Life Sciences Student Journal
2025, Vol3, No2 pp 45-54
DOI: 10.22034/LSSJ.2025.149

The evaluation of the impacts of *Rosa damascena* extract on the physiochemical, functional and sensory properties of soy milk-based yogurt

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Article Type: Research article
Received: 25 May 2025
Revised: 1 Jul. .2025
Accepted: 10 Jul. 2025
ePublished: 30 July. 2025

Keywords: Soy yogurt, *Rosa damascena* extract, Sensory properties, Antioxidant activity

Background: Soy yogurt provides essential protein but is often rejected by Iranian consumers due to undesirable taste and smell. Improving its sensory properties could enhance its acceptability .

Objectives: This study examined the impact of Rosa damascena extract on the microbial, functional, and sensory characteristics of soy yogurt .

Methods: Rosa damascena extract was prepared, and its total phenol content (24.31 ± 4.6 mg gallic acid/g dry extract) and antioxidant activity (against DPPH radicals) were measured . -Soy yogurt was prepared with 5% and 10% extract added before pasteurization .The physicochemical properties (pH, acidity, protein, fat, and antioxidant activity) and syneresis were analyzed .A 10-member panel conducted sensory evaluation .

Results: pure soy yogurt had lower pH, higher acidity, lower fat, and higher protein than extract-supplemented and cow's milk yogurt .Extract addition slightly reduced fat and protein but increased antioxidant activity ,Syneresis was lower in soy yogurt (with or without extract) compared to cow's milk yogurt ,The extract improved flavor, texture, and overall acceptability .The 5% concentration was most effective in enhancing sensory appeal .

Conclusions: Incorporating Rosa damascena extract at 5% improves the sensory quality of soy yogurt while maintaining functional benefits, making it a viable option for plant-based dairy alternatives. This modification could increase consumer acceptance and market potential .

Introduction

Soy is a rich source of high-quality proteins, and its various food products such as soy milk, yogurt, and tofu have gained great popularity in various societies. These products are considered as the functional products due to their high nutritional value and richness in vitamins B and K, as well as minerals such as phosphorus and magnesium, and unsaturated fatty acids (1). In addition, soy has therapeutic effects in various diseases such as hypercholesterolemia, cancer, and especially in cardiovascular diseases. Interestingly, countries with high soy consumption have lower prevalence of the aforementioned diseases (2).

Due to its high protein content, soy consumption has been increasing, and soy-based yogurt has received great attention as a substitute for dairy products (3). Importantly, soy milk can provide a suitable substrate for the growth of bacteria used in starter and facilitate the fermentation process during yogurt production (4). However, soy-based yogurts are not desirable in terms of sensory properties such as appearance and texture due to the difficulty of phase separation (5). Because, during the fermentation process, acidification is intensified, which is accompanied by protein instability and consequently poor gel formation. Also, the unpleasant taste of soy yogurts is another important challenge and one way to overcome it is to add aromatic products to its formulation. For example, Huang et al. (2022) combined soy yogurt with quinoa extract and found that this combination not only improved the fermentation and shelf life of yogurts, but also enhanced the textural and rheological properties of yogurt (6).

Rosa damascena is a plant belonging to the Rosaceae family that is rich in alkanes, alcohols, phenols, terpenes and terpenoids. The most important components of the extract include Linalool, Cirtonellol and Farnesol (7). This plant has antioxidant effects equivalent to α-tocopherol and prevents oxidative stress (8). In addition, its neuroprotective and cardioprotective effects have also been reported (9, 10), indicating that this plant is very valuable in terms of it pharmacological properties. Importantly, the pleasant aroma of *R. damascena* petal extract is favored by Iranian culture and therefore, it has high overall acceptance. Therefore, the aim of this study was to investigate the effect of *R. damascena* extract on the microbial, functional and sensory properties of plant-based yogurt prepared from soy milk.

2. Materials and Methods

2.1. Preparation of *R. damascena* extract

R. damascena petals were purchased from reputable apothecaries and after approval by a specialist botanist, were used to prepare the extract. For this purpose, 10 g of dried and ground petals were placed in 100 ml of hydroethanol solvent for 6 hours and after filtration, placed in a rotary evaporator (temperature 45 °C).

2.2. Anti-oxidant activity of extract

The DPPH method was used to measure the antioxidant activity of the extract. For this purpose, 1 ml of the extract was mixed with 1 ml of DPPH and brought to a concentration of 4 ml using methanol. Then, after vortexing and placing them in the dark, the absorbance intensity was read at a wavelength of 517 nm using a spectrophotometer.

2.3. Total phenols

Total phenols were assessed by the Folin–Ciocalteu method (11). 125 µL of the extracted extracts were mixed with 375 µL of water and 2.5 mL of 10% Folin reagent. After six minutes, two mL of 7.5% sodium carbonate was added to the resulting mixture. The absorbance of the reaction mixture was measured at 765 nm using a spectrophotometer (Unico, USA) after 90 minutes of storage in the dark.

2.4. Preparation of soy yogurt

In the present study, four types of yogurt were prepared: pure soy (A), soy + 5% extract (B), soy + 10% extract, and yogurt made from cow's milk. To prepare soy yogurt, after removing waste materials, soybeans were soaked in water for 180 minutes to facilitate peeling. Then, the seeds were placed in water containing 0.05% sodium bicarbonate and boiled for 300 seconds at 100°C. After removing the boiled water, the soybeans were washed, and 4 L of water per kg of soybeans was added and mixed at high speed for 600

seconds to obtain a slurry. In the next step, the slurry was filtered using cheesecloth and granulated sugar (70 g/kg, 7%) was added. Also, at this stage, *R. damascena* extract was added to the mixture at concentrations of 5 and 10%. Then, pasteurization was performed by heating (90°C) for 20 minutes. Finally, yogurt starter (0.1%) was added and placed at 42°C for 16 hours to complete the fermentation process. Finally, the resulting soy yogurt was placed in a refrigerator at 4°C.

2.5. Evaluation of physicochemical properties

The AOAC method was used to measure the pH and acidity of soy yogurt prepared and stored at 4°C for time intervals of 1, 7, and 14 days (12).

Total protein and fat content of yogurt samples were measured automatically according to standard methods (12) using a Milko-Scan 133B N.Foss Electric.

To measure the dry matter of soy yogurts prepared in this study, the Iranian National Standard method (No.1753) was used. For this purpose, 5 g of soy yogurt was placed in an oven at 100°C for 2 hours to dry completely. Then, weighing was performed to estimate the dry matter percentage. Also, centrifugation method (10 minutes at 5000 rpm) and the difference in weight of sample and supernatant were used to measure syneresis. Finally, the antioxidant activity of pure soy yogurts and those supplemented with extracts was measured at different days of storage using the DPPH method.

2.6. Sensory characteristics

The sensory characteristics of the soy yogurts prepared in this study, such as appearance, aroma, taste, texture, and overall acceptability, were evaluated by panelists (n=10) using a 5-option hedonic questionnaire. It is worth noting that the panelists were trained on how to complete the questionnaire before analyzing the sensory characteristics. They were also asked to rinse their mouths with drinking water after testing each yogurt. Also, the type of yogurt used was completely blinded to the panelists.

2.7. Statistical analysis

After ensuring normal distribution, the data were analyzed using ANOVA procedure and followed by Tukey's multi-range test. Data were reported as mean ± SD and data analysis was performed in GraphPad Prism V.8 software.

3. Results

3.1. Anti-oxidant activity and total phenols of *R. damascena* extract

The antioxidant activities of *R. damascena* extract were evaluated by the DPPH assay and the results indicated that the concentration-dependent antioxidant activity. As the extract concentration increased from 10 to 200 μg/mL, the DPPH free radical scavenging activity increased (Figure 1). However, this free radical scavenging activity was lower than that of ascorbic acid (vitamin C) at all concentrations.

The total phenol content of the *R. damascena* extract was estimated to be 31.24±6.4 mg of Gallic acid per gram of dry extract.

3.2. Physicochemical properties of soy yogurts

Pure soy yogurt had the lowest pH at the beginning of the study (4.47 ± 0.01) and yogurt prepared from cow's milk showed the highest pH (4.59 ± 0.02). Interestingly, the addition of 5% and 10% *R. damascena* extract to the prepared soy yogurt was associated with a significant increase in pH compared to pure soy yogurt at all storage times. However, the findings of the present study indicated a gradual decrease in pH in soy yogurts prepared and supplemented with *R. dama-*

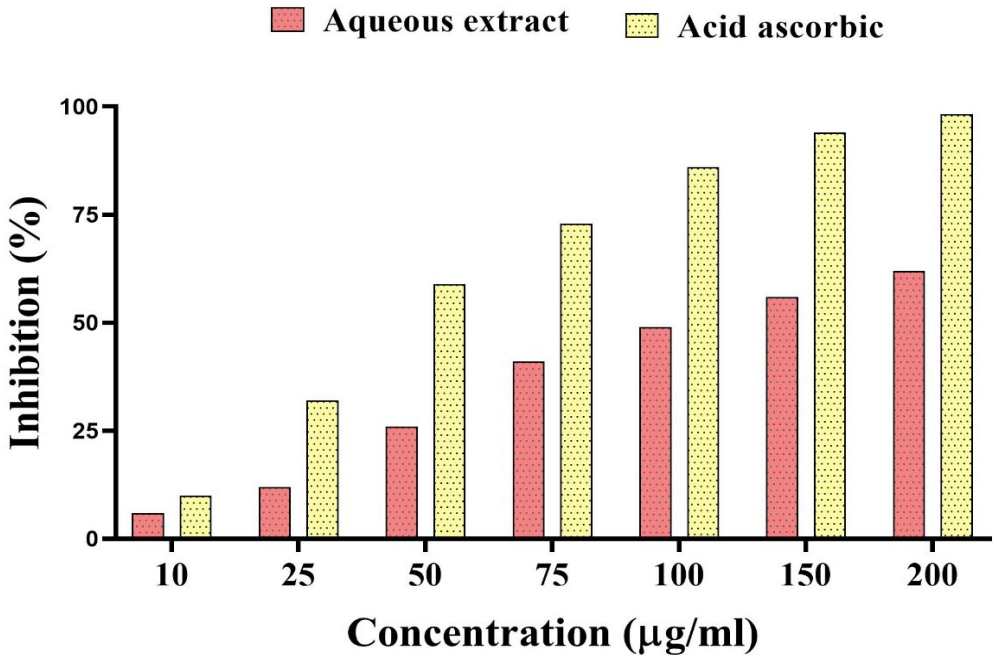

Figure 1. DPPH free radical scavenging activity of *R. damascena* extract in comparison with ascorbic

scena extract with increasing storage time at low temperature (4 °C). For example, the pH of pure soy yogurt reached 4.38 ± 0.006 on the final day of the experiment (day 14), and the pH of yogurts prepared from soy milk and supplemented with *R. damascena* extract at concentrations of 5 and 10% decreased to 4.48 ± 0.012 and 4.53 ± 0.015, respectively (Figure 2a).

Unlike pH, acidity increased with storage at cool temperatures, with the highest acidity being observed on day 14 of storage in all yogurts prepared from soy milk and also from cow's milk (Figure 2b). No significant difference was observed in terms of acidity of yogurts prepared on day 1 of the experiment, however, on day 7 of storage, a significant decrease in acidity was observed in soy yogurt supplemented with 10% *R. damascena* extract compared to yogurt prepared from cow's milk (P=0.043). Importantly, both pure soy yogurt and cow's yogurt had the highest acidity on day 14 of storage, showing a significant difference from both soy yogurt supplemented with 5 and 10% *R. damascena* extract (P<0.05). This indicates that the acidity of yogurt increases with the passage of time during cool storage, and it seems that the extract of *R. damascena* has prevented this increase in acidity to some extent.

2.4. Total dry matter, fat and protein contents

The results of the analysis of the dry matter, fat and total protein content of the soy yogurts prepared in this study are given in Table 1. Although the percentage of dry matter of the different soy yogurts prepared in this study increased slightly with increasing storage time, these differences were not statistically significant. The findings also indicated that the percentage of dry matter of soy yogurt was lower compared to yogurt prepared from cow's milk and it seems that the addition of *R. damascena* extract at the concentrations used (5 and 10 percent) has reduced the percentage of dry matter of soy yogurts.

Also, the findings of the study indicated that soy yogurts were higher in protein and lower in fat compared to yogurt made from cow's milk, with the highest pro-

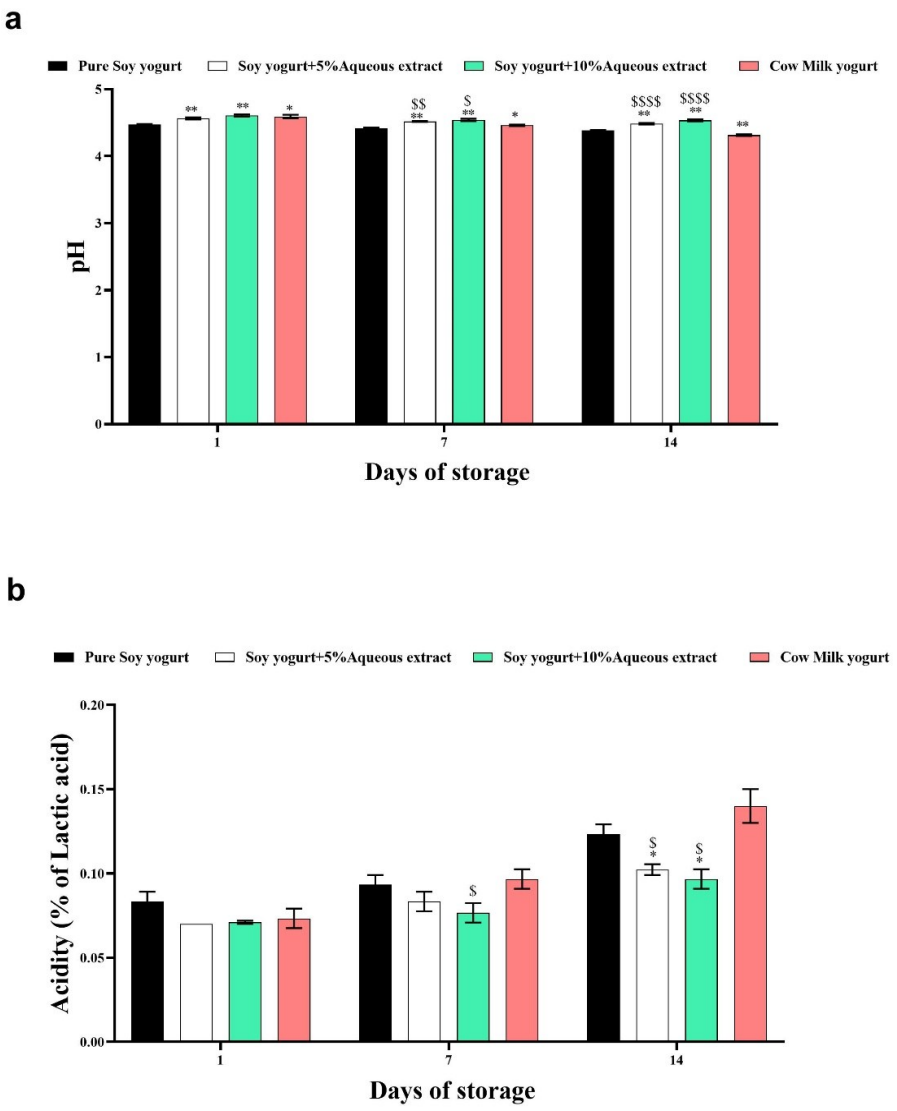

Figure 2. Changes in pH (a) and acidity (b) of pure soy yogurts and those supplemented with *R. damascena* extract (5 and 10%) during 1, 7 and 14 days of storage at 4°C (n=3). Indicates significant differences at the P<0.05 level compared to pure soy yogurt. $ Indicates significant differences at the P<0.05 level compared to yogurt made from cow's milk.

tein content and lowest fat content measured in soy yogurt, and the highest fat content and lowest protein content measured in cow's milk yogurt. Importantly, with increasing the duration of cool storage, both fat and protein contents of the yogurts increased slightly, and adding *R. damascena* extract to soy yogurt reduced both protein and fat. It is worth noting that there was no statistically significant difference in fat and protein content between soy yogurt supplemented with *R. damascena* extract at different concentrations and pure soy yogurt at different storage times (Table 1).

2.5. Syneresis

In the present study, an important indicator of yogurt quality, syneresis, was studied and the results are presented in Figure 3. As can be seen, yogurts made from soy milk and supplemented with *R. damascena* extract had a lower level of syneresis compared to yogurt

Table 1. Changes in dry matter, fat, and total protein contents of prepared pure soy yogurts and those supplemented with *R. damascena* extract, as well as cow's yogur, during 1, 7, and 14 days of storage at 4°C.

Yogurt types/Days	Dry matter (%)	Fat (%)	Protein (%)
Pure soy			
1	10.44±1.4	2.84±0.00	3.86±0.01
7	11.56±2.3	2.88±0.00	3.93±0.01
14	11.68±1.8	2.93±0.01	3.93±0.01
Soy yogurt+5% extract			
1	9.93±2.09	2.73±0.016	3.66±0.011
7	10.08±1.84	2.76±0.011	3.68±0.01
14	10.16±2.23	2.80±0.018	3.70±0.00
Soy yogurt+10% extract			
1	9.33±1.28	2.69±0.01	3.60±0.01
7	9.39±1.51	2.73±0.01	3.62±0.01
14	9.72±1.70	2.78±0.01	3.62±0.00
Cow yogurt			
1	13.34±1.55	3.83±0.02	2.56±0.012
7	13.59±1.24	3.90±0.012	2.61±0.014
14	13.44±1.37	3.93±0.01	2.063±0.012

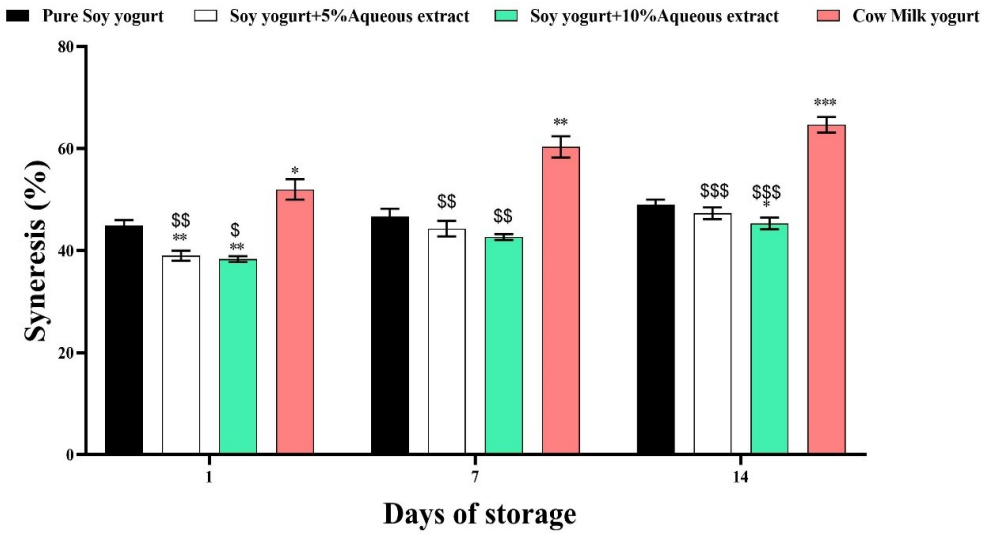

Figure 3. Changes in syneresis of pure soy yogurts and those supplemented with *R. damascena* extract (5 and 10%) during days 1, 7 and 14 of storage at 4°C (n=3)., and indicate significant differences at the probability level of P<0.05, P<0.01 and P<0.001, respectively, compared to pure soy yogurt. $, $$ and $$$$ indicate significant differences at the probability level of P<0.05, P<0.01 and P<0.0001, respectively, compared to yogurt prepared from cow's milk.

made from cow's milk. However, with increasing storage time at 4 °C, the level of syneresis of yogurts increases. Interestingly, adding the *R. damascena* extract was able to partially prevent the increase in this quality indicator of yogurt.

2.6. Anti-oxidant activity of yogurts

Adding *R. damascena* extract to soy yogurt significantly and concentration-dependently increased DPPH scavenging activity, and soy yogurts supplemented with 10% extract of *R. damascena* showed the highest free radical scavenging activity. This indicates an improvement in the antioxidant activity of soy yogurt with the addition of extract of *R. damascena*. It is worth noting that with increasing days of cool storage (7 and 14 days), a sharp decrease in DPPH scavenging activity was observed in all prepared yogurts. However, yogurts supplemented with *R. damascena* extract had antioxidant activity approximately 2 times that of pure soy yogurt and 3 times that of cow yogurt on the last day of storage (Figures 4).

2.5. Sensory characteristics

Sensory evaluation was performed by 10 panelists and the results indicated that there were significant differences in appearance, odor, taste and overall acceptability of the different soy yogurts (Table 2). However, there was no significant difference in texture scores. Pure soy yogurt had the lowest evaluation scores in all sensory parameters except texture compared to soy yogurts supplemented with 5% and 10% *R. damascena* extract. However, soy yogurt supplemented with 5% *R. damascena* extract showed better sensory properties compared to soy yogurt supplemented with 10% extract. It seems that increasing the concentration of the extract is associated with a decrease in acceptability. Therefore, soy yogurt supplemented with 5% extract of *R. damascena* had the best sensory properties and acceptability among the other yogurts.

4. Discussion

The results of the present study showed that the *R. damascena* extract has antioxidant activity against DPPH free radicals, although it was less than the antioxidant capacity of ascorbic acid. Also, the total phe-

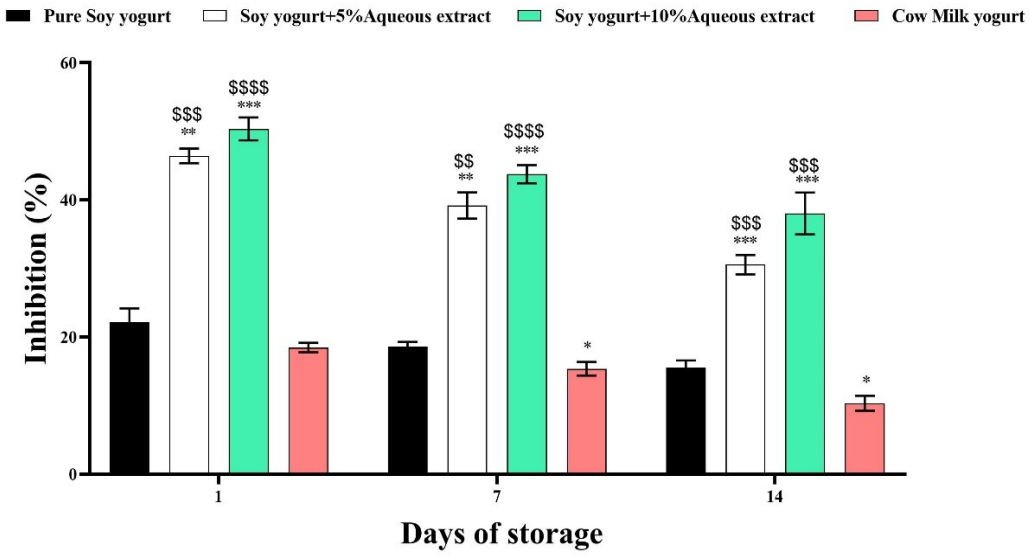

Figure 4. DPPH free radical scavenging by pure soy yogurts and those supplemented with *R. damascena* extract (5 and 10%) as well as cow's milk during days 1, 7 and 14 of storage at 4°C (n=3). , and indicate significant differences at the probability level of P<0.05, P<0.01 and P<0.001, respectively, compared to pure soy yogurt. $$, $$$ and $$$$ indicate significant differences at the probability level of P<0.01, P<0.001 and P<0.0001, respectively, compared to yogurt prepared from cow's milk.

Table 2. Sensory analysis of soy yogurt samples prepared in this study, evaluated by 10 panelists.

Attributes	Types	Min	Max	Mean	p-values
Appearance	Soy	2	5	3.28±0.44	0.029
	Soy+5% Extract	3	5	4.28±0.38	
	Soy+10% Extract	2	5	3.83±0.29	
Aroma	Soy	2	4	2.69±0.11	0.008
	Soy+5% Extract	3	5	4.89±0.42	
	Soy+10% Extract	2	5	3.24±0.31	
Taste	Soy	1	4	2.44±0.56	0.044
	Soy+5% Extract	2	5	3.82±0.38	
	Soy+10% Extract	2	5	3.22±0.78	
Texture	Soy	2	5	3.96±0.26	0.476
	Soy+5% Extract	2	5	3.86±0.14	
	Soy+10% Extract	2	5	3.73±0.33	
Overall acceptance	Soy	1	5	3.08±0.19	0.023
	Soy+5% Extract	2	5	4.15±0.22	
	Soy+10% Extract	2	5	3.69±0.33	

nol content of the *R. damascena* extract was estimated to be 24.31±4.6 mg of Gallic acid per gram of dry extract. In the next step of the study, pure soy yogurt supplemented with *R. damascena* extract was developed and the results indicated a low pH and high acidity in soy yogurt compared to soy yogurt supplemented with *R. damascena* extracts and cow yogurt. However, this trend was reversed with the passage of storage time and cow yogurt showed the greatest decrease in pH and increase in acidity. However, soy yogurt supplemented with *R. damascena* extracts showed less pH changes during cool storage. The lowest fat content and the highest protein content were observed in pure soy yogurt, and the addition of *R. damascena* extract led to a slight decrease in both fat and protein content in soy yogurt. However, 2 weeks of cold storage resulted in a slight increase in the fat and protein content of all yogurts prepared in this study. Also, less syneresis was observed in pure and *R. damascena* supplemented soy yogurts compared to yogurt prepared from cow's milk. Furthermore, the addition of *R. damascena* extract resulted in the highest antioxidant activity in soy yogurt, although a decrease in antioxidant activity was observed in all yogurts during cold storage. Furthermore, *R. damascena* extract improved the sensory properties and overall acceptability of soy yogurt compared to pure soy yogurt.

The results of the present study indicated that pure soy yogurt had low pH and high acidity, and the addition of *R. damascena* extract was associated with a slight increase in pH and a decrease in acidity. It has been shown that the type of bacteria used during the fermentation process in yogurt production has an impact on the physicochemical properties of the product (13). During the fermentation process, hydrophilic proteins are removed from soy milk, which leads to the formation of a gel network in the product. The results

of this study also showed that the pH of pure soy yogurt and supplementation with *R. damascena* extract decreased after fermentation, and on the other hand, the acidity increased. In addition, syneresis was also measured as an important quality factor in yogurt production, and the results showed that the percentage of syneresis increases with storage time, which can be attributed to the reduction of peptide bonds between proteins and the destruction of the gel network (14). Therefore, the increase in syneresis in prepared soy yogurts can be attributed to the decrease in the capacity of soy proteins to retain water in the yogurt gel network. However, the addition of *R. damascena* extract prevented the increase in this important indicator of yogurt quality. It seems that the compounds in *R. damascena* extract prevented protein denaturation and destruction of peptide bonds in soy yogurt, which helped retain water in the yogurt gel network. Also, the bacteria in the yogurt starter have the ability to use soy oligosaccharides as a substrate for fermentation (15), which produces lactic acid and increases the acidity of the product. For fat and protein content, soy milk had the lowest fat and highest protein content, and cow's milk yogurt had the highest fat and lowest protein content, which is mainly due to the high fat content of cow's milk and the high protein content of soy, which is consistent with the results reported by (16).

The soy yogurt sample supplemented with 5% *R. damascena* extract had the highest score for taste and aroma, followed by the soy yogurt sample with 10% *R. damascena* extract for both taste and aroma during storage periods. This finding indicates that the addition of *R. damascena* extract can be considered as a supplement to improve the sensory properties of yogurt and, as a result, overall acceptance.

5. Conclusion

In general, it was concluded that the addition of 5% *R. damascena* extract can be considered in soy yogurt formulation without negatively affecting the optimal physicochemical properties, which is accompanied by improved sensory properties and overall acceptance of soy yogurt.

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