DEVELOPMENT OF VEGAN MOZZARELLA-LIKE CHEESE WITH EXTRUDED FAVA PROTEIN

A Thesis

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by

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ABSTRACT

Fava protein-based cheese prototypes, mimicking properties of conventional Mozzarella cheese, were prepared with extruded and unextruded fava protein and studied to find out if extrusion processing could improve the functionality of fava protein isolate. Both fava samples were prepared with 10% fava protein isolate and analyzed for their stretchability and meltability profiles. Part-skim low moisture Mozzarella from local grocery store and Follow Your Heart (FYH) dairy-free shredded Mozzarella were also analyzed for comparison. Results indicated that part-skim Mozzarella cheese has the best stretch and melt among all samples. The extruded fava sample scored higher in both the stretch and melt tests compared to FYH Mozzarella; it also performed better in stretch test compared to unextruded fava sample. Even though the results from texture analyzer suggested that the textural properties of part-skim low moisture Mozzarella and extruded fava sample were different when heated, extruded fava sample was able to achieve a desirable stretching property. Overall, the extruded fava-based Mozzarella prototype demonstrated that the idea of functionalizing fava through extrusion is promising and offers a potential option for generating sustainable dairy alternatives with stretching and melting properties that consumers desire.

BIOGRAPHICAL SKETCH

Chen Zeng is currently in his 2nd year of study in Food Science Program at Cornell University. He is a member of the Rizvi Lab in the Food Science Department. He will graduate in December 2021 with a Master of Food Science degree. He received a Bachelor of Science degree in Food Science from University of California, Davis. He is interested in plant-based food and development of novel and sustainable food products.

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Abbreviation	Meaning
FYH	Follow Your Heart
wt%	weight percentage
fava 90	90% fava protein isolate

1. Introduction

The plant-based food industry has grown significantly in recent years because of the increasing demand for more diverse product lines. Along with the growth in demand, consumers are also having higher expectations on these vegan products, especially meat and dairy analog. While many meat analogs are able to recreate the chewy and fibrous meat texture relatively easily, the texture of dairy analog, especially cheese analog, is still to be desired and has space for improvement. An important aspect of why people consume meat and dairy products is the need for nutrition intake. Meat and dairy products are good protein sources and can easily fulfill one's daily protein requirement. Their alternatives, however, do not have the same kind of nutritional profile. Meat alternatives can simply add plant proteins to achieve a higher protein profile, but it is a different story for dairy alternatives. Currently, most of the available vegan cheese products sold in the grocery store have 0 gram of protein per serving. The reason is that plant proteins do not process the same kind of functionality as casein found in milk, leading to the use of modified starch to mimic the stretching and melting qualities of casein in these cheese alternatives. While some nut-based vegan cheese products have 2 to 3 grams of protein per serving, they are also unable to provide the same kind of stretching and melting properties as the dairy cheese and they are not suitable for people with nut allergy.

The objective of this project is to develop a vegan Mozzarella cheese product with plant protein, providing good stretchability and meltability at the same time. Fava bean protein is the choice of protein for the study because it is a widely grown legume with better sustainability compared to proteins like soya (Tsolakis et.al, 2019). Similar to other plant proteins, fava bean protein is a globular protein, and it does not possess the ability to stretch and melt like casein protein on its own. Therefore, some types of modifications are needed to improve the functionality of fava protein. Currently, common strategies for modifying plant protein include the application of physical, chemical, and enzymatic processing. As consumers are demanding more clean labeled and chemical-free products, physical processing methods offer the option that best fulfills these requirements. Extrusion processing is one of the various physical processing technologies that can be applied to fava bean protein. Because of the high shear and temperature conditions throughout the extrusion process, proteins are subjected to denaturation and their hydrophobic residues are exposed (Akharume et al., 2021). This process also weakens the tertiary and quaternary structures of the native proteins that were once stable and it enables protein molecules to unfold and align themselves towards the flow within the extruder (Camire, 1991). The altered orientation of proteins and changes of hydrophobicity during this process could help to improve protein functionality. Since fava bean protein was chosen for this study, extrusion processing was used for the treatment of fava bean protein to determine if extrusion improves the overall functionality of fava bean protein, allowing more protein to be accommodated in the formula while maintaining good stretch and melt. This study was conducted to evaluate if extrusion processing can bring about potential improvement in the texture of fava bean protein-based cheese in terms of stretchability and meltability.

2. Materials and Methods

2.1. Materials

All ingredients needed for cheesemaking were obtained within the lab. Follow Your Heart (FYH) dairy-free shredded Mozzarella and part-skim low moisture Mozzarella block were bought from local grocery store; they were used for comparative analysis.

2.2. Fava-based Mozzarella cheese formulation

Water is the main ingredient in both dairy cheese and vegan cheese. A high water weight percentage (wt%) helps to better emulsify the protein and starch and it also improves the meltability of the end product but too much water would result in a paste-like consistency with decreased shreddability and stretchability. USDA specifies the moisture content of low-moisture part-skim Mozzarella cheese should be more than 45% but not more than 52% (USDA, 2012). Considering the difference between milk protein and plant proteins as well as the presence of starch in fava cheese formulation, the ideal %wt in fava cheese ranges between 40% to 55%. In this study, 43.7% of water is chosen for both extruded sample and control sample. Oil is also an important ingredient that is responsible for the meltability of cheese. A 19 to 25 wt% of oil is ideal for meltabilty and stretchability. Even though extrusion is used as a pre-treatment method of the fava protein to improve the functionality, starch is still a required component for the stretchiness in the fava cheese formulation. Among all the starches, modified potato starch provides the best texture and consistency. 15 to 20 wt% of modified potato starch in the formulation delivers ideal shreddability, meltability and stretchability. Tapioca starch offers similar melting properties but is lacking in shredding and stretching properties. Maltodextrin is a polysaccharide that is usually used to improve the flavor, thickness, or shelf life of a product; the addition of maltodextrin in formulations shows improvement on the stretch quality of the fava cheese samples at concentrations of 3% to 6%. For the protein source, fava 90 was used in the formulation with wt% at around 10% to achieve the best stretchability. Extruded treated fava 90 was dried thoroughly in a dehydrator and ground to 150 to 450 μm in particle size before cheesemaking and extruded fava 90 was used as received. A detailed breakdown of each ingredient used for both the extruded and unextruded formula could be found in Table 1. The extrusion conditions used for the treatment of fava 90 can be found in Table 2.

Table 1
Composition of 10% extruded fava 90 and 10% unextruded fava 90.

Ingredient	Weight Percentage %
Water	43.7
Cellulose Nanofibril	3.5
Coconut oil	20.7
Lecithin	0.3
Protein	9.9
Modified Potato Starch	16.6
Sodium Citrate	0.25
Sodium Bicarbonate	0.25
Glycerin	1.5
Lactic Acid	0.25
Maltodextrin	3.2

Table 2
Extrusion conditions of 90% fava protein isolate.

Parameter	Liquid Flow Rate (kg/h)	Feed Rate (kg/h)	CO2 Flow Rate (kg/h)	Knife Speed (rpm)	Screw Speed (rpm)	Head pressure (psi)	Product temperature (˚C)
	9-10	30	0	595	107	1800	75

All fava cheese samples are prepared with Thermomix. Water was added to Thermomix and heated to 110 °F first. After that, dry powder mixture (protein, salts, sodium bicarbonate and modified potato starch) were added and stirred for 4 to 5 min on medium speed. Melted coconut oil was added then; temperature and speed setting of the Thermomix was increased to 150 °F and high. After the mixture was stirred for 2 to 3 min, cellulose nanofibril, glycerin and lactic acid were added with temperature increased to 185 °F and was continually stirred for 2 to 3 min. The final product was kneaded with a spoon for a minute before storage. Samples were then placed in plastic containers and refrigerated for at least 24 h to rest before testing.

2.3. Texture analysis

A Model TA.XT*plus* texture analyzer (Stable Micro Systems, Texture Technologies Corp., Surrey, UK) affixed with a 50 kg load cell was used to perform texture analysis on the different cheese samples. To prepare samples, hanger was placed inside the metal baseplate first and each baseplate was filled with about 10 g shredded of cheese sample. Samples were then heated for 6 min ± 30 sec until melted in a 220 °C convection oven with a glass jar covering the plate to prevent excessive moisture loss during heating. After heating, the samples were carried to the testing room with around 1 to 2 min to cool down. Cheese temperature while bubbling directly out of the oven was 105 ± 5 °C and it reduced to 65 ± 5 °C when the ring was inserted into cheese, reducing cheese temperature to 65 ± 5 °C. Samples were analyzed with the hanger traveling at a fixed speed of 30 mm/s to the height of 250 mm.

2.4. Cheese stretch test

Stretch test was performed on all cheese samples using the texture analyzer. Region A did not have any resistance as the hook was moving up to the hanger. When the hook was in contact with the hanger as it moved up, the bulk yield of cheese samples would be observed in elastic deformation as Region B had shown. After the veil point, the stretch began to break and strands would form and stretch in Region C. A plateau-like bulk yield would be indicative of a colder solid-like cheese, whereas an exponentially decaying bulk yield would be indicative of a hotter liquid-like cheese. In Region D, the strands formed were stretch, neck and break. Even the last strand was broken, a force was still observed on the graph as weight of the attached cheese would exert a nonzero ‘force’ on the hook. Also, as Region D was where strands broke, ideally the stretch distance could be observed on the graph. As Mozzarella strands breaking holds significant force with violent tears and snaps, it would be possible to identify the distance accordingly. However, it is not possible for vegan cheeses to have a breaking force above the noise threshold as they flow when breaking, creating a smooth transition. Therefore, a new regime of detecting the stretch distance was needed. Currently, the stretch distance was determined by acquiring the time spent when the last strand break visually which can be used to determine the distance hook traveled before reaching the strand breaking point. While this was not a very precise measurement, it provided a quantitative measurement of the stretch. In the future, adding an extended height instrument might be able to provide more accurate and precise readings of the stretch distance.

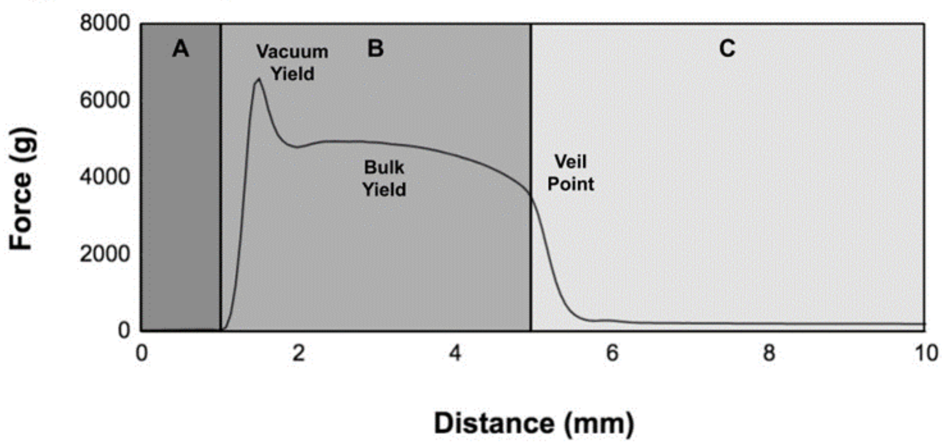

Figure. 1. Typical force-distance diagram of commercially available part-skim low moisture Mozzarella cheese at short distance.

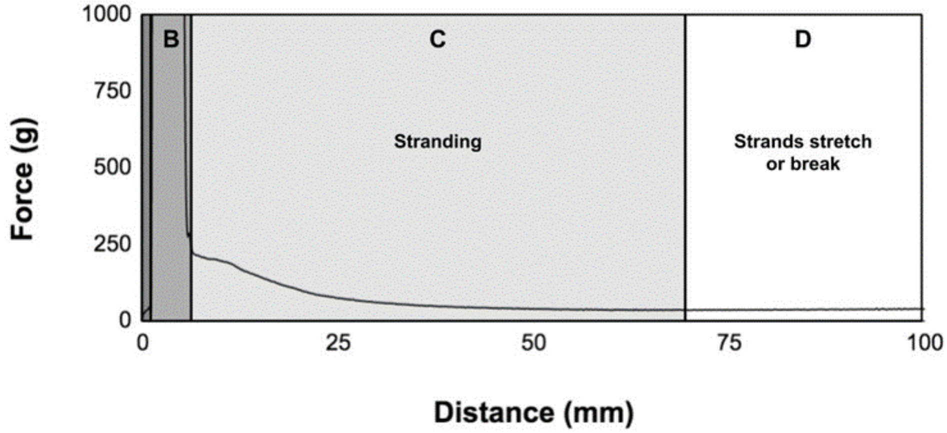

Figure. 2. Typical force-distance diagram of part-skim low moisture Mozzarella cheese at medium distance.

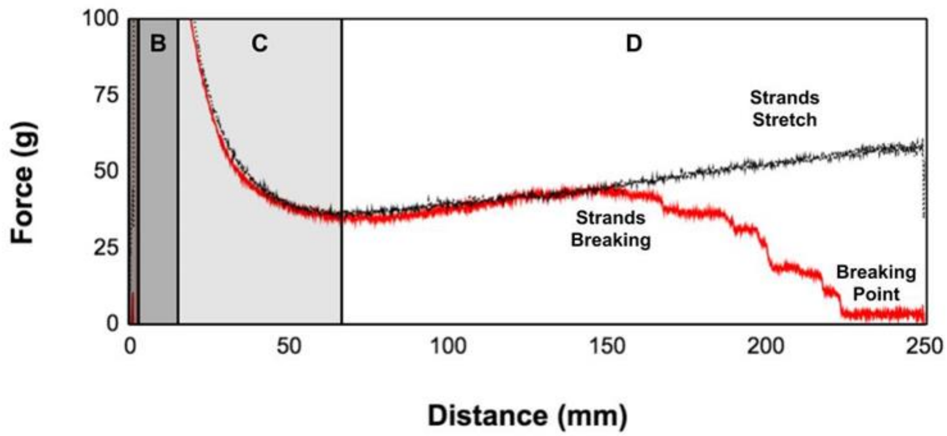

Figure. 3. Typical force diagram of part-skim low moisture Mozzarella cheese at full distance.

2.5. Cheese melt test

Melt test of samples was performed by measuring out about 3 grams of shredded cheese and fitting the samples in a metal ring (40 mm) with a watch glass at the bottom so that the samples were 40 mm in width and 0.5 mm in height. A measurement ring was used to provide a quantitative reading of the spread distance. It consists of a set of rings with different diameters. The smallest ring is 40 mm in diameter and each ring is 2 mm apart.

Readings were generated by measuring the intervals each sample spreads. To start the test, the metal ring was aligned with the inner ring of the measurement ring to make sure all samples were within the smallest ring before testing. After that, the metal ring was removed and the watch glass was covered before heating in the oven to prevent excessive moisture loss.

3. Results and Discussion

3.1. Stretchability Profile

All samples were prepared in the same way and were repeated four times to get an average value. A 10-g sample was shredded and heated with a covered glass jar for 6 to 7 min at 220°C. Distance of stretch was measured for all the samples. Part-skim low moisture Mozzarella displayed the best stretch distance among all the samples, followed by 10% extruded fava 90, FYH Mozzarella and 10% unextruded fava 90. It was worth noticing that as protein content in vegan cheese increased from less than 2% to 10%, the distance of stretch also decreased as more protein was used in place of starch. This general trend was also observed in the FYH sample and fava-based samples even though different types of protein were used. Compared to FYH Mozzarella, 10% unextruded fava 90 sample was about 20 mm shorter in stretchability (Table 3). Even though all dairy-free samples tore during the test and none of them had reached 200 mm as commercial Mozzarella did, these preliminary results indicated that extrusion did improve the stretchability of the fava-based cheese.

Table 3
Stretchability of conventional Mozzarella cheese and plant-based Mozzarella cheese samples.

Sample	**Distance of stretch(mm)**
Part-skim low moisture Mozzarella	>200.0
Follow Your Heart dairy-free shredded Mozzarella	99.3 ± 10.1
10% unextruded fava 90	79.4 ± 5.2
10% extruded fava 90	106.2 ± 6.7

To better understand the texture, data from texture analyzer readings were used to generate force-distance graphs for all samples. Commercial Mozzarella required the highest force during stretchability measurement, indicating a more rigid texture compared to the more flowing texture found in vegan samples. Interestingly, 10% extruded fava 90 was observed to require the lowest force while displaying the most stretch before breaking. This result suggested that high force did not translate to good stretching ability. In Mozzarella cheese, casein protein is responsible for the high force and sudden tear of the strands that were observed in the graph. In vegan cheese, as well as fava-based cheese samples, the use of starch and plant protein tend to create a more fluid-like texture upon stretching and melting.

3.2. Meltability Profile

Melt test was performed under the same oven condition as the stretch test, which was at 220°C but only for 5 min as less amount of sample was placed in the plate. Among all cheese samples, part-skim low moisture Mozzarella was also the highest in spread distance (6 mm). Extrusion did show a slight improvement in the melting quality of the cheese with

10% extruded fava 90 showing a better spread compared to 10% unextruded fava 90 (Table 4).

Table 4
Meltability of conventional Mozzarella cheese and plant-based Mozzarella cheese samples.

Sample	Distance of spread(mm)
Part-skim low moisture Mozzarella	6.0 ± 0.0
Follow Your Heart dairy-free shredded Mozzarella	2.0 ± 0.0
10% unextrued fava 90	3.5 ± 0.0
10% extruded fava 90	4.0 ± 0.0

FYH Mozzarella displayed the least amount of melt. This was very likely due to the addition of calcium sulfate which would increase the rigidity of the sample and the calcium content in final product. FYH Mozzarella was observed to have a higher maximum force compared to the two fava samples (Figure 4).

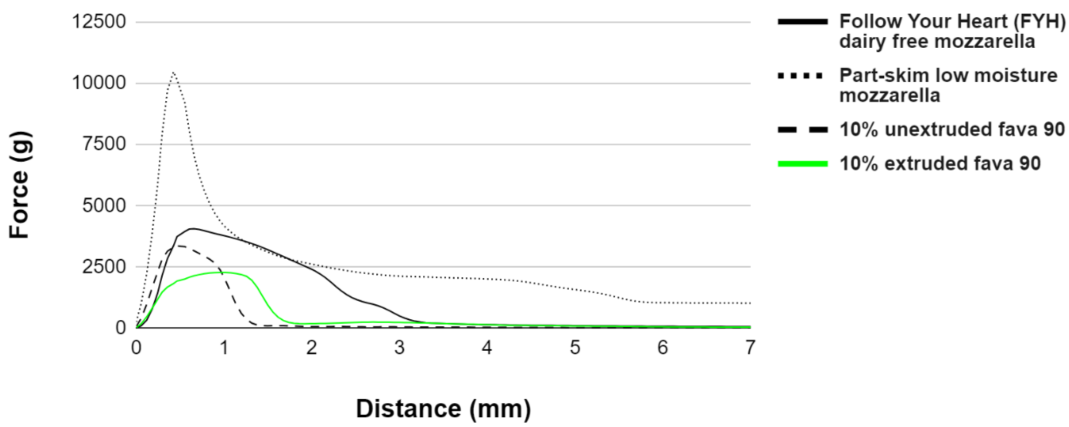

Figure. 4. Force-distance diagram of Follow Your Heart (FYH) dairy-free Mozzarella, part-skim low moisture commercial Mozzarella, 10% unextruded fava 90 and 10% extruded fava 90 at short distance.

While the addition of calcium sulfate better mimics the rigid texture of dairy Mozzarella, it decreases the melting quality of the sample, resulting in a small spread.

4. Conclusion

Overall, the 10% extruded fava 90 sample demonstrated the best results for both the stretchability and meltability among all three plant-based cheese samples that were evaluated, suggesting that extrusion is a promising processing method to improve the functionality of fava protein (Table 3 and Table 4). While the 10% extruded fava 90 sample did not achieve the same stretch and melt as store-bought Mozzarella, the better performance of 10% extruded fava 90 sample compared to FYH Mozzarella makes it a potential direction for an upgrade of commercially available vegan Mozzarella cheese. Despite the promising results from stretch test and melt test, this study did not address much on the evaluation of functionalities of the extruded fava as well as the sensory aspects of the extruded fava cheese samples. Future studies should continue to investigate these two aspects to provide a more conclusive picture of this product and more research is needed to further improve the stretching and melting properties of protein-rich vegan cheese to the next level.

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