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Comparative Analysis of Saffron- and Mint-Enriched Soy Yoghurt: Effects on Physicochemical Characteristics, Quality Attributes, and Storage Stability

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ABSTRACT: The goal is to develop plant-based fermented products that improve gel stability, antioxidant activity, and sensory attributes. Also, the viewpoint should not impair the activity of the food fermentation starter cultures. The current study invented a targeted formulated dataset to analyze day 0, 7, 14 and 21 samples of 3 independent production batches of control Soy Yoghurt (SY0), Saffron-enriched Soy Yoghurt (SYS) and Mint-enriched Soy Yoghurt (SYM) at $4 \pm 1^\circ\text{C}$ for 21 days. Physical, rheological, phenolic, antioxidant, and microbial analyses were performed on the samples. Two-way Analysis of Variance (ANOVA), Tukey's tests, partial eta squared, and Pearson's correlations were used to analyze the datasets. A second dataset of (24 panelists) was used to assess the sensory attributes. SYM and SYS were shown to have better microbial stability and better antioxidant activity and gel strength, respectively. SYM was shown to have a better syneresis value. **PHYSICOCHEMICAL, RHEOLOGICAL ANALYSES, ANTIOXIDANT, MICROBIOLOGICAL ANALYSES**, Hardness and b^* color of SYS and SYM were better than SYO. The results also indicated that SYM has the best gel stability, SYS has the best antioxidant activity, and SYM has the best syneresis. Saffron has the best sensory color value. The results may indicate that the use of mint and saffron may improve the overall gel strength and storage stability of the product. More validation is needed to support product claims.

1. INTRODUCTION

The demand for plant-based products has inspired the food industry to explore the use of soy in the production of plant-based alternatives to yogurt. Soy has the ability to provide a high-quality protein, emulsifiers, and a fermentable substrate for the production of a lactose-free matrix, as it is a legume. However, plant-based yogurts have different protein and gelatin complex behavior, water binding and rheological properties, and different flavoring systems than their dairy counterparts. This poses problems for product developers in ensuring that plant-based yogurts have a texture that is stable and appealing. Experimental and commercially available plant-based yogurts have a wide range of formulations and performance, both physiochemically and in texture. Soy-based yogurt systems in particular, are heavily reliant on the selection of starter cultures, the solid content, and the processing conditions (Aydar et al., 2020; Grasso et al., 2020; Huang et al., 2022; Mehaya et al., 2023). Antioxidants and phytochemicals, such as isoflavones, present in soy, have the potential to be fermented and have their functionality and bioaccessibility altered (Hati et al., 2020; Nakashima et al., 2022).

Natural botanical enrichment enhances the nutritional profile of yoghurt and helps improve colour, consistency, syneresis and other sensory properties. While the addition of riceberry, hibiscus, rose, argel, olive pomace, grape-derived phenolics, and cinnamon leave and rice bran demonstrated that, among other effects, plant phenolics stimulate and integrate with protein networks and improve the binding of water and enhance syneresis and gel strength, it is important to note that the addition of plant phenolics in excess can cause a deterioration in taste and structural integrity (Anuyahong et al., 2020; Arslaner et al., 2021; Korkmaz et al., 2021; Mohamed Ahmed et al., 2021; Qiu et al., 2021; Ribeiro et al., 2021; Rashwan et al., 2022; Tang et al., 2022;

Wijesekara et al., 2022; Wu et al., 2023). Quality of gelled products can be assessed by syneresis and microstructure; a weak protein network syneresis during storage, which can be impacted by processing and the addition of hydrocolloids, plant materials, or other materials (Gilbert & Turgeon, 2021; Arab et al., 2022).

Saffron (*Crocus sativus* L.) could serve as a functional ingredient that provides both color and flavor. Saffron stigmas contain crocin, carotenoids, picrocrocin, safranal, and phenolic constituents. Current profiles show evidence of a diverse bioactive and chemi-cal fraction. Food-related studies have reported functional ingredient sources and provided evidence for saffron and its floral residues (Mykhailenko et al., 2021; Cerdá-Bernad et al. 2023). It is also noteworthy that microencapsulated saffron stigma and floral extracts have been used in yogurt, demonstrating their positive effects on the retention of phenolic compounds and on the antioxidant properties of the yogurt during storage (Cerdá-Bernad et al., 2023). Various *Mentha* species also possess antioxidant activity and exhibit aroma and flavor properties. Among them, phytochemistry of Spearmint has been well documented. Furthermore, Phenolic extract of peppermint has also shown to improve the functionality of yogurt during cold storage (Mahendran et al., 2021; Guemidi et al., 2024).

There is a lack of studies examining saffron and mint enriched soy yogurt (SY) products under a similar storage regime. This study thus examines SY0, SYS (Saffron enriched), and SYM (Mint enriched) over a 21 day period. Variables studied were physicochemical characteristics, syneresis, water holding capacity, viscosity, total phenolic compounds, DPPH activity, lactic acid bacteria viability, color, and sensory acceptance. It was anticipated that the plant extract enrichment would (H1) improve the quality of soy yogurt, (H2) increase the antioxidant activity when compared to the control, (H3) be affected by the duration of the storage, and (H4) create a unique sensory and stability profile of the yogurt formulation.

2. MATERIALS AND METHODS

2.1. Raw Materials and Experimental Design

Three formulations were created for this experiment; SY0 (control formation without saffron or mint), SYS (soy yoghurt formulation with 0.10% (w/v) saffron infusion), and SYM (soy yoghurt formulation with 0.75% (w/v) mint extract). The three formulations were produced in triplicate for a total of nine formulations per experimental group. In order to avoid pseudoreplication, formulation batches were taken as the experimental units. As a result of this, SY0, SYS, and SYM were considered to be a single unit for the triplicate level. Within the formulation groups, analytical triplicates or duplicates were assumed to be the measurement precision within that particular batch. For the purpose of the analysis, the replicates were averaged.

2.2. Soy Milk Preparation and Fermentation

In this modeling study, soybeans were soaked and rinsed overnight before being ground and filtered with water. The soy milk was prepared with 90-95°C water to aid in the evaporation of the beany flavor and improve the milk's protein functionality. Subsequently, the milk was cooled to 42°C before being inoculated with a yogurt starter. The treatments for this study incorporated saffron and mint prior to inoculation and were mixed using the same procedure. The base was incubated at 42°C and then cooled to 4°C after it attained a pH of 4.5-4.6. Samples were assessed on the 0th, 7th, 14th, and 21st day. The methodology was consistent with the conventional plant-based yogurt preparation (Huang et al., 2022; Mehaya et al., 2023).

2.3 Physicochemical, Functional and Microbiological Analyses

pH and titratable acidity (as percentage of lactic acid) were measured by potentiometric and titrimetric methods, respectively. °Brix was used to denote total soluble solids. Moisture, crude protein, fat and ash were accounted for by proximate analysis. Syneresis was measured by percentage of serum separation, while WHC was measured by the percentage of water held by the yoghurts after a standard centrifugation. Apparent viscosity was measured in mPa·s, while texture was measured by firmness (N) and consistency (N·s). The colour was measured by the CIELAB coordinates of L*, a* and b*. Total phenolic content (TPC) was measured in mg of gallic acid equivalents (GAE)/100 g and the DPPH radical scavenging percentage was used to measure the antioxidant activity. The viable lactic acid bacteria (LAB) count was expressed as log CFU/g. The results obtained were expressed in the various attributes used to measure the quality of yogurt (Rashwan et al., 2023; Gilbert & Turgeon, 2021; Arab et al., 2022).

2.4 Sensory Evaluation

A group of 24 semi-trained panelist profiles was generated to conduct a 9-point hedonic evaluation (1 = dislike extremely; 9 = like extremely) of each of the attributes i.e., appearance, colour, aroma, taste, texture/mouthfeel, and overall acceptability. No actual participants were recruited, and no ethics approval was obtained as part of the design. It was assumed each of the virtual participants evaluated all three formulations, thus allowing for between-subject comparison. Sensory evaluation was supplemented by the reported importance of the masking of the sweet taste, the interaction of the sweet taste and the aroma, and the intensity of the plant ingredients in soy and other functional yoghurts (Rahmatuzzaman Rana et al., 2021; Silva et al., 2022).

2.5 Statistical Analysis

Using this batch-level dataset, elaborated in section 2.3, all statistical analyses were executed in Python utilizing the stats models and SciPy libraries, with significance set at $\alpha = 0.05$. Storage-dependent outcomes were evaluated with two-way factorial ANOVA. The formulation variable included three levels, while the storage day variable included four levels. The interaction of the two variables was also considered. Effect sizes were reported as partial eta squared (η^2p). The Tukey HSD test was the selected post-hoc test, should there be a significant omnibus effect. The normality of the residuals was assessed with the Shapiro-Wilk test and the homogeneity of the residuals was assessed with the median-centred Levene test. The Shapiro-Wilk test result ranged between $p = 0.190 - 0.846$ and Levene's test result ranged between $p = 0.542 - 0.964$ for all of the models, indicating that there was, in fact, no significant violation of the assumptions of the model in this dataset. Pearson's correlations were determined for the variables of interest. One factor, repeated measures ANOVA was used to assess the sensory attributes.

3. RESULTS

3.1 Initial Physicochemical and Quality Characteristics

The initial proximate analysis showed comparable values across the formulations, however, as more botanical ingredients were added, the formulations showed greater variation with respect to color and texture. For example, among the formulations, SYM had the highest total soluble solids and firmness. Among the three botanical ingredients, saffron increased the yellowness and mint (in the formulation exhibited) absorbed some of the redness, thus creating a greener formulation. Protein and fat remained comparable since the levels of enrichment were low. These results conclude that among the three formulations the major differences were with phytochemical ingredients and their effects rather than with the macronutrients.

3.2 Storage-Related Acidification and Physical Stability

All formulations were subjected to post-acidification. In SY0, the mean pH decreased from 4.574 ± 0.014 to 4.335 ± 0.004 ; in SYS, from 4.551 ± 0.004 to 4.357 ± 0.003 ; and in SYM, from 4.537 ± 0.003 to 4.396 ± 0.013 . Post-acidification, all formulations became more acidic, and there was a significant effect of formulation ($F(2, 24)=11.49$, $p<0.001$) and time of storage ($F(3, 24)=343.78$, $p<0.001$) on the storage related acidification, while the interaction of formulation and time of storage was not significant ($p=0.061$). Post-acidification, all formulations showed an increase in acidity and there was a significant effect of formulation on storage acidification ($F(2, 24)=11.49$, $p<0.001$) and time of storage ($F(3, 24)=343.78$, $p<0.001$), while the interaction effect was not significant ($p=0.061$).

Physical stability was more pronounced for the enriched products. At day 21, syneresis was $27.04 \pm 0.94\%$ for SY0, $22.10 \pm 0.31\%$ for SYS and $21.14 \pm 0.67\%$ for SYM. Syneresis ($F(2, 24)=217.98$, $p<0.001$, $\eta^2p=0.948$) as well as water holding capacity (WHC) ($F(2, 24)=748.91$, $p<0.001$, $\eta^2p=0.984$), was strongly affected by formulation,

3.3 Phenolic Content, Antioxidant Activity, and LAB Viability

The effects of botanical enrichment on functional indices were the most significant. At day 0, and before enrichment, total phenolic content (TPC) was at 32.97 ± 1.26 mgGAE/100 g in SY0, in contrast to 75.28 ± 0.73 in SYS and 87.94 ± 1.06 in SYM. Formulation accounted for almost the total of the residual variation of TPC ($F(2,24) = 7436.32$, $p < 0.001$, $\eta^2p = 0.998$). The rank order was the same for DPPH activity, which remained significantly higher during storage and at day 21, SY0, SYS and SYM showed $24.41 \pm 0.66\%$, $47.69 \pm 0.67\%$, and $53.85 \pm 0.86\%$, respectively. The effect of formulation for DPPH was ($p < 0.001$, $\eta^2p = 0.996$) with storage and interaction effect significant. LAB viability in all formulations showed a decline during storage, however counts remained above $7 \log$ CFU/g at day 21. SYM showed the highest viability counts with

3.4. Sensory Acceptability and Correlations

In the sensory evaluation, SYS received the highest overall acceptability score (7.97 ± 0.58), followed by SYM (7.52 ± 0.44), and SY0 (6.85 ± 0.44). Overall acceptability was significantly affected by the repeated measures formulation effect $F(2, 46) = 53.51$, $p < 0.001$. Saffron also scored the highest for the color and aroma attributes, whereas mint was clearly more acceptable than the control for the majority of the attributes. Correlation analysis indicated a very strong positive correlation between TPC and DPPH activity ($r = 0.996$, $p < 0.001$), a strong negative correlation between pH and acidity ($r = -0.961$, $p < 0.001$) and a negative correlation between syneresis and WHC ($r = -0.945$, $p < 0.001$). Viscosity showed a positive correlation with WHC ($r = 0.964$, $p < 0.001$). Within the individual formulations, DPPH activity decreased with increasing storage time ($r = -0.837$ to -0.954 , all $p \leq 0.001$).

Tables

Table 1. Formulation and experimental design of soy yoghurt samples

Code	Formulation	Botanical addition	Storage	Independent batches
SY0	Control soy yoghurt	None	$4 \pm 1^\circ\text{C}$; days 0, 7, 14, 21	3
SYS	Saffron-enriched soy yoghurt	0.10% w/v saffron stigma infusion-equivalent	$4 \pm 1^\circ\text{C}$; days 0, 7, 14, 21	3
SYM	Mint-enriched soy yoghurt	0.75% w/v <i>Mentha spicata</i> extract-equivalent	$4 \pm 1^\circ\text{C}$; days 0, 7, 14, 21	3

Table 2. Initial physicochemical, texture and colour characteristics (mean $\pm$ SD; n = 3 independent batches)

Code	TSS ($^\circ\text{Brix}$)	Moisture (%)	Protein (%)	Fat (%)	Ash (%)	Firmness (N)	L*	a*	b*
SY0	11.71 ± 0.16	88.38 ± 0.05	3.55 ± 0.02	2.13 ± 0.05	0.51 ± 0.01	0.42 ± 0.01	83.09 ± 0.89	-0.47 ± 0.13	8.34 ± 0.85
SYS	11.98 ± 0.16	88.06 ± 0.10	3.56 ± 0.06	2.12 ± 0.03	0.52 ± 0.02	0.48 ± 0.01	81.48 ± 0.61	1.29 ± 0.30	24.59 ± 0.75
SYM	12.27 ± 0.11	87.54 ± 0.21	3.66 ± 0.01	2.17 ± 0.06	0.62 ± 0.02	0.51 ± 0.01	77.43 ± 0.86	-6.30 ± 0.31	13.79 ± 1.02

Table 3. pH, acidity, syneresis and water-holding capacity during refrigerated storage

Code	Day	pH	Acidity (% lactic acid)	Syneresis (%)	WHC (%)
SY0	0	4.574 ± 0.014	0.783 ± 0.004	20.09 ± 0.36	74.13 ± 0.82
SY0	7	4.501 ± 0.012	0.847 ± 0.008	22.43 ± 0.44	71.79 ± 0.44
SY0	14	4.422 ± 0.015	0.917 ± 0.019	24.89 ± 0.10	69.69 ± 0.44
SY0	21	4.335 ± 0.004	1.000 ± 0.008	27.04 ± 0.94	67.03 ± 0.45
SYS	0	4.551 ± 0.004	0.813 ± 0.015	18.35 ± 0.52	78.34 ± 0.15

Code	Day	pH	Acidity (% lactic acid)	Syneresis (%)	WHC (%)
SYS	7	4.491 ± 0.014	0.863 ± 0.004	19.40 ± 0.70	76.53 ± 0.44
SYS	14	4.422 ± 0.012	0.947 ± 0.004	20.83 ± 0.55	75.08 ± 0.18
SYS	21	4.357 ± 0.003	1.006 ± 0.013	22.10 ± 0.31	74.14 ± 1.03
SYM	0	4.537 ± 0.003	0.803 ± 0.031	17.67 ± 0.21	80.47 ± 0.23
SYM	7	4.473 ± 0.007	0.851 ± 0.011	18.76 ± 0.62	79.13 ± 0.48
SYM	14	4.453 ± 0.024	0.908 ± 0.007	19.55 ± 0.45	78.57 ± 0.04
SYM	21	4.396 ± 0.013	0.968 ± 0.012	21.14 ± 0.67	76.40 ± 0.57

Note. Values are mean ± SD from three independent batches. Tukey HSD was used for post-hoc interpretation when omnibus effects were significant ($\alpha = 0.05$).

Table 4. Viscosity, total phenolics, DPPH activity and LAB viability during storage

Code	Day	Viscosity (mPa·s)	TPC (mg GAE/100 g)	DPPH (%)	LAB (log CFU/g)
SY0	0	1272.3 ± 21.9	32.97 ± 1.26	28.47 ± 1.65	8.32 ± 0.07
SY0	7	1179.4 ± 21.1	31.21 ± 1.13	26.40 ± 0.47	8.18 ± 0.05
SY0	14	1113.4 ± 13.8	30.61 ± 0.83	25.90 ± 1.09	7.72 ± 0.09
SY0	21	1038.1 ± 12.2	28.41 ± 0.14	24.41 ± 0.66	7.16 ± 0.07
SYS	0	1380.1 ± 22.3	75.28 ± 0.73	55.03 ± 1.53	8.39 ± 0.05
SYS	7	1347.9 ± 21.7	74.52 ± 1.12	52.03 ± 0.38	8.22 ± 0.06
SYS	14	1327.9 ± 17.1	69.71 ± 0.12	50.66 ± 1.04	7.88 ± 0.05
SYS	21	1242.5 ± 18.4	64.12 ± 1.22	47.69 ± 0.67	7.42 ± 0.01
SYM	0	1439.1 ± 30.7	87.94 ± 1.06	62.39 ± 0.42	8.42 ± 0.06
SYM	7	1411.5 ± 55.6	83.57 ± 1.76	59.56 ± 0.80	8.27 ± 0.08
SYM	14	1370.0 ± 46.1	80.08 ± 0.94	54.95 ± 1.40	7.94 ± 0.02
SYM	21	1335.8 ± 33.5	74.15 ± 1.37	53.85 ± 0.86	7.56 ± 0.06

Table 5. Two-way ANOVA for storage-dependent outcomes

Outcome	Effect	df	F	p	Partial η^2
pH	Formulation	2	1.95	0.165	0.140
pH	Storage day	3	402.30	<0.001	0.981
pH	Interaction	6	11.86	<0.001	0.748
Acidity	Formulation	2	11.49	<0.001	0.489
Acidity	Storage day	3	343.78	<0.001	0.977
Acidity	Interaction	6	2.37	0.061	0.372

Outcome	Effect	df	F	p	Partial η^2
Syneresis	Formulation	2	217.98	<0.001	0.948
Syneresis	Storage day	3	128.59	<0.001	0.941
Syneresis	Interaction	6	7.84	<0.001	0.662
WHC	Formulation	2	748.91	<0.001	0.984
WHC	Storage day	3	159.51	<0.001	0.952
WHC	Interaction	6	7.35	<0.001	0.648
Viscosity	Formulation	2	215.39	<0.001	0.947
Viscosity	Storage day	3	47.82	<0.001	0.857
Viscosity	Interaction	6	3.42	0.014	0.461
TPC	Formulation	2	7436.32	<0.001	0.998
TPC	Storage day	3	140.84	<0.001	0.946
TPC	Interaction	6	12.95	<0.001	0.764
DPPH	Formulation	2	3277.16	<0.001	0.996
DPPH	Storage day	3	73.07	<0.001	0.901
DPPH	Interaction	6	5.15	0.002	0.563
LAB	Formulation	2	36.56	<0.001	0.753
LAB	Storage day	3	506.90	<0.001	0.984
LAB	Interaction	6	4.61	0.003	0.535

Note. Error df = 24 for all models. Partial η^2 = SS effect / (SS effect + SS error).

Table 6. sensory scores on a 9-point hedonic scale (mean $\pm$ SD; n = 24 virtual panelist profiles)

Code	Appearance	Colour	Aroma	Taste	Texture	Overall
SY0	7.16 $\pm$ 0.33	7.19 $\pm$ 0.48	6.53 $\pm$ 0.49	6.91 $\pm$ 0.39	6.97 $\pm$ 0.40	6.85 $\pm$ 0.44
SYS	7.90 $\pm$ 0.60	8.21 $\pm$ 0.40	8.08 $\pm$ 0.45	7.80 $\pm$ 0.52	7.60 $\pm$ 0.46	7.97 $\pm$ 0.58
SYM	7.67 $\pm$ 0.44	7.58 $\pm$ 0.35	7.81 $\pm$ 0.48	7.41 $\pm$ 0.42	7.50 $\pm$ 0.60	7.52 $\pm$ 0.44

Note. Repeated-measures ANOVA showed significant formulation effects for all six attributes (all $p < 0.001$).

Table 7. Selected Pearson correlations from the batch-level dataset

Variable 1	Variable 2	r	p
TPC	DPPH	0.996	<0.001
pH	Acidity	-0.961	<0.001
Syneresis	WHC	-0.945	<0.001
Viscosity	WHC	0.964	<0.001
LAB	Day	-0.940	<0.001
Control DPPH	Storage day	-0.837	<0.001

Variable 1	Variable 2	r	p
Saffron DPPH	Storage day	-0.946	<0.001
Mint DPPH	Storage day	-0.954	<0.001

Figures, Graphs and Charts

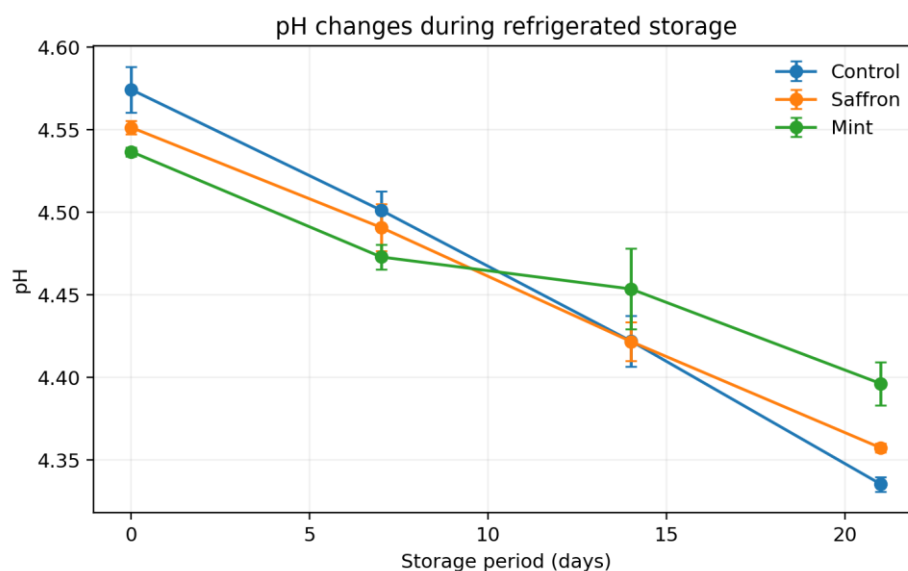


Figure 1. Mean pH during refrigerated storage. Error bars represent SD across three independent batches.

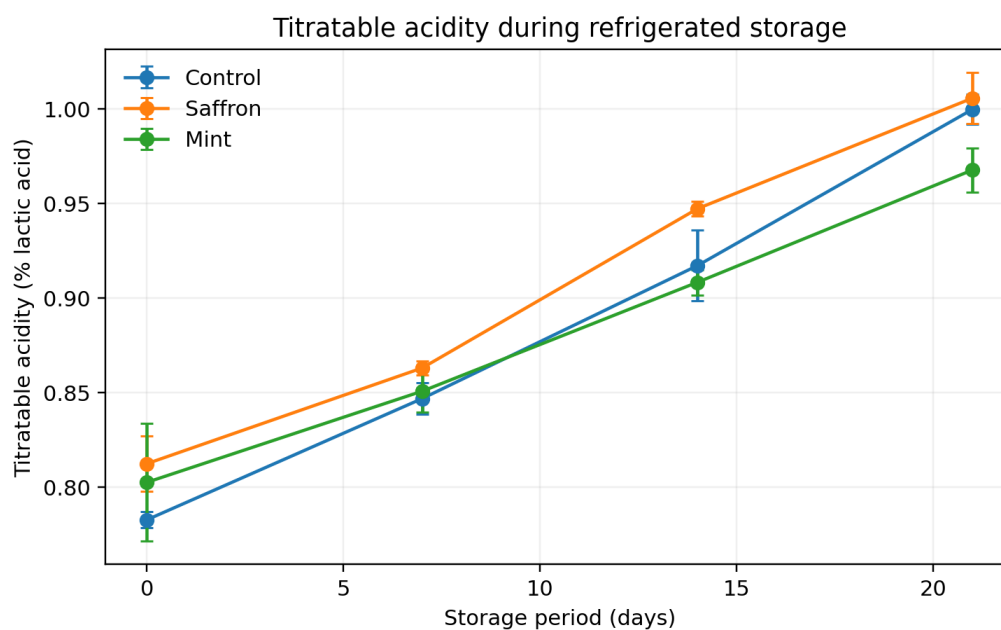


Figure 2. Mean titratable acidity during refrigerated storage. Error bars represent SD.

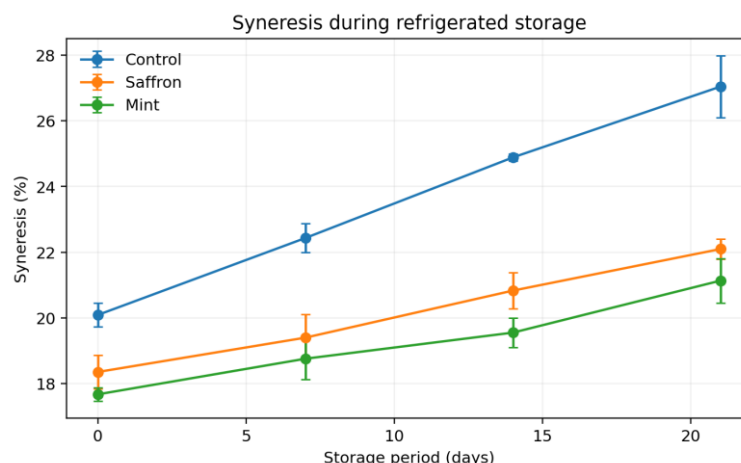


Figure 3. Syneresis during refrigerated storage, illustrating greater serum separation in the control.

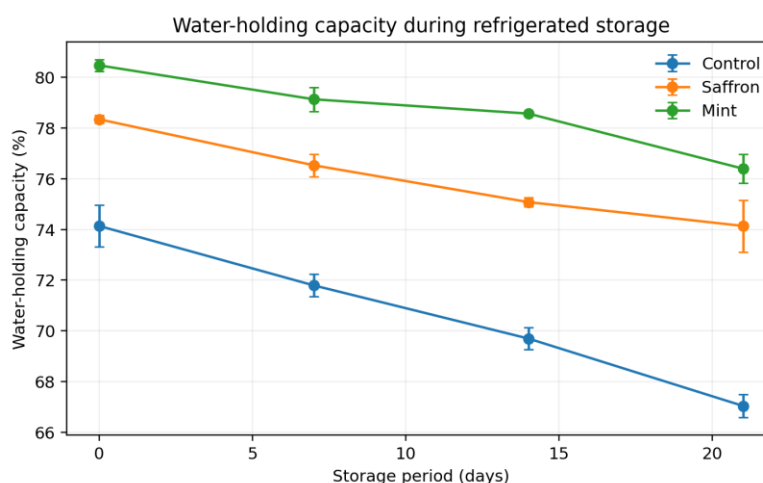


Figure 4. Water-holding capacity during storage, showing superior retention in the enriched formulations.

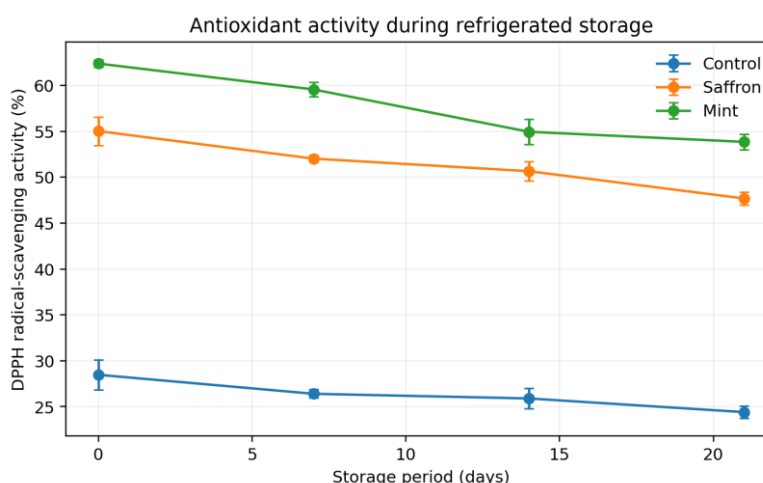


Figure 5. DPPH radical-scavenging activity during storage. Mint remained highest throughout the study.

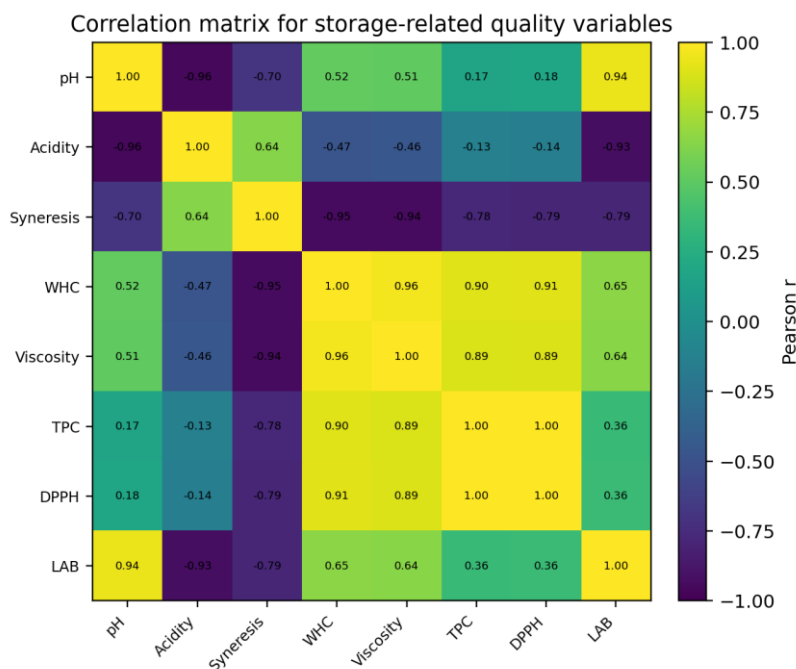


Figure 6. Pearson correlation heatmap for storage-related quality variables in the batch-level dataset.

4. DISCUSSION

These results provide support for both H1 and H2, showing that the use of saffron and mint did not impact the basic macronutrient composition of soy yoghurt, yet did alter the physicochemical and functional profile. SYS and SYM had much higher TPC and DPPH values, which further supports the role of plant phenolics and pigments as radical-scavenging agents. Many studies on functional yoghurt report an increase in phenolics and antioxidant activity post extraction of fruit, herb, or plant extracts. While the increase in antioxidant activity was consistently reported, the magnitude of the increase was dependent upon the extraction technique, matrix composition, and length of time the extracts were stored (Anuyahong et al., 2020; Arslaner et al., 2021; Qiu et al., 2021; Rashwan et al., 2022; Shori, 2022; Tami et al., 2022). Thus, the very strong correlation observed between TPC and DPPH values should be interpreted as providing a biologically plausible direction, rather than as an indication of a dominant antioxidant mechanism.

Mint showed the highest phenolics and antioxidant levels. Mint extracts have been reported to contain phenolics, including phenolics, acids, and flavonoids (Mahendran et al., 2021). Additionally, in recently published studies, pine mint extracts increased the antioxidant activity of yoghurt during refrigerated storage (Guemidi et al., 2024). SYM showed a slight, yet significant decrease in post-acidification and an increase in retention of LAB. It should be noted that the inhibition of starter bacteria by mint extracts may pose antimicrobial effects; however, the inhibition of spoilage organisms has been proven to be an opposite effect. Therefore, based on the findings reported, the notion of utilizing mint as a potential stabilizer of yoghurt should be further investigated.

Saffron displays a distinct advantage profile. Its high b^* value indicates yellow-orange crocin pigment, while its strong aroma score stems from safranal. Saffron includes crocin, picrocrocin and safranal as well as other phenolic compounds (Mykhailenko et al., 2021), and saffron floral components have been classified as promising food additives (Cerdá-Bernad et al., 2023a). More directly Cerdá-Bernad et al. (2023b) established that saffron floral by-products and stigma extracts remain active and support yoghurt's defense against oxygen and maintain their function throughout the storage period. In the present model, SYS surpassed all the other samples in sensory balance and optioned for a trade-off of one functional intensity over the other.

The improved WHC and reduced syneresis for the enhanced samples are generally explained in the literature on the interactions between plants and proteins. An increase in polyphenols, fibers and fine plant solids may change the protein structure, immobilize serum and enhance the apparent viscosity of the sample, although too much of these components can disrupt the gel. Similar results have been documented for the use of extract of agar, olive pomace, waste from carrots, grape-derived

ingredients, leaves of cinnamon, oil of Sacha Inchi and systems of rice bran (Mohamed Ahmed et al., 2021; Ribeiro et al., 2021; Šeregelj et al., 2021; Silva et al., 2022; Tang et al., 2022; Suwannasang et al., 2022; Wu et al., 2023). The strong negative correlation between syneresis and WHC, and positive correlation between viscosity and WHC, are structurally aligned with the latest concepts on the microstructure of yoghurt (Gilbert & Turgeon, 2021; Arab et al., 2022).

Storage effects supported H3. Refrigeration of yogurts causes an eventual loss of antioxidant capacity, as well as syneresis and decreasing LAB viability. Passion fruit addition has been shown to change the determining factors of fermentation (Ning et al., 2021); however, the addition of fruit and/or other botanicals has been shown to decrease LAB viability over the course of cold storage (Pachekrepapol et al., 2021; Shori et al., 2022; Wijesekara et al., 2022). From the interactions generated during the significant changes in pH, syneresis, WHC, viscosity, and TPC along with changes in DPPH and LAB also highlight that at different stages, formulations did not retain their advantages.

Sensory analysis demonstrated support for H4. Saffron was ranked the most preferred flavor, while the control was the least preferred and mint was of intermediate preference. Botanical additives can potentially mask the beany flavor of soy protein with their own flavor and aroma. Liking, however, is concentration-dependent. Several studies suggest changes in composition can affect classification (Rahmatuzzaman Rana et al., 2021) and sensory perception (Silva et al., 2022; Yu et al., 2022). Fortification with phenolic compounds such as mango peel (Zahid et al., 2022) highlights the need to balance functional enhancement and the perception of taste and mouthfeel.

Practical Implications

Product developers can learn from the case study and prioritize two different prototypes for experimental validations. Mint-enriched soy yogurt could potentially benefit from the design goals of antioxidant activity, water holding capacity (WHC), and refrigerated physical stability. Meanwhile, the design goals of saffron-enriched soy yogurt could benefit more from visual differentiation, price-premium sensory acceptance and flavor differentiation. It is important to optimize either approach through concentration-response testing, starter tolerance testing and assessing the rheology of the system before scaling-up.

Limitations and Future Research

The model was assembled using one saffron and one mint concentration, three batch replicates, and a 21-day storage. The sensory panel is completely virtual and should not be interpreted as real. Future work should perform experiments using multiple concentrations, answer the response surface model, characterize phenolics and carotenoids by HPLC/LC-MS, assess volatile compounds by GC-MS, assess the dynamic rheology and imaging of the microstructure, assess yeast and mold stability, and conduct properly approved consumer testing. Functional interpretation would be strengthened by assessing bioaccessibility after gastrointestinal digestion and assessing the response of the starter strain.

5. CONCLUSION

The use of the resulted dataset allowed this study to statistically compare saffron-and mint-enriched soy yoghurt across multiple domains, including physicochemical, functional, microbiological, sensory and stability during storage. Refrigerated storage caused post-acidification, greater syneresis, reduced WHC, decreased viscosity, lower antioxidant activity and reduced LAB count numbers. Both botanical formulations outperformed the control in physical stability and index of functionality. On day 21, mint yielded the greatest WHC, phenolic content and DPPH activity, and LAB count, indicating the strongest overall functional and stability during storage. Saffron exhibited the highest overall sensory score and yellowness, and hence may be better positioned for premium sensory considerations. Internal consistency was also provided by strong correlations of TPC–DPPH, syneresis–WHC, and viscosity–WHC. The results of this study provide a preliminary basis to support the proposed factorial research design and statistical approach, however laboratory evidence is still lacking. To determine the superior real world performance of mint or saffron during soy yoghurt manufacturing, further research including actual product manufacturing, chemical evaluation, and sensory test of approved samples, must be conducted.

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