Effects of various emulsification agents in fat emulsions for plant-based meat analogs

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Abstract

Over the past 40 years, global meat production has increased four-fold, leading to concerns regarding the environmental toll of increasing production to feed the growing global population, which just hit 8 billion in 2022 and continuing to increase exponentially. The development of plant-based meat can reduce this environmental damage, being 5-10 times more environmentally friendly compared to livestock production, and eliminates the resulting animal welfare concerns from mass livestock production. It has been found that plant-based meats can retain fats more efficiently through the use of oil-in-water (o/w) emulsions, which allows for improved textural properties that can better mimic real meat. Methylcellulose is a commonly used emulsification agent, but its synthetic nature creates negative consumer perception and a need to find a plant-based alternative. The goals of this study were to determine the effects of using various emulsification agents (pea protein isolate, soy protein isolate, guar gum, and methylcellulose) in fat emulsions for use in plant-based meats, and to see if there are any similarities between natural emulsification agents and methylcellulose. It was found in a blind tasting that soy protein isolate was most promising as a replacement for methylcellulose in fat emulsions. The data from this research can be used to further optimize fat emulsions for plant-based meats.

Introduction

The global population just hit 8 billion in 2022 and will continue to increase due to innovations in healthcare (United Nations, 2022); however, this leads to issues regarding the global consumption of meat. In addition to the growing population, the advancement of economically developing countries has also led to global meat production to increase by over four times since 1961, and global per capita consumption has also increased (Ritchie & Roser, 2019). This raises global concerns regarding the expansion of livestock breeding to meet consumer demands.

Animal-based food production currently accounts for about 57% of global greenhouse gas emissions and 80% of global land use in 2013 (Xu et al., 2021; Stehfest et al., 2013), and will continue to grow to meet population demands. In addition, the use of CAFOs (Concentrated Animal Feeding Operations) have allowed for thousands of animals to be confined in small spaces, forced to stand for long periods of time, consume feed, and be injected by steroid hormones that allow them to fatten and grow quickly for efficiency. The use of petroleum-based fertilizers in the rapid growth of animal feed for this large-scale industrial farming also heavily contributes to greenhouse gas emissions (Emmanouilidou, 2022).

Due to these concerns, the development of successful meat analogs is necessary in order to maintain food security and reduce environmental damage. A promising meat analog is plant-based meat. Plant-based products can be 5-10 times more environmentally friendly compared to their animal-based counterparts (Clark et al., 2022). In addition, plant-based meat can eliminate the animal welfare concerns that have risen from the expansion of livestock production.

Plant-based meat is readily available to purchase in the United States, with companies such as Beyond Meat and Impossible Foods having gained success and consumer recognition through their plant-based burgers. Another example is Ikea, which has also released their own "plant balls", a plant-based alternative to their iconic Swedish meatballs. Comminuted meat products, which mimic products such as sausages, meatballs, and patties rather than full cuts of meat, are the most common and focused on in development as the technology involved in mimicking the fibrous structure of a cut of meat still needs development (Ismail et al., 2020).

Plant-based meat typically utilizes textured vegetable proteins, such as soy or wheat gluten as the main ingredient to replace meat protein for texture/mouthfeel, appearance, and nutrition. Coloring agents such as beetroot powder can be used to add a red hue and mimic the myoglobin present in real meat. Spices and flavorings allow for flavor enhancement or masking of unwanted flavors, such as a bean-like flavor that could come from soy proteins. Binding agents, such as wheat gluten, gums, or starches, allow for texture and water binding properties. Finally, fats and oils are used to allow for better texture/mouthfeel, succulence, and also browning of the product when cooked (Bakhsh et al., 2021b).

It has been found that the use of oil-in-water (o/w) emulsions in plant-based meat can allow for increased oil content in high-moisture extrudates, and improved textural properties that can better mimic real meat (Wang et al., 2022). Reports have shown that o/w emulsions have been created using perilla and canola oils and soy protein isolate as a replacement for animal fat in emulsion-type meat products (Utama et al., 2018).

One emulsification agent commonly used in plant-based meat is methylcellulose, a modified polymer created from cellulose. Its amphiphilic properties allow it to create a thermoreversible gel when dissolved in water and gel when heated (Nasatto et al., 2015).

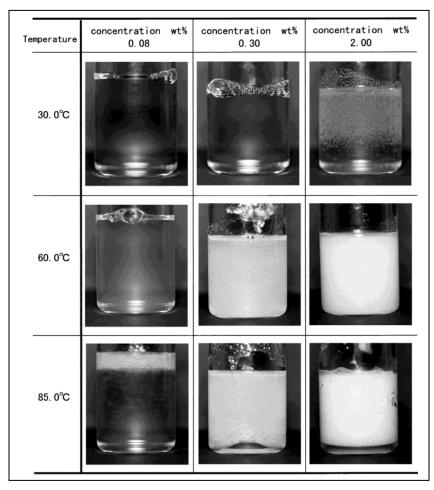


Fig 1: Shows the heat-set gelation property of methylcellulose at different temperatures (Takahashi et al., 2000).

Due to this property, methylcellulose is useful as a binder in plant-based meats, where heat-set gelation, water-binding, and fat-binding are essential properties for mimicking meat and creating desired textural properties. Previous studies have shown methylcellulose to be used successfully as a binding/emulsification agent in both real meat and plant-based meat (Husak et al., 2018), (Bakhsh et al., 2021a). However, methylcellulose's synthetic nature creates a negative consumer perspective around its

use in food products (Varela & Fiszman, 2013). Therefore, it is important to consider other natural, plant-based emulsification agents, such as soy and pea protein isolate.

The goals of this study were to determine the effects of using various emulsification agents (pea protein isolate, soy protein isolate, guar gum, and methylcellulose) in fat emulsions for use in plant-based meats, and to see if there are any similarities between natural emulsification agents and methylcellulose that could potentially replace methylcellulose through a panel. Methylcellulose is expected to stand out compared to the other emulsification agents due to its unique gelling properties.

Methodology

Creating the Fat Emulsions

Using instructions from Kitchen Alchemy's Plant Based Burger recipe, 4 fat emulsions (FEs) were created using proportions of 2:100:300 grams emulsification agent (methylcellulose, pea protein isolate, soy protein isolate, and guar gum), water, and purified coconut oil to compare to beef tallow obtained from a local butcher shop (B&M Meat Market, Park Ridge, NJ) ("Kitchen Alchemy," 2019).

Refined coconut oil (Amazon.com Services, Inc, Seattle, WA) was heated until fully melted. Water and a chosen emulsification agent were placed in a food processor and processed until the mixture began to thicken. The heated coconut oil was then slowly added into the water-emulsification agent mixture and emulsified in a food processor, creating the fat emulsion. The fat emulsion was then poured into a bowl and refrigerated until firm. Once firm, it was scooped out into individual 1 oz plastic sample cups (TashiBox, Pittsfield, MA), with individual samples weighing roughly about 4 grams. This process was done a total of 4 times to create fat emulsions using high viscosity methylcellulose (Modernist Pantry, Eliot, ME), pea protein isolate (138 Foods, Inc, Claremont, CA), soy protein isolate (Bulk Supplements.com, Henderson, NV), and guar gum (138 Foods, Inc, Claremont, CA) as emulsification agents.

Setting up the Triangle Test

Nine, three-digit numbers were generated between 001 and 999 using a random number generator. For each round of the triangle test, participants were instructed to grab three cups from a table, each labeled with different 3-digit numbers so that they can examine and swap them in any order they would like. For the first round, cups with the fat emulsion containing methylcellulose (MC) were labeled as "947". The two other labeled cups were filled with guar gum emulsions (GG1 and GG2), and were labeled as "844" and "398", respectively. For the second round, cups with MC were labeled as "190". The

other two cups were filled with pea protein isolate emulsions (PP1 and PP2), and were labeled as "204" and "561", respectively. For the third round, two cups were filled with methylcellulose emulsions (MC1 and MC2) and were labeled as "621" and "122", respectively. The other batch of cups were filled with soy protein isolate emulsions (SP) and were labeled as "975".

Sensory Testing

A triangle test was used to see if there is an overall difference between methylcellulose-based fat emulsions and other fat emulsions containing either guar gum, pea protein isolate, or soy protein isolate, and to determine if these changes lead to recognizable changes in the fat emulsions. Triangle tests are a type of discrimination test that are used in order to compare two samples for any identifiable differences. They are done by preparing three samples for a participant, two of which are the same and the third being different. The participant would then be instructed to choose the odd sample out of the three (Johnson, n.d.). Data was collected through Google Forms.

Three rounds of triangle tests were used to compare each of the three different emulsification agents (pea protein isolate, soy protein isolate, and guar gum) to methylcellulose. For each test, three groups of cups were presented to the panelists, each labeled with a different randomly generated 3-digit number. Two of the three groups were the same, while one was different. Participants were instructed to choose one cup from each group and were allowed to inspect them in any order to ensure randomization. On the Google Form, participants were instructed to identify the odd sample out of the three and record the 3-digit number for identification. Following this, participants filled out a 6-point likert-scale questionnaire by being prompted with the question and task: "For the next four questions, rank the similarities (if any) of the odd sample to the two alike samples." This was done for visual similarity, flavor, texture, and overall similarity. Lastly, participants were given a text box to describe any other qualitative observations or opinions regarding the sample fat emulsions.

398							
844							
947							
For the next 4 quest samples. Description (optional)	ions, ran	k the sin	nilarities	(if any) o	of the od	d sample	to the two alike
Visual similarity *							
	1	2	3	4	5	6	
No differences	0	0	0	0	0	0	Many differences
Flavor *							
	1	2	3	4	5	6	
No differences	0	0	0	0	0	0	Many differences
Texture *							
	1	2	3	4	5	6	
No differences	0	0	0	0	0	0	Many differences
Overall similarity							
	1	2	3	4	5	6	
No differences	0	0	0	0	0	0	Many differences
Is there anything els	-	uld like t			about the	e sample:	s? (Example: *

Fig 2: Questionnaire participants were given

Additional Information on Human Participants

There were 18 participants in total, 14 were high school students and 4 were high school teachers, mostly Caucasian but with varying demographic backgrounds. No identifiable vulnerable populations were involved in this research. Participants were recruited from the Nyack High School Science Research Program. Identifiable risks included potential allergic reactions to the ingredients in the FEs. Potential allergens included soy, pea, tree nuts, peanuts, wheat, milk, and egg. This risk was minimized by listing these ingredients in the initial permission slips/sign up form and screening participants for allergies prior to the panel. Participants with allergies were instructed to assist in the experiment rather than participate.

Identifiable information of the participants was not collected, and questionnaires were submitted anonymously by participants through a Google Form. Participants were informed about the purpose of the study, what they were going to be asked to do, and that their participation was voluntary and that they had the right to stop at any time through the permission slips, where they were able to view this information and print out informed consent forms to turn in as well as prior to data collection.

Colorimetry

Color Name AR, a colorimeter iPhone application by Vlad Polyanskiy, was used to compare the color of the FEs to the beef tallow. All FEs and beef tallow were placed under controlled lighting to take images of each sample.

Materials-Based Assay

The melting points of all fat emulsions were measured and compared to the melting point of beef tallow, using a ThermPro⁺ Digital Thermometer (ThermPro, Duluth, GA). Each fat emulsion was slowly heated in a glass bowl placed over a pot of boiling water while measuring their temperatures with the thermometer until they started to melt to find the melting points.

Results

Pea Protein Isolate

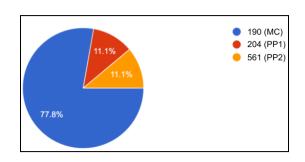
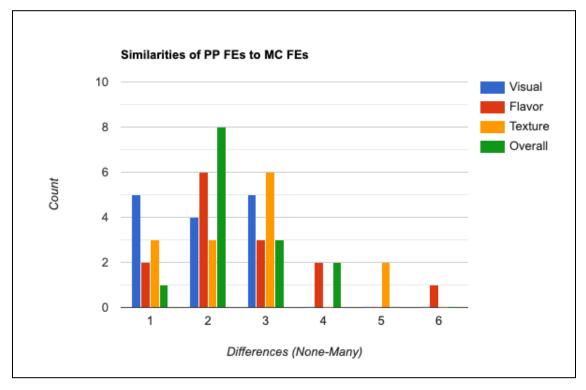


Fig 3: Percent correct identification of MC FEs compared to PP1 and PP2 FEs

Figure 3 illustrates that 77.8% of the 18 participants were able to correctly distinguish the MC FEs to the PP1 and PP2 FEs. The PP FEs are significantly different from the MC FEs (p=0), shown through an upper-tailed p-test.



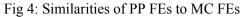


Figure 4 illustrates the perceived differences of the PP FEs to MC FEs.

Soy Protein Isolate

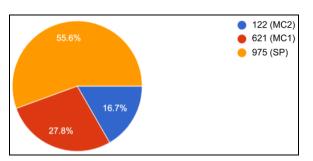


Fig 5: Percent correct identification of SP FEs compared to MC1 and MC2 FEs

Figure 5 illustrates that 55.6% of the 18 participants were able to correctly distinguish the SP FEs to the MC1 and MC2 FEs. The SP FEs are significantly different from the MC FEs (p=0.043), shown through an upper-tailed p-test. The SP FEs are directionally closer to significance compared to the GG FEs or PP FEs.

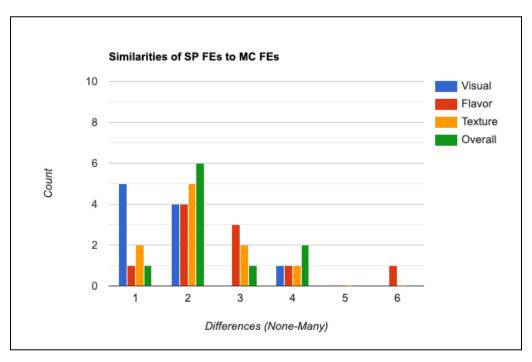


Fig 6: Similarities of SP FEs to MC FEs

Figure 6 shows the perceived differences of the SP FEs to MC FEs.

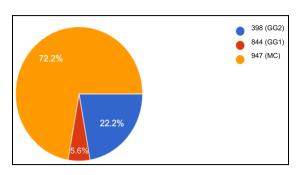


Fig 7: Percent correct identification of MC FEs compared to GG1 and GG2 FEs

Figure 7 shows that 72.2% of the 18 participants were able to correctly distinguish the MC FEs to the GG1 and GG2 FEs. The GG FEs are significantly different from the MC FEs (p=0.001), shown through an upper-tailed p-test.

Guar Gum

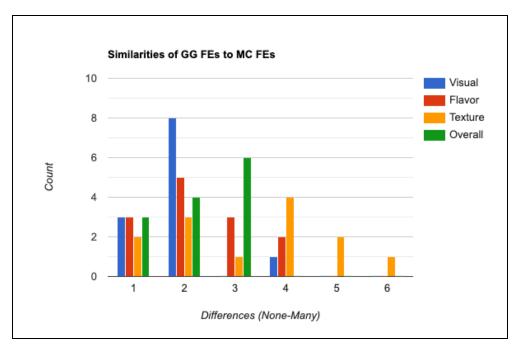


Fig 8: Similarities of GG FEs to MC FEs.

Figure 8 illustrates the perceived differences of the GG FEs to MC FEs.

	Visual	Flavor	Texture	Overall
РР	2.00	2.64	2.64	2.43
SP	1.70	2.80	2.20	2.40
GG	1.77	2.31	3.31	2.23

Table 1: Shows the average values of similarities of PP, SP, and GG FEs to MC FEs. Values were obtained from the Likert-Scale questionnaire, where 1=No differences and 6=Many differences.

Based on Table 1, SP FEs performed best regarding visual similarity and textural similarity, but were more easily distinguished compared to the other FEs. GG FEs had the greatest difference in texture compared to MC FEs.

Qualitative Data

Qualitative data was also collected via Google Forms. Participants were asked to note any other observations or opinions they had for each FE. This will be further discussed in the conclusions.

	Image	Color (HEX Color Code)	Melting Point
Pea Protein Isolate		#D9BFA0	21ºC
Soy Protein Isolate		#D4BA9A	21.2°C
Guar Gum		#DDC3A3	21.5 °C
Methylcellulose		#E2CBB0	21.4 °C
Beef Tallow		#AB8D65	47°C

Colorimetry

Table 2: Displaying images of each fat emulsion and beef tallow and their colors and melting points.

The beef tallow has a much darker color compared to the FEs. The FEs are all similar in color, though the MC FEs appear to be slightly lighter in color compared to the other three FEs.

Materials Based Assay

All of the FEs showed similar melting points, with a mean of 21.3°C. This is much lower compared to the measured melting point of the beef tallow, which was 47°C.

Discussion and Conclusion

The results from this study show that soy protein isolate may potentially be used as a replacement for methylcellulose in fat emulsions for plant-based meat products. The percent number of participants who correctly identified the odd sample for the triangle tests was the least (55.6%) when comparing the SP FEs to MC FEs. This is very different from the percentages for the GG FEs (72.2%) and PP FEs (77.8%), which shows that the SP FEs are similar enough to MC FES where differences are not easily distinguishable. Additionally, SP FEs had the least average differences regarding visuals (1.70) and textures (2.20) compared to the GG (1.77 and 3.31) and PP (2.00 and 2.64) FEs. However, SP FEs had the highest average differences regarding flavors (2.80) compared to the GG (2.31) and PP (2.64). One participant noted that SP FE "had less flavor" compared to the MC FEs, and two participants noted that the SP FE had a slight yellow hue that made it distinguishable from the MC FEs. However, this could be beneficial for use in plant-based meat analogs. Through the colorimetry, it was found that the SP, PP, and GG FEs were all similar in color and were all slightly darker than the MC FE. The beef tallow in comparison was the darkest, so the non-MC FEs have closer colors to beef tallow than the MC FE, making them more suitable for use in plant-based meat analogs than the MC FE.

These results reflect the results of other research done, such as that of replacing animal fat with a perilla-canola o/w emulsion using soy protein isolate in meat emulsions and replacing pork backfat with vegetable oil o/w emulsions using soy protein isolate (Utama et al., 2018) (Asuming-Bediako et al., 2014).

This study can be redone using a larger sample size and different combinations of emulsification agents and fats in order to gain a wider scope of the effects of different emulsification agents or fats in o/w emulsions. However, further research is required to optimize the concentrations of soy protein isolate and to test the fat emulsion in an actual comminuted plant-based meat product.

Fat emulsions are important to PBMAs as they allow for greater textural properties in plant-based meat products compared to using only solid plant-based oils such as coconut oil or palm oil as they can retain more oil/moisture (Wang et al., 2022). In order to boost consumer perception and product value, a replacement for methylcellulose is needed (Varela & Fiszman, 2013). This study has confirmed that plant-based emulsification agents can be a viable alternative to methylcellulose, and continues our goals to create a better plant-based meat alternative.

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