



## A Research on the Composition of Pomegranate Molasses

Bige İncedayi<sup>1\*</sup>, Canan Ece Tamer<sup>1</sup>, Ömer Utku Çopur<sup>1</sup>

<sup>1</sup>Department of Food Engineering, Faculty of Agriculture, Uludag University,  
16059. Bursa, Turkey.

\*e-posta: bige@uludag.edu.tr

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**Abstract:** In this study, seven different brands of pomegranate molasses purchased from the local markets of Bursa, Turkey, were analyzed for the chemical components, some of which are important for nutrition and health. Four samples were found to contain glucose syrup, one of which was found to have very high amount of hydroxymethylfurfural (1524.98 mg/kg). Analyses showed that protein, invert sugar, total sugar and total polyphenol contents of the samples were ranged 0.08-1.54 g/100g, 21.60-57.60 g/100 g, 44.80 - 65.3 g/100 g and 551.61-9695.17 mg/kg, respectively, while the antioxidant activity of the samples were found to be between 0 to 46.31%. The samples were also found rich in minerals e.g. potassium (450-4700 mg/100g), calcium (71.88-1803.63 mg/100g), magnesium (7.48-409.10 mg/100g) and iron (1.05-22.99 mg/100g). As some of the components were found in more amounts than the limits of the relevant standard, it is suggested that the production techniques for pomegranate molasses should be improved and optimized.

**Key Words:** Pomegranate, Pomegranate Molasses, Polyphenols, Antioxidant Activity.

### Nar Ekşilerinin Bileşimi Üzerine Bir Araştırma

**Özet:** Bu çalışmada, Bursa piyasasındaki marketlerden temin edilen 7 farklı marka nar ekşisinin kimyasal bileşimi ve insan beslenmesi ve sağlığı açısından önem taşıyan bazı bileşenler analiz edilmiştir. Sonuçta, 4 farklı marka nar ekşisinin glikoz şurubu içerdiği; bunlardan birinin oldukça yüksek hidroksimetilfurfural içeriğine (1524.98 mg/kg) sahip olduğu ortaya konmuştur. Aynı zamanda nar ekşilerinin 0.08-1.54 g/100g arasında protein, 21.60-57.60 g/100 g arasında indirgen şeker, 44.80-65.3 g/100 g arasında toplam şeker, 551.61-9695.17 mg/kg arasında toplam polifenol içerdiği; % 0 - 46.31 arasında antioksidan aktiviteye sahip olduğu saptanmıştır. Nar ekşilerinde yüksek düzeyde mineral madde bulunmuş, örneklerin sırasıyla potasyum (450-4700 mg/100g), kalsiyum (71.88-1803.63 mg/100g), magnezyum (7.48-409.10 mg/100g) ve demir (1.05-22.99 mg/100g) yönünden zengin olduğu ortaya konmuştur. Bu çalışmadan elde edilen verilere göre, piyasada bulunan nar ekşilerinin bazı bileşenler bakımından ilgili standartta yer alan limitlere uygunluk göstermediği; dolayısıyla üretim tekniklerinin iyileştirilmesi ve optimize edilmesi gerektiği sonucuna varılmıştır.

**Anahtar Kelimeler:** Nar, Nar Ekşisi, Polifenoller, Antioksidan Aktivite.

## Introduction

Pomegranate is consumed both as fresh fruit and fruit juice. It is also being used in the production of jam, wine, liqueur, food coloring agent and flavor enhancer. The kernels are also used as a garnish for desserts and salads (Al-Maiman and Ahmad, 2002). The pomegranate molasses is commonly used in salads and many dishes in Turkey (Marti et al., 2001; Poyrazoğlu et al., 2002; Altan and Maskan, 2005; Maskan, 2006).

Turkey is one of the leading producers of pomegranate e.g. 127.760 tons in 2008 (Anonymous, 2009). In recent years, pomegranate has become a widely known product. The production growth, consumption and trading volume have been increased because of the significant developments in growing techniques, nutritional technologies, storage and transportation means (Vardin and Abbasoğlu, 2004).

Traditional methods are still being used to produce pomegranate molasses. It is concentrated simply by boiling, without the addition of further sugar or other additives (Tekeli, 1965; Gökçen et al., 1982). A typical processing requires cleaning, crushing, extraction, filtration, clarification and evaporation in open vessel or under vacuum, though different type of fruits requires different production methods (Vardin and Abbasoğlu, 2004; Kaya and Sözer, 2005).

Pomegranate is a rich source of anthocyanins, ellagitannins and other phenolic compounds which are already proved to have antioxidant and antitumoral activity (Perez-Vicente et al. 2004). Major hydrolysable tannins in pomegranates are gallotannins, ellagic acid tannins and gallagyl tannins (these are termed as “punicalagin”s). These compounds are found abundantly in the pomegranate juice (< 2g/L) (Karaali et al., 2006). It is also stated that punicalagins have the highest antioxidant activity (Seeram et al., 2005). De Nigris (2005) reported that the antioxidant level in pomegranate juice was higher than found in other fruit juices, e.g. blueberry, cranberry, orange, and red wine. Recently, the bioavailability of these compounds has been studied in vitro and in vivo analyses. Pomegranate juice has also important clinical implications, and it has been recommended in the treatment of AIDS (Perez-Vicente et al., 2004). Researchers found that daily consumption of pomegranate juice may improve stress-induced myocardial ischemia in patients who have coronary heart disease and they also reported that the pomegranate juice not only prevented hardening of the arteries by reducing blood vessel damage, but also reversed the progression of this disease (Sumner et al., 2005; De Nigris, 2005). According to Mori-Okamoto et al. (2004), kernels of pomegranate are good source of phyto-estrogens. By strong antioxidant and anti-inflammatory effects pomegranate juice suppresses cancer activity. The polyphenolic phytochemicals in the pomegranate can play an important role in the modulation of inflammatory cell signaling in colon cancer cells (Adams et al., 2006). Pomegranate fruit extract (PFE) possesses remarkable antitumor-promoting effects in mouse skin. Malik et al. (2005) suggested that pomegranate juice may have cancer-chemopreventive as well as cancer-chemotherapeutic effects against prostate cancer in humans. Polyphenols also have neuroprotective effects. Hartman et al. (2006) reported that pomegranate juice had beneficial effect on animal model of Alzheimer's disease.

The composition of the fruit varies with type, growing conditions, climate, maturity and storage conditions. The edible part of pomegranate contains significant amounts of acids, sugars, vitamins, polysaccharides, polyphenols and minerals (Cemeroğlu et al., 1988; Cemeroğlu et al., 1992; Ünal et al., 1995; Melgarejo et al., 2000). Pomegranate molasses is

a highly nutritive product because the product is a concentrate and specially the presence of high mineral contents makes it more nutritious. The strong antioxidant activity of it is also important for human health. Composition of pomegranate (Anonymous, 2006), composition of natural pomegranate juice (Velioglu et al., 1997) and the general characteristics of pomegranate molasses according to Turkish standard of pomegranate molasses (TSE 12720) were shown in Tables, 1, 2, and 3 respectively.

## Material and Methods

In this study, seven different market brands of pomegranate molasses were analyzed. The samples were analyzed as triplicate at room temperature. Water soluble dry matter, viscosity, the presence of glucose syrup, total acidity, pH, hydroxymethylfurfural (HMF), protein, invert and total sugars, total polyphenols, antioxidant activity and some minerals (Fe, K, Ca, Mg) of the samples were determined.

The water soluble dry matter content (brix) of the samples was expressed as g/100g by using an Abbe refractometer (Cemeroğlu, 1992). Presence of glucose syrup was verified qualitatively 10 g of sample was diluted with 10 mL distilled water then filtered. 1-2 drops of 10% ammonium oxalate solution were added. After heating for a short time, activated carbon included and again it was heated. Solution was filtered and 2 mL of the filtrate was mixed with 2 drops of 37.5 % HCl and 20 mL 96 % ethyl alcohol. If the solution contained glucose syrup, solution become blurred the color changed to white (Cemeroğlu, 1992). Viscosity was measured in mPa.s using a rotary viscosimeter (NDJ-1, Shanghai Precision & Scientific Inst. Co. Ltd.). Total acidity was determined as citric acid, by titrating samples against NaOH solution of known normality (AOAC, 1984). pH was measured potentiometrically by a Nel-pH 890 model pH meter. HMF was determined by measuring the color variation of the samples by barbituric acid and p-toluidine, spectrophotometrically (Cemeroğlu, 1992). Protein was identified as nitrogen, by burning and distilling fresh samples (AOAC, 1984). Invert and total sugars were determined by the Luff-Schoorl method (Uylaşer and Başoğlu, 2000). The total polyphenol content was calculated via the gallic acid curve of the spectrophotometric measurement result of the absorbance of the blue color at 765nm, which is the result of the redox reaction of the reduction of the Folin-Ciocalteu reagent by phenolic compounds and turning into oxidized forms in an alkali medium (Spanos and Wrolstad, 1990). The results were expressed as mg/100g dry matter. Antioxidant activity was determined spectrophotometrically by using the 2,2-diphenylpicrylhydrazyl (DPPH) radical. In this method, extracted samples were reacted with the radical solution for 30 minutes at room temperature and the absorbance at 517 nm was measured, and the inhibition percentage of 0.1 mM DPPH free radical were calculated (Moon and Terao, 1998). Minerals that are present in significant amounts were determined according to the wet ashing method in the TS 4887-05/1986 by using Shimadzu AA 6701 F atomic absorption spectrometer and expressed as mg/100g (Anonymous, 1986).

The statistical analyses of the results were studied by the MSTAT-C software tool, by using ANOVA and LSD, within 95% confidence.

**Table 1.** Composition of pomegranate (Anonymous, 2006)

<b>Components</b>	<b>Amount</b>
Water	80.97 g/100g
Protein	0.95 g/100g
Carbohydrate	17.17 g/100g
Total sugar	16.57 g/100g
<b>Minerals</b>	
Calcium, Ca	3 mg/100g
Iron, Fe	0.30 mg/100g
Magnesium, Mg	3 mg/100g
Phosphorus, P	8 mg/100g
Potassium, K	259 mg/100g
Sodium, Na	3 mg/100g

**Table 2.** Composition of natural pomegranate juice (Velioglu et al., 1997)

<b>Components</b>	<b>Mean (n=120)</b>
Relative density (20/20°C)	1.068
Brix	16.30
Titration acidity (as citric acid) (g/L)	8.58
pH	3.53
Ash (g/L)	3.907
Formol number (mL 0.1 N NaOH/100 mL)	9
Glucose (g/L)	64.80
Fructose (g/L)	71.50
Glucose/Fructose	0.92
Invert sugar (g/L)	153.20
Citric acid (g/L)	5.39
L-Malic acid (g/L)	0.87
D-isocitric acid (mg/L)	54.90
Chloride (mg/L)	500
Proline (mg/L)	7.70
Sulphate (mg/L)	133.4
Phosphate (mg/L)	270
Calcium (mg/L)	20
Magnesium (mg/L)	45
Sodium (mg/L)	9
Potassium (mg/L)	1209

**Table 3.** General composition of the pomegranate molasses (Anonymous, 2001)

Components	Amount
Water Soluble Dry Matter, %, minimum	68.0
Titration acidity (as citric acid), %, minimum	7.5
pH	3.0
HMF, mg/100g, maximum	50
Saccharose	Not allowed

The aim of this study was analyzing of different market brands of pomegranate molasses for composition and the components that are of importance for nutrition and health.

### Results and Discussion

Physical and chemical analyses results of pomegranate molasses and quantities of some minerals in the samples were shown in Table 4 and Table 5 respectively. The differences between the components of the samples were found to be statistically significant ( $P < 0.05$ ).

The water soluble dry matter content of pomegranate molasses was found to vary between 58.25-74.50 g/100g. Only one sample had water soluble dry matter content (58.25 g/100 g) which was less than the specified amount (68%) of the standard (Anonymous 2001). Yılmaz et al. (2007) have found the moisture content of the pomegranate molasses as 24.4%. Vardin et al. (2008) reported that brix values ranged from 50.1 to 77.3 for pomegranate juice concentrate.

Glucose syrup was determined in four of the samples. It is suspected that this was a mean to increase the dry matter content.

Viscosities of the samples were found between 200 and 1800 mPa.s. Yılmaz et al. (2007) reported that pomegranate molasses exhibits a Newtonian flow behavior.

Pomegranate molasses samples were found to have total acidity between 5.11-9.83 g/100 g. According to the pomegranate molasses standard, titration acidity must be minimum 7.5% (Anonymous 2001). In four of the samples acidity was below than the limit. Vardin et al. (2008) determined total titratable acidity (% citric acid) in pomegranate juice concentrate between 5.80 and 14.27 %. The variety of the pomegranate used and evaporation applied in the production caused an increase in the acidity of some samples.

pH of the samples changed between 0.87 and 1.98. Yılmaz et al. (2007) measured pH of the pomegranate molasses as  $1.74 \pm 0.09$ . Vardin et al. (2008) determined pH of the pomegranate juice concentrate as 1.34 – 2.90.

The hydroxymethyl furfural (HMF) content is important since it indicates the degree of heating of the treated products during processing and the quantification of this compound is considered as a quality parameter for concentrated food products (Marcy and Rouseff, 1984; Babsky et al., 1986; O'Brien and Morrisey, 1989; Ramirez-Jimenez et al., 2000; Rada-Mendoza et al., 2002; Kus et al., 2005). One sample contained a very high amount of

HMF as 1542.98 mg/kg. All but three samples had HMF contents that were much higher than the standard value (50 ppm). It was suspected that this was the direct result of open air heating of the products for a long time. Since the pH values were low (0.87-1.90), this high acidity might accelerate the formation of HMF. Kus et al. (2005) obtained a higher range of HMF concentrations (514 - 3500 ppm at 68 - 71 Brix) in boiled pomegranate juices and they determined that the formation of HMF in boiled juice is highly dependent on pH. Orak (2008) determined extremely higher amount of HMF as 4003 ppm in traditionally produced and two months stored concentrated pomegranate juice.

**Table 4.** Physical and chemical analysis results of the pomegranate molasses\*

Sample	Water Soluble Dry Matter (g/100g)	Presence of glucose syrup	Viscosity (mPa.s)	Total acidity (g/100 g) (as citric acid)	pH	HMF (mg/ kg)	Protein (g/100 g)	Invert sugar (g/100 g)	Total sugar (g/100 g)	Total polyphenol (mg GAE/kg)	Antioxidant activity (%)
1	74.25 ab	-	1360 c	7.72 b	1.35 d	498.43 c	0.40 d	57.60 a	65.30 a	551.61 g	26.76 b
2	69.25 d	+	500 e	5.76 e	1.44 c	40.06 e	0.40 d	28.80 d	58.80 b	2771.31 f	20.36 d
3	58.25 e	+	200 g	5.11 f	1.90 b	104.58 d	0.55 c	26.40 e	46.80 d	4968.94 d	1.42 f
4	72.25 bc	+	1500 b	6.30 c	0.95 e	590.49 b	0.29 e	22.80 f	60.30 b	5149.87 c	22.04 c
5	68.50 d	-	250 f	7.76 b	1.98 a	18.56 f	1.21 b	40.80 c	44.80 d	9695.17 a	46.31 a
6	70.50 cd	+	1150 d	6.14 d	0.87 f	45.78 e	0.08 f	21.60 g	54.30 c	9227.40 b	0 g
7	74.50 a	-	1800 a	9.83 a	1.43 c	1542.98 a	1.54 a	45.60 b	46.50 d	3653.89 e	17.36 e

\* Different letters in the same column indicate significant differences  $P < 0.05$ . LSD, least significant difference.

**Table 5.** Quantities of some minerals in the pomegranate molasses (mg/100 g)\*

Sample	Fe	K	Ca	Mg
1	4.79 c	762 f	400.81 d	84.96 e
2	4.72 c	2720 c	318.07 e	130.83 d
3	4.06 d	2627 d	459.03 c	204.11 c
4	1.05 e	1102 e	86.65 f	27.35 f
5	14.18 b	4700 a	1803.63 a	409.10 a
6	0.25 f	450 g	71.88 g	7.48 g
7	22.99 a	3861 b	617.66 b	238.90 b

\* Different letters in the same column indicate significant differences  $P < 0.05$ . LSD, least significant difference.

Protein content of the samples was found to range in between 0.08-1.54 g/100 g. Yılmaz et al. (2007) determined the protein content of the molasses as  $0.23 \pm 0.06$  and according to the researchers, filtration and clarification of pomegranate juice during commercial processing may be the main reason for low protein content of pomegranate

molasses in comparison to fresh pomegranate. According to Anonymous (2006), protein content is 0.95 g/100g for pomegranate. The protein content of some samples is increased percentage wise via concentration of the products, while some had lower via denaturation of proteins through heating.

Invert sugar and total sugar were determined between 21.60 - 57.60 g/100g and 44.80 - 65.30 g/100g, respectively. It was observed that the acidity of pomegranate molasses samples was very high, while it was found that a significant part the total sugar of the samples with unknown glucose syrup content was subjected to inversion via thermal processes. Poyrazoğlu et al. (2002) found total sugars as 148.75 g/L by analyzing the composition of freshly squeezed pomegranate juices of 13 different types. Orak (2008) determined invert sugars in concentrated pomegranate juice as 46.4%. Evaporation of water during the production of molasses increases the concentration of carbohydrates in the final product (Yılmaz et al., 2007).

Total polyphenol contents were found to be in between 551.61 and 9695.17 mg/kg. Karaali et al. (2006) identified this value to be 3100 mg/kg in pomegranate pulp and 1860 mg/L in juice. Alper et al. (2005) determined the total phenolic substance content of pomegranate juice as 2294.14 mg/L. Markh and Lysoger (1973) reported that the fruit contains 0.22-1.05 % anthocyanins and polyphenols from various phenolic acids. Total phenolic content of pomegranate molasses was very high in the research of Yılmaz et al. (2007) and reported as 52.6 mg gallic acid equivalent (GAE)/g. Orak (2008) found the total phenolics to be 9870 µg/ml (GAE) in pomegranate juice concentrate.

Phenolic compounds give the color, astringency and bitter taste to pomegranate juice. Also these compounds cause cloudiness during concentration process and storage (Bayındırlı et al., 1994; Alper et al., 2005). Since pomegranate molasses is a concentrated product, it was anticipated that the phenolic substances would increase by percentage, despite losses during thermal processing. In some samples, this result was observed, while some had lower results due to the composition of the raw material.

Antioxidant activity was measured spectrophotometrically by using the 2,2-diphenylpicrylhydrazyl (DPPH) radical within 0 - 46.31 % in molasses samples that were diluted to 14 brix. It was the result of the long term thermal processing of samples with lower antioxidant activity. The research of Singh et al. (2002) revealed this value as 23.2% (at 100 ppm) in the seed extracts. Antioxidant activity of commercial pomegranate juice was also determined by Gil et al. (2000) and Karaali et al. (2006) as 26.4 % and 54.8 % (at 5 mg/mL) respectively. Orak (2008) determined the antioxidant activity (percentage of inhibition of on peroxidation in a linoleic acid system) of pomegranate juice and concentrated pomegranate juice as 79.06% and 85.91% respectively. It is stated that the higher antioxidant activity of concentrated pomegranate juice is due to the phenolic content in spite of anthocyanin being destroyed by the effect of heating. The high antioxidant activity of concentrated pomegranate juice might be appeared due to a high phenol concentration.

It was revealed that the samples were rich in minerals, especially in potassium (450 - 4700 mg/100g), calcium (71.88 - 1803.63 mg/100g), magnesium (7.48 - 409.10 mg/100g) and iron (1.05 - 22.99 mg/100g). Yılmaz et al. (2007) determined some minerals in pomegranate molasses such as potassium (20 mg/kg), calcium (280 mg/kg), magnesium (28 mg/kg), iron (16 mg/kg), phosphorus (16 mg/kg) and zinc (7 mg/kg). According to the

results of the elemental analysis of concentrated pomegranate juice; sodium, potassium, iron, magnesium, zinc, copper and lead were determined as 326.25, 1685.80, 5.88, 51.25, 1.76, 0.70 and 0.05 mg/kg respectively. Cadmium, mercury, manganese, cobalt, arsenic and boron were not determined. The mineral contents of pomegranate juice were variously affected from the concentration process. The sodium, iron, zinc, copper and lead amounts determined were lower than those of pomegranate juice. Percentages of losses were calculated as 44%, 71%, 62%, 68%, and 69%, respectively. On the other hand, the potassium and magnesium contents increased during concentration as 2% and 24%, respectively (Orak, 2008).

The difference between the results might be originated from variety of the fruits used and processing steps applied such as clarification and filtration. Fadavi et al. (2005) reported in the work on the composition of 10 different types of pomegranates grown in Iran that potassium was the highest mineral content (80.0–160.6 mg/100g) and other minerals like sodium (11.3 – 54.5 mg/100g), calcium (13.05 – 30.60 mg/100g), magnesium (2.75 – 5.20 mg/100g), iron (0.03 – 0.21 mg/100g), zinc (0.037 – 0.084), copper (0.013 – 0.081 mg/100g) and manganese (0.012 – 0.021 mg/100g) were found in various amounts. It was clearly seen that the pomegranate molasses was richer in minerals than the fruit itself, since it was a concentrated product.

## Conclusion

Pomegranate molasses is a highly nutritive product, since it is processed as concentrate. Especially the presence of mineral content of the fruit are in higher concentration of the molasses, gives it an exceptional nutritional characteristic. The results showed that pomegranate molasses are also rich in phenolics with high health and nutritional values, if it is produced in the correct way. It was found to have good antioxidant activity, an important aspect for human health. From the results of the analysis, it was found that some components were within the limits of the standard (Anonymous 2001), while others (HMF, pH, total acidity) were higher. Based on the results of this study, it is required to improve the production techniques of pomegranate molasses. Especially to limit the HMF content with adverse health effects, which was the result of long term thermal processing, it is required to utilize vacuum for the concentration process. In order to detect adulteration of the pomegranate molasses, analytical techniques such as fourier transform infrared (FTIR) spectroscopy and chemometrics should be used and pomegranate juice concentrate standard should be revised and detailed.

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