Maintaining Physicochemical, Microbiological, and Sensory Quality of Pineapple Juice (Ananas comosus, Var. 'Queen Victoria') through Mild Heat Treatment. *Processes*. https://doi.org/doi:10.3390/pr8091186

Pearson, O. (2019, December 5). *Foods Containing Bromelain*. LEAFtv. https://www.leaf.tv/6202111/foods-containing-bromelain/.

What Is Jello Made Of? Ingredients and Nutrition. (n.d.). https://www.healthline.com/nutrition/what-is-jello-made-of.

Hypothesis:

The gelatin made with canned pineapple juice will be the most stable since it has gone through the process of pasteurization and heated to very high temperatures in the canning process, but the least preferred flavor of gelatin since it is not fresh. The gelatin made with fresh pineapple juice heated to 166 degrees Fahrenheit will be the least stable and may not set all the way. The gelatin made with fresh pineapple juice heated to 176 degrees Fahrenheit will have the most favorable flavor and will set up successfully since bromelain is denatured at 176 degrees Fahrenheit.

Experimental Design:

Variable:

- *Heat*

Product(s) Prepared:

- *Gelatin made with fresh pineapple juice heated to 166 degrees Fahrenheit*
- *Gelatin made with fresh pineapple juice heated to 176 degrees Fahrenheit*
- *Gelatin made with fresh pineapple juice heated to 186 degrees Fahrenheit*
- *Gelatin made with canned pineapple juice*

Control Product(s)

- Gelatin made with canned pineapple juice

Test Product(s):

- *Gelatin made with fresh pineapple juice heated to 166 degrees Fahrenheit*
- *Gelatin made with fresh pineapple juice heated to 176 degrees Fahrenheit*
- *Gelatin made with fresh pineapple juice heated to 186 degrees Fahrenheit*

Procedure:

Formula(s):

Pineapple Gelatin
Yield: 329g of liquid

Ingredients	***Weight (in gm)***	***% of Formula***
Pineapple juice	285 g	87%
Water, boiled	36 g	11%
Gelatin	8 g	2%

TOTAL Wt: 329 g

Methods of Preparation:

1. Gather all of your ingredients along with a juicer, 4 mixing bowls, 4 small saucepans, 4 whisks, and small plastic cups for your gelatin to set in
2. Use the juicer to juice all of your fresh pineapples
3. Heat 3 different saucepans of fresh pineapple juice to varying temperatures; 166 Fahrenheit, 176 Fahrenheit, and 186 Fahrenheit
4. Boil a separate pot of water to add to your gelatin
5. Add your gelatin to each mixing bowl
6. After the water is boiled, pour it over each of the mixing bowls with gelatin and stir until it is completely dissolved
7. Stir in your pineapple juices (the three heated, fresh pineapple juices as well as canned) into each mixing bowl
8. Pour the mixtures into plastic cups, where they will set up
9. Refrigerate for a few hours, or until the gelatin feels firm and does not stick to your fingers

Procedure:

- Use a juicer to juice all of your fresh pineapples
- Divide the pineapple juice into 4 containers
- Heat 3 of the containers of pineapple juice; one to 166 Fahrenheit, 176 Fahrenheit, and 186 Fahrenheit
- Conduct a blind taste test of the three juices once they have cooled to find whether there is a noticeable difference in taste
- Conduct another blind taste test with fresh pineapple juice, fresh heated pineapple juice to 176 Fahrenheit, and canned pineapple juice to find whether there is a noticeable difference in taste
- Make gelatin with each of the three fresh, heated juices as well as the canned juice
- Conduct a third blind taste test with each of the gelatins you made to find which is the most favorable

Evaluation of Experiments

Results Tables and Graphs:

Gelatin Experiment

Gelatin		*Texture*	*Flavor*	*Did it set?*		
Control Canned pineapple juice		*Firm / chewy*	*Not so fresh*	*Yes, completely*		
Gelatin 166f heated fresh pineapple juice		*Completely liquid*	*Very fresh, tart*	*No*		
Gelatin 176f heated fresh pineapple juice		*Completely liquid*	*Fresh, slightly watered down*	*No*		
Gelatin 186f heated fresh pineapple juice		*Lumpy/soft*	*Fresh, sweet but tart*	*Partially*		

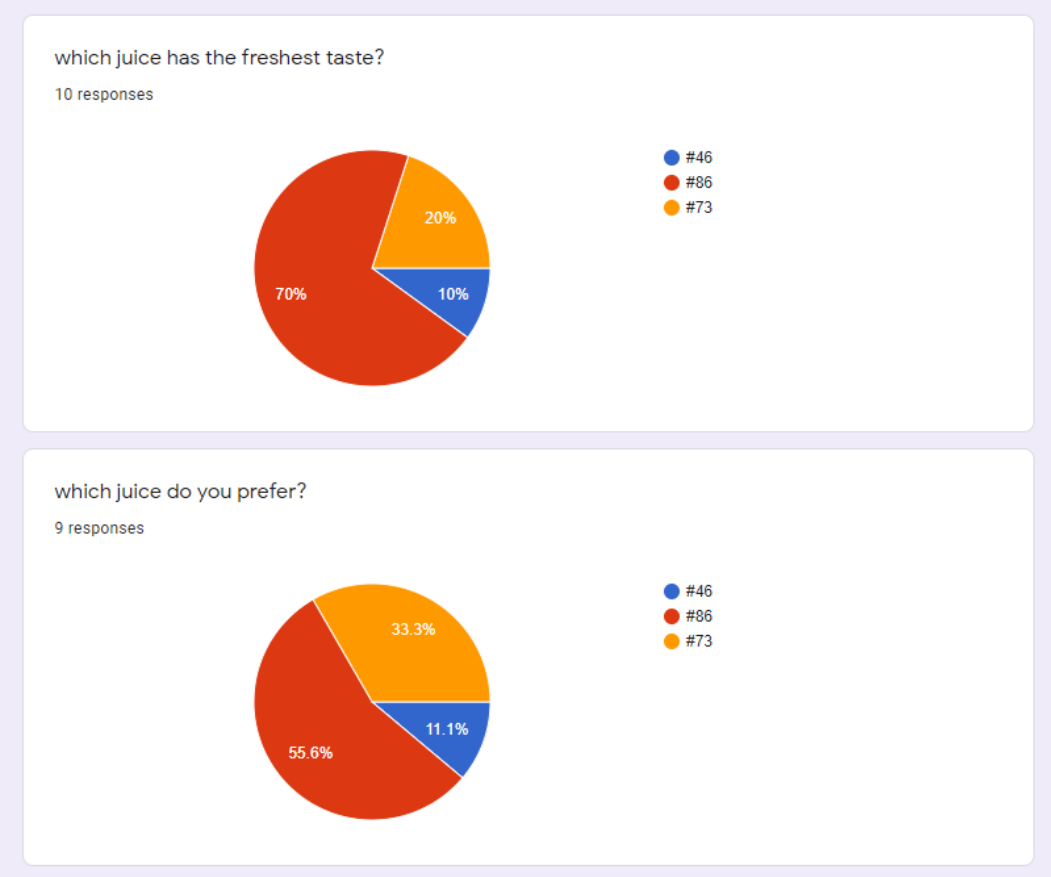
Instrument Test: Brix

Juice	Brix
Canned pineapple juice	13
Fresh pineapple juice	16
Fresh juice heated to 166 degrees F	16.9
Fresh juice heated to 176 degrees F	17

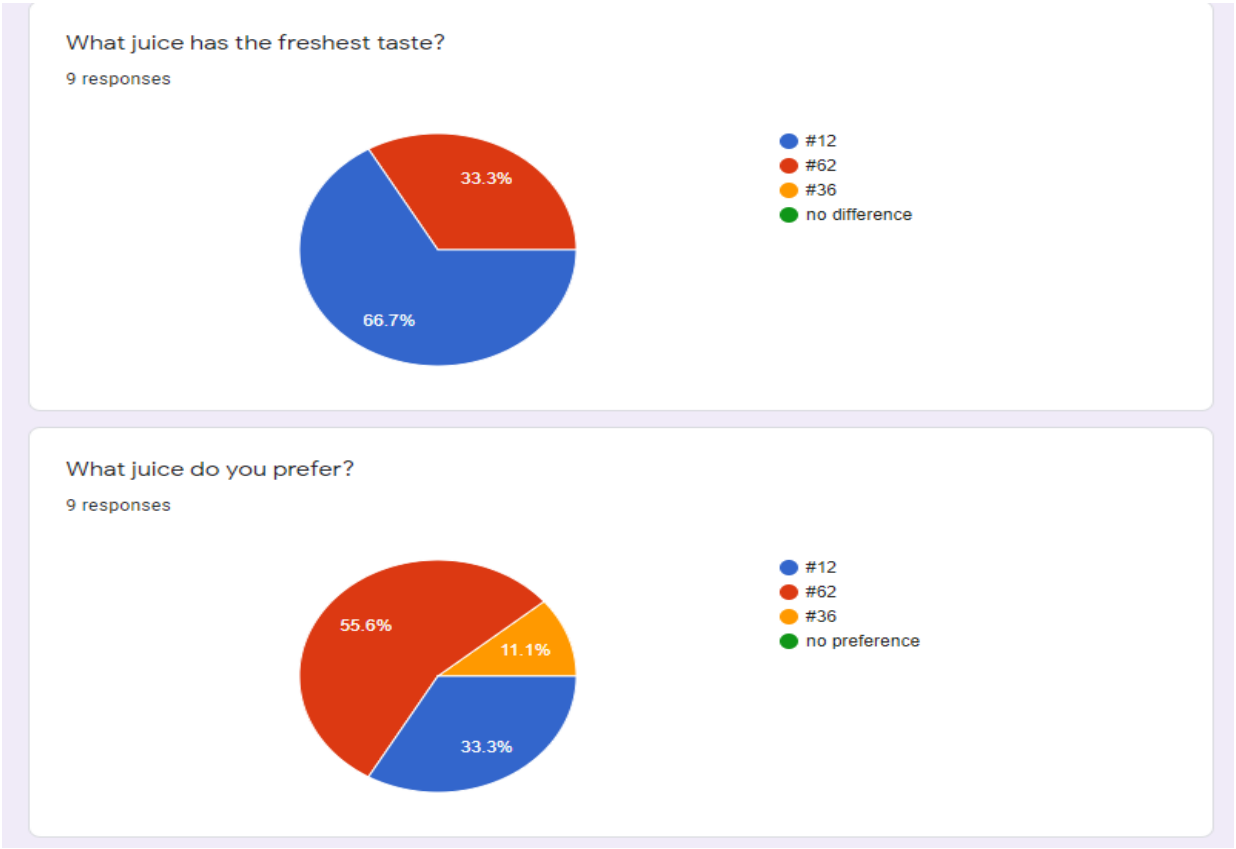
Fresh juice heated to 186 degrees F	17

Taste Test 1 Results:

Commented [1]: Comment under ALLLLLL test results on what the group believes the answers should be based on research, comment what the group truly liked answers, say what the number mean, etc.



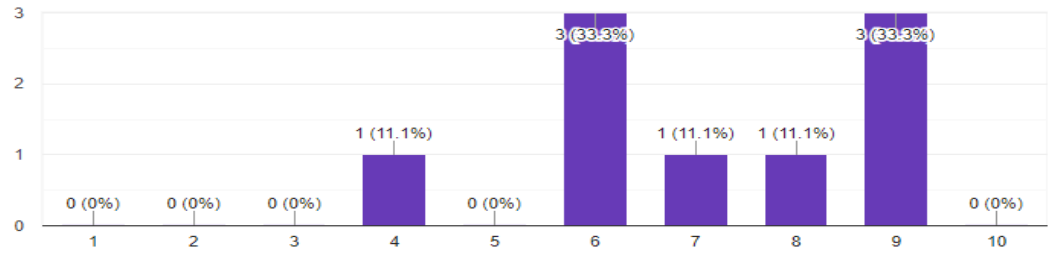
Taste Test 2 Results:



Taste Test 3 Results:

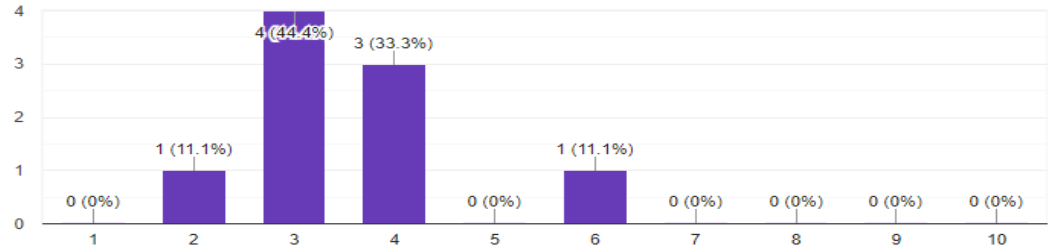
independent of texture, what is your overall favorability of flavor of 34?

9 responses



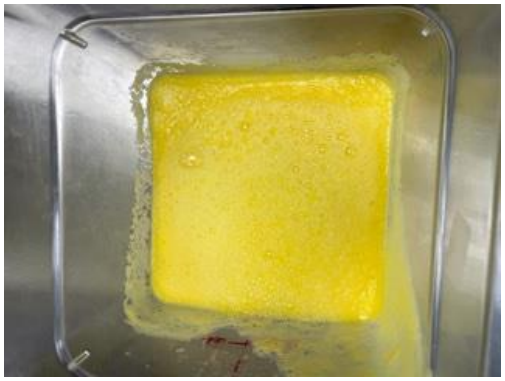
independent of texture, what is your overall favorability of flavor of #2

9 responses



Results Tables and Graphs:

Photos of Results:



Fresh Pineapple Juice from Juicer.

All Fresh Pineapple Juice being Heated to Varying Temperatures (166f, 176f and 186f)

Right off the stove, Fresh Pineapple Juice heated to 166f. Notice more bubbles and foam on top.

The Juices used for the gelatin (canned, fresh heated to 166f, fresh heated to 176f, and fresh heated to 188f).

The first Taste Test consists of canned pineapple juice, fresh pineapple juice and fresh pineapple juice heated to 176f?

The two forms of gelatin that affected the protein (canned and heated to 186f?)

Commented [2]: just want to make sure this is what we tested in Test one.

Commented [3]: yes

Commented [4]: just making sure this is right.

Commented [5]: yes!

The canned pineapple juice gelatin.

186f heated fresh pineapple juice gelatin.

Commented [6]: making sure this is the correct info.

Commented [7]: yes

Descriptions of Experimental Error:

Random:

- During a taste test, an informant notified our group that they did not fill out the questions correctly, and accidentally pressed the wrong answer choice. This is for test one (canned, fresh, heated fresh to 176).
- While the pineapple juice that was heated to 186 degrees was being heated, the thermometer turned off. This could have resulted in incorrect temperatures and incorrect times.

Systematic:

- We did not strain the juices, so the texture could have affected the people's choices during the taste test.

Discussion of Results:

The Brix measurements showed minimal results about the sugar content of each product. The canned juice had the least amount of sugar because, during the canning process, canning companies add water back into the pineapple juice instead of using straight pineapple juice because water is significantly cheaper. The control product (gelatin made with canned pineapple juice) is the only product that is set completely. This was basically guaranteed because the canned juice had gone through the pasteurization process which completely denatures the bromelain. The product made with fresh pineapple juice heated to 186 degrees partially set up but did form complete jello, as it was still a little loose. This means that the bromelain was not completely denatured during the heating process. Both of the other variables (fresh pineapple juice heated to 166 degrees and fresh pineapple juice heated to 176 degrees) remained in the liquid form. This means that the bromelain was either not denatured at all or it was only denatured a little bit. The texture of the final control product was firm and chewy, exactly as a gelatin dessert should be. The texture of the final product of the variable heated to 186 degrees was lumpy and soft, an undesirable texture for a gelatin dessert. The other two variables remained liquid, so the texture was the same as before we refrigerated them. The survey was taken by everyone who remained in class. Out of the fresh pineapple juice, the fresh juice heated to 176 degrees, and the canned juice, the results for the freshest taste unsurprisingly ended up being the fresh pineapple juice that was not heated. The unheated fresh juice was also the most preferred juice. The canned juice tasted the least fresh and was also the least preferred. Out of the three heated juices, the one heated to 166 degrees had the freshest taste, which was also predicted. However, the most preferred juice out of these three ended up being the one heated to 176 degrees. The fresh juice heated to 186 degrees did not appear very enjoyable. Of the two final products that did set (either completely or

partially), the flavor of the partially set gelatin was more favorable than the flavor of the gelatin made with canned juice. The flavor of this was rated significantly higher, which shows that using fresh pineapple juice to make gelatin dessert might be beneficial in some situations.

Conclusion:

Since most of our products did not set, we conclude that if using fresh pineapple juice, it is best to heat the fresh juice to a higher temperature than 186 degrees to make sure the bromelain is completely denatured. The safest route to go when making pineapple gelatin would be to use canned pineapple juice because it will definitely set. Therefore, the first part of our hypothesis was correct, as the gelatin made with canned juice was the most stable and the gelatin made with fresh juice that was heated to 166 degrees was also the least stable. However, the rest of our hypothesis was refuted because the gelatin made with fresh juice that was heated to 176 did not set up at all and it did not have the most favorable flavor. For a future experiment like this, we would suggest cooking the juices to higher temperatures, such as 186 degrees, 196 degrees, and 206 degrees to see the lowest temperature at which the bromelain will be completely denatured instead of partially denatured. We would also like to test whether holding the juices at varying temperatures for longer periods of time such as thirty seconds to one minute instead of turning off the heat as soon as it reaches the given temperature would make a difference. The flavor of the partially set gelatin made with fresh pineapple juice heated to 186 degrees was rated significantly higher than the control made with canned juice, so using fresh pineapple juice does impact the flavor of the gelatin dessert. However, using fresh pineapple juice might not be worth it considering the fact that the canned juice is guaranteed to set and make a gelatin dessert. This would also depend on the cost and the purpose of the gelatin dessert.