Effect of black carrot size usage on the quality of shalgam (Şalgam): A traditional Turkish lactic acid fermented beverage

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In this study, the effect of black carrot size on the quality of shalgam was researched. For this purpose, the experiments of shalgam production using traditional production method were carried out by cutting black carrots 3, 6 and 9 cm and also lengthwise in size. Dough fermentation was done for 3 days and carrot fermentation for 9 days. According to the results obtained from the ready-to-serve shalgams; total acidity as lactic acid were found between 7.15 to 7.75 gm/L, lactic acid between 5.6 to 6.3 gm/L, pH between 3.45 to 3.53, anthocyanin as cyanidine-3-glycoside between 120.18 to 145.6 mg/L, total phenolic compounds as OD₂₈₀ between 23.3 to 28.99. Sensory analysis showed that the most preferred sample was the one done by using 3 cm size of black carrot. The results stated that the smaller size of black carrot usage favourably affected the anthocyanin content, phenolic composition and sensory properties of shalgam.

Keywords: Black carrot, Size, Shalgam, Fermented beverage, Lactic acid fermentation

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Traditional foods and beverages production is one of the oldest manufacturing and preservation methods, dating back to old days¹. Shalgam is a red coloured, cloudy and sour soft Turkish fermented beverage which is obtained by lactic acid fermentation². It is traditionally made at home or at the home-scale level. Shalgam is also produced commercially on small and large scales²-³. The essential process consists of preparation of raw materials, fermentation, and packaging⁴. Shalgam is highly popular in Southern Turkey, but it has also become a popular drink in other parts of Turkey. Lately, it is also sold in markets of some European cities⁵. The black carrot size is important for the production of shalgam, because some parameters such as anthocyanin and total phenolics levels are greatly influenced by the size of black carrot.

The objective of this research was to evaluate the effect of black carrot size usage on the quality of shalgam production using traditional method, and to determine the most suitable size of black carrot in shalgam production.

Methodology

Shalgam production by traditional method was carried out according to Erten et al.³ and Erten & Tanguler⁴. The traditional method consisted of 2 stages: Dough fermentation and carrot fermentation. The mixture of bulgur flour (3%), rock salt (0.2%), sourdough (0.2%) and adequate drinkable water was kneaded for the formation of dough. The dough obtained was left for the first fermentation at 25°C for 3 days. After that time, drinkable water was added into fermented mixture, blended well and extracted for 15 min. The extraction was repeated 4 times. The extracts were used for carrot fermentation. The black carrots were chosen in 2–3 cm diameter and in 10–12 cm size and then, the experiments were carried out by cutting the black carrots into 3 cm (sample T), 6 cm (sample S), 9 cm (sample N) and also lengthwise (sample L). Those sorted and chopped black carrots (20%) were combined with rock-salt (1%) and extracts obtained from dough fermentation in a 10 L of closed glass jar. Fermentation was carried out at 25°C in duplicate and followed by measuring pH and total acidity (TA) as lactic acid. After the completion of fermentation, shalgam was stored at 4°C.
Enumeration of microorganisms

For enumeration of LAB, samples were taken from the centre of 10 L of closed glass jar, serially diluted (10^{-1} to 10^{-8}) and spread-inoculated onto MRS Agar (Merck), supplemented with 50 mg/L cycloheximide to prevent the growth of yeasts. Plates were incubated at 30°C for 3-5 days in jars anaerobically using Gas Pack (Anaerocult® A, Merck). PCA (Merck) was used for the count of total mesophilic aerobic bacteria (TMAB) and incubation was conducted at 30°C for 3 days. Yeasts were enumerated on PDA supplemented with 0.1 gm/L oxytetracycline to prevent the growth of bacteria. L-lysine agar was used to count non- 
S. cerevisiae yeasts (N-SY). The plates were incubated at 25°C for 4-5 days. S. cerevisiae spp. was calculated by subtracting the total count of N-SY from the count of total yeast. Coliform bacteria (CB) were enumerated on VRBA agar, after incubation at 30°C for 1-2 days.

Chemical analysis of ready-to-serve shalgam

TA was measured using digital pH meter and expressed as grams of lactic acid/L. Total solid, total sugar and ash were analysed according to AOAC. Reducing sugar was determined using DNS method and salt was measured according to Cemeroglu. The contents of total phenolic compounds were determined by measuring the absorbance value at 280 nm. For determination of total anthocyanins as cyanidin-3-glycoside, the spectrometric method of Wrolstad was used. Colour values of L*, a* and b* measured with a Minolta Chroma Meter CR-100. Hue angle (a=tan b/a) and Chroma (a^2+b^2) values were also calculated.

Determination of organic acids and ethanol content of ready-to-serve shalgam

Ethanol, acetic and lactic acids were analysed by a HPLC (ShimadzuLC-20AD, Japan) using an Aminex HPX-87H column (Bio-Rad, 300x7.8 mm) at 50°C. Shalgam samples were centrifuged and filtered before being injected onto column. The eluent was 5 mM H_2SO_4 in high-purity water at a flow rate of 0.6 mL/min. Concentrations of ethanol were calculated from RI detector and acetic and lactic acids from UV detector. Standards (Merck) were used to determine lactic acid, acetic acid and ethanol amounts. All measurements were performed in duplicate.

Sensory analysis

Ranking test was done according to Barillere & Benard using a taste panel of 13 assessors (5 females and 8 males between 22 and 53 yrs old). Samples were numbered from 1 (very good) to 4 (very poor) and served in mixed order. Bran bread and water were given to the panelists to neutralize taste between the sample evaluations. They were ranked from the most preferred to least preferred one. For the descriptive test, panelists evaluated to colour, smell, taste and overall impression of shalgam samples on a horizontal 10-cm scale.

Statistical analysis

Results obtained were evaluated for statistical significance by 2-way analysis of variance (ANOVA) using the windows programme SPSS version 10.0 (SPSS Inc., Chicago, USA). If the analysis indicated a statistically significant difference, Duncan Multiple Range test was applied to compare the differences. For statistical analysis of sensory evaluation, data was analysed by Kruscal-Wallis test.

Results and discussion

In the production of shalgam, no comparative research has been realized before, concerning different sizes of black carrot. For this reason, shalgam fermentations were carried out for the determination of the effect of different sizes of black carrot on the quality of shalgam. Fermentations were followed daily by measuring pH and TA as lactic acid. The initial total sugar, TA and pH of bulgur flour were found as 18.20 gm/kg, 1.36 gm/kg and 5.76, respectively. In addition, those properties were determined as 70.9 gm/kg, 0.28 gm/kg and 6.03, respectively for black carrot. Similar results were reported in previous studies.

During the dough fermentation process, the pH gradually declined, due to the accumulation of lactic acid. TA as lactic acid increased from 3.6 gm/kg at the beginning to 11.8 gm/kg at the end. pH value decreased from 5.63 to 5.01 after 3 days of fermentation during the dough fermentation. Similar results for TA (8.05 and 11.9 gm/kg) and lower results for pH (4.01 to 4.37) were reported at the end.
of fermentation in previous trials using traditional method for shalgam production\textsuperscript{21-25}. Total LAB, TMAB, \textit{Saccharomyces} spp. and N-SY counts of dough were determined as 7.1 log cfu/gm, 7.17 log cfu/gm, 7.12 log cfu/gm and 6.36 log cfu/gm, respectively (data not shown). Results in this study were found lower than previous data published by Gunes\textsuperscript{23} and Aydar\textsuperscript{26} for total LAB and TMAB, but N-SY counts slightly higher than reported counts by Tanguler\textsuperscript{25}.

At the end of the dough fermentation, the dough was extracted four times with water and extracts obtained were combined. Its TA expressed as lactic acid was 0.39 gm/L and pH was 6.46 (results not shown). pH value obtained in dough was higher and TA was lower than reported data\textsuperscript{23,25}. Then, the extract was added into tank containing chopped black carrots (3, 6, 9 cm and lengthwise) and rock salt to carry out the carrot fermentation.

\textbf{Microbial changes during the carrot fermentation}

Fig. 1 shows the growth of total LAB during the fermentations. As can be seen from the Figure, at the beginning of fermentation, total LAB counts were found between 7.31 and 7.49 log cfu/mL and after fermentations started, the amounts of LAB increased rapidly. The highest numbers of 8.75 log cfu/mL for samples T and S, and 8.73 log cfu/mL for sample N were achieved within the 2\textsuperscript{nd} and 3\textsuperscript{rd} days of fermentation, respectively. After maximum growth, populations in all samples decreased slightly until the 7\textsuperscript{th} day of fermentation. Instead of the classical stationary phase after maximum growth, LAB had a gradual decrease during fermentation. LAB counts were found between 7.30 to 7.49 log cfu/mL at the end of fermentations. The data reported in the present study were similar to the works reported by Tanguler and Erten\textsuperscript{2} and Gunes\textsuperscript{23}.

Fig. 1—The growth of lactic acid bacteria during the carrot fermentation
Addition of black carrot size of 3 cm (−Δ−), 6 cm (−□−), 9 cm (−○−) and lengthwise (−♦−)

The amount of TMAB at the beginning of carrot fermentations ranged from 7.28 to 7.49 log cfu/mL (Results not given). Their value increased rapidly, and the highest numbers were achieved within the 3\textsuperscript{rd} day of fermentation (except for sample N). At the end of fermentation, TMAB counts were found between 7.08 and 7.64 log cfu/mL (Results not shown). The data obtained in this study are similar to the results obtained by Tanguler\textsuperscript{23} and Aydar\textsuperscript{26}. However, TMAB counts found in shalgam samples in the present study were 2 log units higher than that of given by Turkish shalgam standards TS 11149\textsuperscript{27}.

The occurrence of yeasts in carrot fermentation may originate from the surfaces of raw materials (including the bakers’ yeast or sourdough) and surfaces of shalgam equipments\textsuperscript{3}. The development of total \textit{Saccharomyces} spp. during the carrot fermentation is given in Fig. 2. At the beginning of fermentation, total \textit{Saccharomyces} spp. count was ranged from 6.32 log cfu/mL to 7.29 log cfu/mL. Its amount in all experiments increased until the day 3 and the highest values were obtained that day. Later on, total \textit{Saccharomyces} spp. count decreased slightly and their value was determined between 7.13 to 7.59 log cfu/mL.

In present study, N-SY count at the beginning of fermentations ranged from 4.48 to 5.54 log cfu/mL and their population rose quickly from the beginning of fermentation to day 2 and the highest numbers were obtained in all samples (Data not given). Later on, populations in all trials decreased slightly. However, at the end of fermentation, the highest and lowest non-\textit{Saccharomyces} values were determined as 6.02 and 6.47 log cfu/mL in samples T and N, respectively (Results not given). N-SY counts obtained in this study were found slightly higher than reported counts by Tanguler\textsuperscript{25} who found the last

Fig. 2—The growth of \textit{Saccharomyces} spp. during the carrot fermentation
Addition of black carrot size of 3 cm (−Δ−), 6 cm (−□−), 9 cm (−○−) and lengthwise (−♦−)
counts in the range of 4.21 and 5.19 log cfu/mL at the end of fermentation. In contrast, Ozhan\textsuperscript{28} has stated that yeast was not observed in shalgam.

The initial CB counts were found between 2.78 and 3.51 log cfu/mL (Results not shown). With the start of carrot fermentation, their counts increased only for first day, reaching a maximum of 4.69-4.88 log cfu/mL. But, in following days, their numbers decreased steadily during fermentation with an increase in the acidic content, therefore a drop in pH, and they disappeared by day 3 (sample L), 4 (sample N) and 5 (samples T and S) of the fermentations. Total coliforms seemed to be more sensitive than were LAB and TMAB to the inhibition effect of lactic and acetic acids produced during fermentation. The data is matched with the results obtained by Gunes\textsuperscript{23} and Ozer\textsuperscript{29}, Tanguler & Erten\textsuperscript{24} and Yener\textsuperscript{20} reported that coliforms were detected as 0.85–2.20 log cfu/mL at the end of fermentation in commercial shalgam samples. These different CB results were probably attributable to microbiological load found at raw materials and inadequate sanitary conditions carried out during processing.

Development of total acidity and pH during the carrot fermentation

As can be seen from the Fig. 3, pH and the amounts of TA at the beginning varied from 6.05 to 6.27 and 0.48 to 0.6 gm/L, respectively. As expected, pH decreased rapidly after fermentations started, whereas TA increased. This situation was due to formation of organic acids with the fermentation of sugars by mostly LAB. Increase in surface of black carrot contacted with extract accelerated the formation of TA. Especially, acid production rate was very high in samples T and L. However, acid production rate slowed down towards the end of fermentation in those samples, but increased in samples S and N. At the end of fermentation, the amounts of TA were determined between 7.15 and 7.75 gm/L and pH values were 3.45 and 3.53, respectively. There was significant (\(P<0.05\)) effect of black carrot size on TA and pH value. According to the Turkish shalgam standards\textsuperscript{27}, TA in shalgam must be at least 6.0 gm/L as lactic acid and pH between 3.3 and 3.8. Our findings for TA and pH are in agreement with Turkish Standard\textsuperscript{27}.

The change in phenolic composition and anthocyanin content

Phenolic compounds contribute to sensory qualities of fruit, vegetables and fermented products\textsuperscript{31}. Total phenolic compounds were determined from 4.10 to 4.95 at the beginning of fermentation (Results not shown). After a start of fermentation, the amounts increased steadily throughout fermentation and reached the maximum levels on the 9\textsuperscript{th} day ranging from 23.30 (sample N) to 28.99 (sample T) (Data not given). It could be said that the increase in the surface of black carrot contacted with extract accelerated the amount of phenolic compounds passing in to the liquid. Similar results for shalgam were presented by Tanguler\textsuperscript{25} and Nesanir\textsuperscript{32}.

Recently, shalgam has been in focus because of its high anthocyanin content. The main raw material used in shalgam production is black carrot which contains high amount of anthocyanins\textsuperscript{33}. The main anthocyanin pigments presented in black carrot and thereby shalgam, are cyanidin-3-glycosides\textsuperscript{3}.

After cutting black carrots into 3, 6 and 9 cm sizes and also lengthwise, they were combined with rock-salt (1\%) and extracts obtained from dough fermentation in a 10 L of closed glass jar and mixed by homogeneous. At the beginning of fermentation, total anthocyanin contents were determined between 0.30 to 2.55 mg/L (Fig. 4). Their values ranged from 120.18 to 145.6 mg/L at the end of fermentation (\(P<0.05\)). The highest value was obtained in sample T (145.6 mg/L) which was added 3 cm size of black carrot and then, in sample L (140.12 mg/L) which was added lengthwise in size black carrot. The lowest amount of anthocyanin was found in sample N which was added 9 cm in size black carrot. As it was expected, increase on the surface of black carrot contacted with extract accelerated the amount of anthocyanin passing into the liquid. The results
obtained in this study are similar to the results given by Gunes and Nesanir.

The effect of black carrot size on general composition of shalgam

Table 1 shows the general composition of ready-to-serve shalgam. This is the first study on the effect of the addition of different black carrot size for composition of shalgam. As can be seen in Table 1,

![Graph showing change in total anthocyanin amount](image)

**Fig. 4**—The change in total anthocyanin amount of the shalgams

Addition of black carrot size of 3 cm (—Δ—), 6 cm (—○—), 9 cm (—♦—) and lengthwise (—□—)

only some parameter values were dependent on the size of black carrot. ANOVA results showed that addition of different black carrot size did not affect the contents of acetic acid, total sugar and residual sugar, total solid and salt of shalgam samples.

Lactic acid, which accounts for the majority of TA, is the main end product of shalgam fermentation. During dough and carrot fermentations of shalgam, lactic acid formed by mostly homofermentative and heterofermentative LAB helps to preserve the beverage and enhances taste and aroma of shalgam.

In present study, the amounts of lactic acid were found close to each other, except for sample L. The shalgam made from lengthwise cut black carrot showed a markedly lower concentration of lactic acid (5.6 gm/L) compared to other shalgams made from 3, 6 and 9 cm sizes of black carrot (P<0.05). According to the Turkish shalgam standards, the concentration of lactic acid should be between 4.5 and 5.5 gm/L. In the present study, the lactic acid levels of shalgam samples were determined higher than the standards of Turkish shalgam and Tanguler & Erten. However, the concentrations of lactic acid in shalgam samples collected from commercial plants were reported as 2.61 to 8.75 gm/L by Yener and Ozturk. During the shalgam fermentations, acetic acid is also formed in minor amounts. Acids such as lactic and acetic acids have antimicrobial effects on microorganisms in fermented products. However, the amounts of high acetic acid are generally problem for fermented products. For this reason, they are not desirable from sensorial properties and quality aspects.

In this study, acetic acid concentrations of shalgams ranged from 0.48 gm/L to 0.56 gm/L. These findings for acetic acid are also evident from other reports given by Ozturk and Arici. Although lactic acid was the major product in the carrot fermentation, some ethanol was also produced. In present study, its concentration ranged from 3.00 to 3.72 gm/L and was significantly affected from black carrot size (P<0.05). Yeasts were generally responsible for ethanol production during carrot fermentation. At the same time, ethanol could also have been produced with less extent by heterofermentative LAB such as *Leuconostoc* spp. Our findings for ethanol are generally in good agreement with the results obtained by Tanguler and Erten and Ozturk.

In the present study, total and reducing sugar levels were observed between 90 to 200 mg/L and 13.65 to 15.25 mg/L, respectively. Their amounts were

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**Table 1**—General composition of ready-to-serve shalgam

| Samples | T  | S  | N  | L  | S  |
|---------|----|----|----|----|----|---|
| **Chemical properties** |    |    |    |    |    |---|
| Total acidity (gm/L) | 7.25<sup>b</sup> | 7.65<sup>a</sup> | 7.75<sup>a</sup> | 7.15<sup>b</sup> |    | *  |
| Lactic acid (gm/L) | 6.02<sup>a</sup> | 6.3<sup>a</sup> | 6.23<sup>a</sup> | 5.6<sup>b</sup> |    | *  |
| Acetic acid (gm/L) | 0.36 | 0.55 | 0.50 | 0.48 | ns |    |
| pH | 3.52<sup>ab</sup> | 3.48<sup>ac</sup> | 3.45<sup>c</sup> | 3.53<sup>a</sup> |    | *  |
| Ethanol (gm/L) | 3.42<sup>ab</sup> | 3.0<sup>b</sup> | 3.37<sup>ab</sup> | 3.72<sup>a</sup> |    | *  |
| Total sugar (mg/L) | 200 | 95  | 90  | 155 | ns |    |
| Residual sugar (mg/L) | 14.95 | 15.25 | 15.15 | 13.65 | ns |    |
| Total solid (gm/L) | 23.55 | 23.6 | 23.8 | 22.65 | ns |    |
| Ash (gm/L) | 14.12<sup>c</sup> | 15.30<sup>b</sup> | 15.30<sup>b</sup> | 16.25<sup>c</sup> | ** |    |
| Salt (gm/L) | 12.00 | 13.35 | 13.30 | 13.49 | ns |    |
| L* | 20.98 | 21.41 | 24.83 | 24.30 | ns |    |
| a* | 52.31 | 52.01 | 56.47 | 56.22 | ns |    |
| b* | 35.79 | 36.51 | 42.34 | 41.57 | ns |    |
| Hue | 34.38<sup>b</sup> | 35.07<sup>ab</sup> | 36.86<sup>a</sup> | 36.45<sup>a</sup> |    | *  |
| Chroma | 63.38 | 63.55 | 70.58 | 69.93 | ns |    |

*: as lactic acid, <sup>a</sup>: T: 3 cm black carrot; S: 6 cm black carrot; N: 9 cm black carrot; L: lengthwise cut black carrot; S: Significance, ** and * display the significance at 1% and 5% by LSD, respectively. a–c Values not sharing the same superscript letter within the horizontal line are different according to Duncan test. ns: not significant.
determined below 200 mg/L, showing that sugars were fermented. Similar low levels were reported by Gunes. However, Tanguler reported slightly higher values.

One of the important quality characteristics of shalgam is colour. L* is called lightness index and ranges from 0 (completely opaque) to 100 (completely transparent). In other words, an increase in L* value points the loss of the colour. As can be seen from Table 1, L* value was determined as 20.98 and 24.83 in the shalgams. It could be said that, L* value increased with the increasing size of black carrot. a* is a redness index and the highest value was determined as 56.47 in sample N. While a* represents the red colour, yellow colour is represented by b* value. b* value also increased with the increasing size of black carrot and found between 35.79 to 42.33. All parameters were determined as positive. For this reason, it could be said that there was yellowness in red colour. A parameter frequently used to characterise colour in food products is hue value. In addition, it is also used for comparison and evaluation of colour parameters in various vegetables and fruits. Hue value ranged from 34.38 to 36.86. Chroma is a parameter that indicates the contribution of a* (redness) to b* (yellowness). Values of Chroma were determined as 63.38 and 70.58. Gunes reported slightly lower values for L*, a*, b*, Hue and Chroma in ready to serve shalgams. The results showed that the effect of black carrot size had no significant effect on L*, a*, b* and Chroma values.

**Sensory analysis of ready-to-serve shalgam**

Shalgam is widely consumed with food and as a refreshing beverage. Shalgam complements the local tastes. It completes them in terms of taste. The effect of black carrot size in terms of sensory evaluation was analysed by thirteen assessors. According to the descriptive test (results not shown), sample T received the highest score (7.77) for colour. On the other hand, colour score of sample N was significantly lower than that of other samples. The taste of samples was affected from the addition of different size of black carrot. While the samples T (7.33) and S (7.31) received the best preference for the taste, sample N (6.73) received the lowest score. When the smell of shalgam samples by the panelists evaluated, the highest score was obtained with sample T (7.17). On the other hand, the most preferred sample, after sample T, was found as sample L by panelists. However, the lowest score was determined as 6.34 for sample N. The highest overall impression was obtained from sample T (7.67) and it was scored slightly higher than samples L (7.1) and S (7.08).

**Table 2—Sensory analysis of shalgam with the addition of black carrot at different size**

<table>
<thead>
<tr>
<th>Points of classification</th>
<th>3 cm</th>
<th>6 cm</th>
<th>9 cm</th>
<th>(P &lt; 0.01)</th>
</tr>
</thead>
<tbody>
<tr>
<td>T: 3 cm black carrot</td>
<td>a</td>
<td>b</td>
<td>c</td>
<td>20 &lt; 24 &lt; 28</td>
</tr>
<tr>
<td>S: 6 cm black carrot</td>
<td>b</td>
<td>c</td>
<td>a</td>
<td></td>
</tr>
<tr>
<td>N: 9 cm black carrot</td>
<td>c</td>
<td>a</td>
<td>b</td>
<td></td>
</tr>
</tbody>
</table>

In contrast, sample N which was added 9 cm size of black carrot ranked the worst overall impression. It could be said that increasing size of black carrot decreased to overall impression of shalgams. However, black carrot size had no statistically significant effect at P>0.05 on colour, smell, taste and overall impression (Table 2).

Shalgam samples were ranked from the most preferred to least preferred one with ranking test (Table 2). Statistical analysis of ranking test revealed that there was a significant (P<0.01) effect on the addition of different size of black carrot. From the point of sensory evaluation with ranking test, addition of 3 cm size of black carrot was found to be the one of the most preferred samples.

**Conclusion**

Recently, there is a growing interest on shalgam consumption. This work was prepared in order to determine the suitable black carrot size on the quality of shalgam. After fermentation completed, the amounts of total acidity, anthocyanin content and phenolic composition were determined between 7.15 to 7.75 gm/L, 120.18 to 145.6 mg/L and 23.30 to 28.89, respectively. This research demonstrated the smaller size of black carrot usage favourably affected the anthocyanin content, phenolic composition and sensory properties of shalgam and especially 3 cm size of black carrot can be used for the production of shalgam.

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