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Development of High Protein Tomato Soup Using Milk Protein Concentrate and Pea Protein Isolate

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This study aimed to explore the development of a high protein tomato soup to address the nutritional needs of older adults who require higher levels of protein compared to the general population. The specific objective of this study was to explore how the addition of a dairy protein (Milk Protein Concentrate - MPC), a plant-based protein (Pea Protein Isolate - PPI), or their combination affected the techno-functional properties and sensory acceptance of tomato soup. A control soup and three soups containing MPC, PPI and their combination were prepared, and their techno-functional properties were tested. A group of 33 naïve consumers older than 18 years, scored overall liking, liking of appearance, taste, aroma and mouthfeel of the soups using 9-point hedonic scales. A significant increase in pH, apparent viscosity, and L\*and b\* values and a decrease in a\*value for colour were observed with the addition of protein in the tomato soup. The sensory acceptability test of this pilot study showed that the participants liked the taste, aroma, colour, appearance and mouthfeel of the control soups significantly more compared to the soups with added protein. There was a significant negative correlation between changes in colour with overall liking, liking colour and appearance of samples. The age of participants had a significant effect on overall liking and liking of aroma, as participants older than 55 years liked the samples with added protein less compared to the control.

* 1. Introduction

Good nutrition is one of the contributors to good health and well-being of older adults contributing to healthy ageing (WHO, 2020). Sufficient protein intake could potentially help limit the reduction in muscle mass that is a result of ageing (Deutz et al., 2014). Current guidelines suggest that the minimum amount of 0.8 g of dietary protein per kilogram of body weight daily is sufficient to meet the minimum nutritional requirement (European Food Safety Authority (EFSA), 2012). While some factors such as age, physical activity, and physiological changes may increase the need for protein to 1 - 1.2 g/ kg of body weight/day (Deutz et al., 2014). Baugreet et al. (2017) recommended an amount of protein between 1.2 and 1.5 g/kg of body weight/day for older adults with sarcopenia. Recent evidence shows that consuming proteins incorporated into foods as part of a normal eating pattern is a more efficient way to increase protein intake compared to the consumption of protein powder or supplements (Burd & Murphy, 2022). According to the regulation on Nutrition and Health Claims, a product can bear the claim "source of protein" and "high in protein" when at least 12% and 20% of the energy of the food is provided by protein, respectively (Regulation (EC) No 1924/2006, 2006). Milk protein concentrate (MPC) is one of the complete dairy proteins rich in essential amino acids, e.g., regularly used as an ingredient for nutritional or functional purposes in food systems (Agarwal et al., 2015). The demand for the replacement of animal proteins is increasing due to the increase of animal-based food consumption by consumers due to the increase of the population in the world (Searchinger et al., 2019), the increasing consumer’s awareness and concern about environmental sustainability and the health benefit of plant-based proteins (Aschemann-Witzel et al., 2020). Due to the low allergenicity, high availability and low cost of pea protein, it has attracted increasing attention as an ingredient in a number of food products (Lam et al., 2018). However, plant proteins in general have lower nutritional value due to the lack of some essential amino acids (AA) such as methionine and cysteine (Konieczny, 2019). One of the strategies to improve this nutritional limitation is to combine them with other proteins such as animal proteins (Alves & Tavares, 2019). The strategy of combining pea protein and milk protein concentrate is explored as a way to overcome the techno-functional and nutritional limitations of plant proteins, while increasing sustainability. Previous research exploring which foods could be fortified with protein, has shown that tomato soup was one of the most preferred products by older adults (Jamshidvand et al., 2022). Moreover, tomato soup can be consumed as an appetizer (Vashista et al., 2003) during lunch or dinner. Therefore, this study focused on protein fortification of tomato soup and aimed to investigate the effect of the addition of protein from two different sources; MPC and PPI individually or combined, on the techno-functional properties and the sensory acceptability of the final products.

* 1. Methodology
     1. Materials

Milk protein concentrate (MPC, 84% protein) and Pea Protein Isolate (PPI, 77% protein) were kindly provided by Kerry Co., Kildare, Ireland. Dehydrated tomato soup powder (Knorr®) was purchased from a local market.

* + 1. Sample Preparation

Fourteen samples of tomato soups that could bear the claim “high in protein” (Regulation EU No 1924/2006) were developed initially using protein powders (MPC and PPI), however four samples, a control (tomato soup without added protein) and three protein-fortified samples were selected for the sensory acceptability test by a convenience sample of naïve consumers, as described in Table 1. For each selected sample for the sensory evaluation there were three replicates. The ratios were chosen in order to provide the daily Leucine requirement to an older adult who consumes one portion of soup (WHO/FAO/UNU Expert Consultation, 2007). Samples were mixed with deionised water, boiled at 90°C for 5 min using a heating plate (Aluminium, VELP Scientifica Srl, Italy) and cooled to 60°C (common serving temperature for soup (Mattes, 2005)) before subjecting them to further testing.

Table 1 Formulations of tomato soup (TS) developed with Milk Protein Concentrate (MPC) and Pea Protein Isolate (PPI), percentage of energy that derives from protein and mg EAA Leucine/kg/day in 100ml reconstituted sample.

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Sample | MPC  (g) | PPI  (g) | Soup powder (g) | Water (ml) | % of Calories from protein\* | mg Leucine  (per100g sample) \*\* |
| TS (Control) | 0 | 0 | 9.18 | 90.81 | 11.76 | 86 |
| TS + PPI | 0 | 3 | 8.91 | 91.08 | 29.85 | 287 |
| TS + MPC | 3 | 0 | 8.91 | 91.09 | 32.50 | 316 |
| TS + MPC + PPI | 0.5 | 2.5 | 8.91 | 91.08 | 30.27 | 292 |

\* *Energy Conversion Factors of protein (% of Calories from protein) = ((g of protein per 100g of product x 4 (kcal/g)) / Total Energy per 100g of product in kcal) x 100*

*Based on Regulation EU No 1169/2011, g or protein provides 4 kcal of energy.*

\*\* *Leucine (mg/100g sample) = ((g of added protein per 100g sample) x (mg of Leucine per g protein)), 1g of MPC contains 7.66 mg of Leucine and 1 mg of PPI contains 6.7 mg Leucine.*

* + 1. Techno-functional properties of tomato soup

The apparent viscosity (η app) of all samples was measured at 60°C using a Brookfield (DV2T, Brookfield, Toronto, Canada) viscometer at a rotational speed of 100 RPM, spindle No 21 and shear rate 93 s-1. The pH of each soup at 60 °C was measured using a calibrated pH meter (EUTECH pH 700, Thermo Scientific, Sweden). A colourimeter (Lovibond LC 100/ RM 200, Colour Measurement, Tintometer Group, United Kingdom) was used to assess the colour of tomato soups with and without protein fortification at room temperature. Three L\*, a\* and b\* values were recorded within the CIELAB colour space. L\* represents the light dark axis (where 0 corresponds to black and 100 corresponds to white), a\* represents the green-red axis (+100 corresponds to red and -80 to green), and b\* represents the blue-yellow axis (+70 corresponds to yellow and -80 to blue) (Chavan et al., 2015). All measurements of the techno-functional properties were conducted in triplicate, unless stated otherwise.

* + 1. Sensory Evaluation

Three protein-fortified tomato soups and one control (tomato soup without protein) were all tested by 33 (17 male and 16 female) naïve consumers who were all older than 18 years (n=14 (18-25 y.o.), n=4 (26-35 y.o.), n=7 (36-45 y.o.),n=2 (46-55 y.o.), n=6 (older than 55 years)). The sensory acceptability test was carried out in the designated individual sensory booths under artificial white light in ATU, Sligo, Ireland. The participants were asked to provide information on their age, gender and knowledge about the functionality of protein before testing the samples. The above information was collected in order to investigate which samples are liked by different consumer groups. Then they received the samples monadically in a balanced random order. Samples were offered in white paper cups labelled with three-digit codes at a serving temperature of 60°C. Mineral water and plain crackers were given to participants to cleanse their palates between each sample.  Participants scored overall liking and liking of the appearance, taste, aroma, colour and mouthfeel of the sample using a nine-point liking scale (1 - dislike extremely to 9 - like extremely). Ethical approval for the study to proceed was obtained from the research ethics committee of the Department of Health and Nutritional Sciences in ATU, Sligo.

* + 1. Data Analysis

The statistical software IBM SPSS Statistics (Version 28) was used to perform statistical analysis of the results. All measurements of the techno-functional properties were reported as means ± standard (SD) of three replicates unless stated otherwise. One-way analysis of variance (ANOVA) followed by Tukey post-hoc (HSD) paired comparison test were applied to compare means of the techno-functional properties, overall liking and liking of the attributes of the four samples. The effect of gender, age and awareness of protein functionality on liking was tested using the non-parametric tests Mann-Whitney and Kruskal-Wallis (when two or mor than two factors were tested respectively). Spearman correlation was used to find correlations between overall liking and liking of the attributes with the techno-functional properties (colour L\*, a\*, b\*, pH and apparent viscosity). The significance for all statistical tests was defined at p< 0.05. The qualitative data derived from the comment boxes where participants were asked to justify their liking scores were analysed to generate codes, and the frequency of these codes was counted.

* 1. Result and discussion
     1. Techno-functional properties of tomato soup

Table 2 shows the techno-functional properties of the control soup and the ones with added protein. As seen in Table 2 addition of protein resulted in a significant increase in the apparent viscosity η app of tomato soup (p<0.05), with the control soup having the lowest (70.33±0.58 mPa.s) apparent viscosity and the soup with MPC having the highest (145.33±2.31 mPa.s). The increase in η app with increasing protein content can be attributed to higher levels of insoluble solids as was also reported by Chavan et al. who investigated the rheological properties of whey-based tomato soup with different total soluble solids (TSS) and found that the η app of soup increased by increasing TTS that resulted in higher levels of insoluble solids (Chavan et al., 2015). Increased viscosity of tomato soup with MPC could also be due to the gelation of MPC when heated at temperatures above 72 °C, as it was reported by (Gruppi et al., 2022). Interestingly the combination of proteins resulted in lower apparent viscosity compared to samples where MPC or PPI were added individually. The results of the present study agree with the finding of (Oliveira et al., 2022) who reported that the suspensions containing a combination of whey (a dairy protein) and pea protein resulted in weaker gels compared to the suspension containing pure whey protein.

Table 2 Techno-functional properties of tomato soup without protein (TS), with Pea Protein Isolate (TS + PPI), with Milk Protein Concentrate (TS+MPC), and with the combination of Milk Protein Concentrate and Pea Protein Isolate (TS+MPC+PPI). Different superscript letters (a, b, c and d) in each column show significant differences between means (p < 0.05).

|  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- |
| Sample | Apparent viscosity (mPa.s) | pH | L\* | a\* | b\* |
| TS (Control) | 70.33±0.58a | 3.20±0.01a | 27.84±0.23a | 20.98±0.10a | 24.82±0.13a |
| TS + PPI | 94.00±0.40b | 4.61±0.03b | 37.42±0.32b | 19.96±1.93b | 31.17±1.61b |
| TS + MPC | 145.33±2.30c | 5.07±0.08c | 45.25±1.40b | 18.84±1.16c | 27.13±1.28c |
| TS + MPC + PPI | 78.33±1.15d | 4.71±0.01b | 41.60±1.61c | 19.12±1.01b | 27.10±0.73c |

The result of the pH of tomato soup with or without protein powders that was measured at 60°C can be seen in Table 2. As seen on the table, the type of protein had a significant effect on the pH of the protein-fortified tomato soup (p<0.05). The tomato soup without protein fortification had the lowest pH (3.2±0.01) and addition of protein led to a significant increase in the pH of protein-fortified tomato soup (p<0.05). Possible differences in the pH of protein-fortified tomato soup using PPI vs MPC could be attributed to the different pH values of the pure protein solutions, e.g., for PPI solutions from 1.5 % w/w to 7.5% w/w the pH varied from 6.8 to 7.6 respectively, for MPC solutions from 1.5% w/w/ to 7.5% w/w/ the pH varied from 6.8-7.0. There were no significant differences in the pH between protein-fortified tomato soup with PPI and the one prepared using a combination of PPI and MPC possibly due to the lower levels of MPC (0.5% w/w) in the combined sample. Table 2 shows the results of the colour measurements of tomato soups with and without protein fortification based on three parameters (L\*, a\* and b\*). The type of protein had a significant effect on the colour of the product (p<0.05). The addition of protein (regardless of the type) significantly increased L\* (lightness) and b\* (blue/yellow colour) values, while it decreased a\* (red colour) values (p<0.05). The protein-fortified soup with MPC was the sample with the lightest colour with the highest L\* value (45.25±1.40) while the control had the lowest L\* value (27.84±0.23). Tomato soup fortified with PPI had the highest yellow colour with the highest b\* value 31.17±1.61) while the control had the lowest b\* value (24.82±0.13). The control had the highest red colour with the highest a\* value (20.98±0.10) and the protein-fortified soup with MPC had the lowest a\* values (18.84±1.16) These changes in the colour are due to the white colour of the MPC powder and the yellow off white colour of the PPI.

* + 1. Sensory evaluation

Since this study found that the addition of protein significantly affected the techno-functional properties of tomato soup the sensory assessment of fortified tomato soup was essential in order to understand consumer overall acceptability and acceptability of the sensory attributes that best described the products. As seen in Table 3 the control tomato soup without protein had the highest scores for overall liking and liking of the different attributes, followed by protein-fortified tomato soup with MPC and protein-fortified tomato soup with PPI while the protein-fortified tomato soup with a combination of PPI and MPC was the least liked samples. However, no significant difference in overall liking (p>0.05) was observed, while there were significant differences in liking of colour, mouthfeel, aroma, taste, and appearance (p<0.05). There was a significant difference in the liking of the soups between different age groups, where people older than 55 years (18.2% of participants) gave the lowest liking scores for overall liking (4.5±0.66) and aroma (4.38±0.82) to the fortified samples compared to the rest of the age groups (p<0.05). On the other hand, participants aged (36-45 yo.) gave the highest score for overall liking (6.6±0.41) and for aroma (6.71±0.57) to the fortified samples. These differences in liking scores could be attributed to changes in the sensitivity for taste, smell, and mouth feel observed with ageing (Norton et al., 2021). This was confirmed by previous studies where it was shown that older adults perceived soups as less creamy compared to younger participants and were less sensitive to changes in flavour profile of soups (Kremer et al., 2005). No differences were indicated in liking between men and women. Moreover, knowledge about the functionality of protein had no effect on how people scored liking of the soups (p>0.05), possibly because more than 70% of participants had good knowledge about protein functionality due to their science background. Qualitative analysis of the participants’ comments showed that the control soups received more positive comments related to their sensory attributes compared to the protein-fortified tomato soups. This further confirms the differences in the liking scores between soups and the low score for the protein-fortified soups as shown in Figure 1. Specifically, the participants did not like the consistency and aftertaste of protein-fortified tomato soups and mentioned that these soups had grainy mouthfeel, especially the one fortified with PPI. The changes in techno-functional properties had a significant effect on the perceived sensory properties and the overall acceptability of the protein-fortified tomato soup. Spearman correlation showed that there is a significant negative correlation between changes in b\* and overall liking (rho=-0.857, p<0.001), liking of colour (rho=-0.629, p=0.029) and liking of appearance (rho=-0.621, p=0.031) of the soup and a negative correlation of L\* with liking of appearance (rho=-0.622, p=0.031), while there was no significant correlation of liking with the rest of the techno-functional properties.

Table 3 Overall liking and liking of the sensory attributes of control tomato soup without added protein (TS), tomato soup with Pea Protein Isolate (TS+PPI), tomato soup with Milk Protein Concentrate (TS+MPC), and tomato soup with the combination of Milk Protein Concentrate and Pea Protein Isolate (TS+MPC+PPI). Different superscript letters (a, b, c and d) in each column show significant differences between means (p < 0.05).

|  |  |  |  |  |  |  |
| --- | --- | --- | --- | --- | --- | --- |
| Sample | Overall liking | Appearance | Colour | Aroma | Taste | Mouthfeel |
| TS (Control) | 6.36±1.9a | 6.31±2.1a | 6.85±1.6a | 6.52±2.1a | 6.76±2.1a | 6.79±1.8a |
| TS + PPI | 5.39±2.2a | 5.42±1.9ab | 5.18±1.9b | 5.03±2.1b | 5.39±2.2b | 5.42±1.9b |
| TS + MPC | 6.06±1.8a | 4.88±2.1b | 5.06±1.9b | 6.12±1.7ab | 6.12±1.9ab | 6.06±1.7ab |
| TS + MPC + PPI | 5.33±2.1a | 5.31±1.9ab | 5.30±1.9b | 5.03±2.1b | 5.45±2.1b | 5.12±2.3b |

* 1. Conclusions

The present study aimed to examine the effect of the addition of proteins from two sources (milk protein concentrates as an animal-based protein and pea protein isolate as a plant-based protein) either individually or their combination, on the techno-functional properties and sensory acceptance of protein-fortified tomato soup. Overall, the addition of MPC and PPI significantly affected the techno-functional properties (apparent viscosity, pH, and colour) of the tomato soup. Specifically, apparent viscosity (η app), pH, lightness (L\*) and yellow colour (b\*) of protein-fortified tomato soups were significantly higher than the control soups while a\* value was significantly lower than the control soup regardless of the type of protein. These changes in techno-functional properties had a significant effect on the perceived sensory properties and the overall acceptability of the protein-fortified tomato soup. There was a negative correlation between changes in colour (b\* values) and the overall liking and liking of colour and appearance of the tomato soups. Despite the limitation of the small number of participants, the sensory acceptability test showed that liking scores of the taste, aroma, colour, appearance and mouthfeel of the control soups were significantly higher compared to the soups with added protein. There were no differences in liking between men and women and knowledge about the functionality of protein had no effect on how people scored liking of the soups (p>0.05) while there was a significant difference in liking between the different age groups of participants. Specifically, people older than 55 liked the protein-fortified soup less than the control. This pilot study, despite the small number of participants from a convenience sample has provided an insight on how product development of the soups should be taken forward. For example, it is suggested to explore further the combination of the two proteins and investigate how solubility, viscosity and turbidity of the mixture change at different ratios of MPC and PPI. It is further suggested to modify proteins via enzymatic hydrolysis since previous studies have shown that this strategy can improve the techno-functional properties such as solubility, viscosity, and turbidity of proteins. In this way, the incorporated hydrolysates could have minimum impact on the techno-functional properties of the soup. However, enzymatic hydrolysis could also affect the taste of the final product since hydrolysates can be bitter. Therefore, sensory evaluation and acceptability testing specifically by the target population group (adults older than 55 years old) would be essential.

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