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# Increasing Shelf Life of Sous-Vide Cooked Rainbow Trout by Natural Antioxidant Effective Rosemary: Basic Quality Criteria

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# ABSTRACT

In the study, the rosemary was applied as a natural antioxidant after grinding. Dust ground rosemary were applied on the fillet surface (0.1% percent of the fillets weight), and then were packaged with the vacuum and sous-vide processed. Every five days, chemical composition, pH, TVB-N, TBARS analyzes and sensorial assessment was carried out until the spoilage. The ash, moisture, protein, and lipid content of raw rainbow trout was 1.63±0.11%, 78.36±0.18%, 16.49±0.20%, and 3.37±0.30%, respectively. The pH value raw fish was 6.29±0.01. TVB-N values were determined between 12.86±0.15 and 21.94±0.29 mg/100g. in RAG. The highest TBARS value was determined as 0.80±0.03 in RAG. Odor parameter was effective to deteriorate the decision of panelists for all the samples The vacuum packaged fresh fish (VFF) was spoiled in the 10<sup>th</sup> days while, the rosemary untreated group (RUG) was spoiled in the 40th days, and rosemary applied group (RAG) was spoiled in the 45<sup>th</sup> days. With the rosemary treatment, the shelf life and acceptability of the samples was extended to at least 5 days. Also, applied rosemary as the natural antioxidant had no negative effects with regard to sensory and nutritional criteria.

Keywords: TVB-N, TBARS, sensorial assessment, chilled storage, spoilage

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#### Doğal Antioksidan Etkili Biberiye ile Sous-Vide Pişirilmiş Gökkuşağı Alabalığının Raf Ömrünün Artırılması: Temel Kalite Kriterleri

**Öz:** Çalışmada, öğütülmüş biberiye doğal bir antioksidan olarak uygulandı. Yüzeyine toz halindeki biberiye uygulanan alabalık filetoları, vakumda paketlendikten sonra sous-vide işlemine tabi tutuldu. Örnekler bozuluncaya kadar beş günde bir kimyasal kompozisyon, pH, TVB-N, TBARS analizleri ve duyusal değerlendirme yapıldı. Çalışmada kullanılan gökkuşağı alabalığı filetolarının kül, nem, protein ve lipid içeriği sırasıyla  $\%1,63\pm0,11$ ,  $\%78,36\pm0,18$ ,  $\%16,49\pm0,20$  ve  $\%3,37\pm0,30$  olarak belirlendi. Taze balık etinin pH değeri  $6,29\pm0,01$ 'di. RAG'de TVB-N değerleri  $12,86\pm0,15$  ile  $21,94\pm0,29$  mg/100g arasında tespit edildi. En yüksek TBARS değeri RAG'de  $0,80\pm0,03$  olarak belirlendi. Koku parametresi, tüm örnekler için panelistlerin bozulma kararı vermesinde belirleyici oldu. Vakum paketlenmiş taze balık (VFF) 10. günde bozulmuşken, biberiye ile muamele edilmemiş grup (RUG) 40. günde bozuldu ve biberiye uygulanan grup (RAG) 45. günde bozuldu. Biberiye muamelesiyle, numunelerin raf ömrü ve kabul edilebilirliği en az 5 güne kadar uzatılmıştır. Ayrıca, yapılan duyusal analizlere göre doğal antioksidan olarak uygulanan biberiyenin duyusal ve beslenme kriterleri açısından olumsuz bir etkisi olmadığı belirlendi.

Anahtar Kelimeler: TVB-N, TBARS, duyusal değerlendirme, soğuk depolama, bozulma

#### Alıntılama

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# Introduction

The desire for a healthy and long life is a commonly accepted merit for humankind. The numbers of health problems are rising in both developed and developing countries with increasing population. The focused research on plant-based diets and functions of phytochemicals for nutrition and ageing brought clarity to progress (Poyrazoğlu and Velioğlu 2005; Meskin et al. 2008). The studies showed polyphenols and carotenoids have riskreducing effects of certain cancers and cardiovascular diseases besides health-promoting properties (Meskin et al. 2008). So, those adverse effects were seen of artificial antioxidants, the need for natural antioxidants has increased and the work in this area has focused on plant-derived antioxidants (Diri 2006a). Recent years, a lot of studies were carried out about antioxidant and antimicrobial properties of rosemary had significant phytochemicals resource (Genena et al. 2008; Klančnık et al. 2009; Abramovic et al. 2012; Altınelataman et al. 2015; Azizkhani and Tooryan 2015).

Fisheries product is a significant natural source of unsaturated fatty acid like as DHA (22:6n-3) and EPA (20:5n-3) (Eseceli et al. 2006; Nollet and Toldrá 2010). But, lipid peroxidation is the most restrictive factor for shelf life of the fish oil. Also, it was admitted the most important factor for shelf life determination of those product if the microbiological effects omitted. Reaction products of lipid oxidation have an adverse effect on the sensorial properties of fish products. The secondary volatile products of oxidation, especially derived from unsaturated fatty acid (like as n-3 group) have adverse effect on the general appreciation even in low quantities (Nollet and Toldrá 2010).

Nowadays, consumers mostly prefer the food, that with fresh tasty, high-quality, low salt contain, non additives, and in a short prepared time. In accordance with those demands, had significantly increased of the production of ready to eat foods, of which minimally processed, and shelf life prolonged by cooling, and heatable with microwave owen, in the European and American markets. Such processed products, also consists sous-vide processed products, are called "new generation chilled products" (Novak et al. 2003).

The sous-vide technology is a pasteurization application for vacuumed foods. Using this technique, ensured the food shelf life with the cold storage, and therefore it has increased importance day to day. To apply this technology, the foods cook in a vacuum package with or without spices or sauces, and could be serviced after by heating with hot water or microwave oven. In this respect, the sous-vide technology or vacuum packed cooking technique is an application that can service to the catering sector. Also, for the preservation of rapidly perishable foods is very convenient. Because fisheries products are easily perishable foods, so, it is reported to be suitable for make packaging using this technology, by researchers (Mol and Özturan 2009).

There were reported that no adequate scientific studies about sous-vide treated fisheries products (Fagan and Gormley 2005; Mol and Özturan 2009). Also, the conducted studies were about microbiological properties of products. According to the results of those studies, any problem microbiologically was not seen, but the deterioration occured along the storage period came true on the sensorial properties (Bolton 1998; Kılınç and Çaklı 2001; Lindström et al. 2003; Novak et al. 2003; Garcia-Linares et al. 2004; Gonzales-Fandos et al. 2004; Gonzales-Fandos et al. 2005; Cobos and Diaz 2007; Diaz et al. 2008; Diaz et al. 2009; Shakila at al. 2009; Diaz et al. 2010;). So, in this study it was aimed to determination of changes on the quality properties, especially sensorial attributes, along the storage period. This study was carried out determination effects of rosemary as a natural antioxidant on the nutritional composition, and quality properties of rainbow trout, which cooked sous-vide method and cold stored.

# **Materials and Methods**

# Materials and sample preparation

This study was conducted on fresh, vacuumed, sous vide processed, and rosemary (Rosmarinus officinalis Linn. Lamiaceae) applied and sous vide processed fish groups. Totally 15 kg rainbow trout in the study were filleted, weighed, equally divided (150 g) (15 packets for every groups), and were kept in deep freezer (-18 °C) until usage (The average total length of the fish is 25 cm and the average weight is 250 gr). The rosemary is a plant, which have phenolic compounds. In this study, the rosemary (pine - leaf) was used as a natural antioxidant additive. The rosemary, reaped in a local garden, cleaned dust by the tap water. Than it was dried in the air flow and shadow place, and stored within nonpermeable bags in the cool and shadow place until use. The vacuum bags (cast PP) were provided by a commercial company (Apack Ambalaj Makine Sanayi ve Tic. Ltd. Sti. in Istanbul-Türkiye). O2 permeability of vacuum bag material is 22.56 cc/m<sup>2</sup>/day (24 °C), and one bag is 2.71 cc/day (24 °C). The CO<sub>2</sub> permeability of material is 95.64  $cc/m^2/day$  (24 °C), and one bag is 11.48 cc/day (24 °C). Also heat resistance of the material is -25/150 °C. Total thickness of one bag is 0.88 mm.

The fillets were melted under the tap water (the water temperature was 14 °C). Grounded rosemary (like dust) was weighed as 0.1% percent of the fillets weight (Selmi and Sadok 2008). Rosemary applied on the fillets surfaces then the fillets were put into the bag and vacuumed. The cooking was made in 85 °C benmari for 35 minutes (SVAC 1991; Peck 1997). Than, the packets were dipped in a bucket with 1/3 water and 2/3 iced, so rapidly cooled to 3-4 °C, in the 30 minutes. All packets were stored in the refrigerator at  $3\pm1$  °C until to spoil.

#### **Analytical methods**

Total antioxidant effect, free radical scavenging capacity and total phenolic content of rosemary was determined. Total antioxidant effect was determined by  $\beta$ -carotene-linoleic acid assey (Dapkevicius et al. 1998; Diri 2006a). Free radical scavenging capacity

was determined 1.1-difenil-2-pikrilhidrazil (DPPH system) (Tepe et al. 2005; Diri 2006b; Selmi and Sadok 2008). Total phenolic content was determined by Folin-Ciocalteu assay (Oktay et al. 2003; Diri 2006b).

The ash and total lipid analysis were conducted according to Lovell (1981). Moisture content was determined by AND MX-50 automatic moisture analysis (Oven Drying Method). Protein contents were determined by Velp UD-20 protein predigested unit and fully automatic Velp UDK 142 distillation unit by Kjeldahl methods (Nx6.25) (AOAC 2000). pH of the samples was measured with automatic pH meter (WTW 320 set digital) diluted by 1/10 (w/v) ratio (Varlık et al. 2007). Total volatile basic nitrogen (TVB-N) determined by a method had informed by Nicholas (2003). The thiobarbituric acid-reactive substances (TBARS) were analyzed using a method modified by Erkan and Özden (2007). Fatty acids analysis was conducted by Perkin Elmer Clarus 500 (GC) in fresh fish and RAG. For this purpose a flame ionization detector (FID) and a silica AGE colon (30m x 0.32 mm ID x 0.25 μm, BP20 0.25 UM, USA) and Supelco FAME 37 mix was used.

#### Sensorial analyses

Sensorial analyses were conducted with a hedonic scale (Huss 1995; Altuğ and Elmacı 2005; Diaz et al. 2009; Shakila et al. 2009). In the every session, the experienced 10 panelists were assessed the micro-wave heated 100 g samples descriptive and affective tests for color, smell, taste, texture, appearance, juiciness, general appreciation. If the group score equal to 2 points or lower were accepted had spoiled (Huss 1995; Altuğ and Elmacı 2005; Diaz et al. 2009; Shakila et al. 2009).

#### Statistical analyses

The analyses were conducted for fresh fish (FF), vacuum packaged fresh fish (VFF), the rosemary untreated group (RUG), and rosemary applied group (RAG) samples to observe changes along the storage period in every 5 days. All analyses and measurements were made triplicate. The data analyzed by ANOVA (Özdamar 2001) using SPSS 15 (95% confidence interval). The appeared differences for the treatments (0<sup>th</sup> day) on the stored samples had determined with Duncan test. Also, of the rosemary treatment effects determined by t test between RUG and RAG along the storage period, and marked on the tables as + (significant), and - (insignificant).

# Results

The rosemary was used as a natural antioxidant in this study. Total antioxidant effect, free radical scavenging capacity and total phenolic content of rosemary was determined as  $97.76\pm1.92$  %,  $8.76\pm0.57$  %,  $128.95\pm0.00$  mg/l gallic acid equivalent, respectively.

No significant effects were determined related to vacuuming, sous vide treatment, or sous vide treatment with rosemary application to the ash content of FF, VFF, RUG, and RAG group (Table 1). Solely, a significant difference was seen between RUG and RAG on the 15<sup>th</sup> day. No significant differences were seen in the in-group values along the storage period.

The moisture content of samples had not effected by the treatment or application. The highest and the lowest moisture values were obtained in the  $35^{\text{th}}$  storage days (Table 2). The differences between in the groups were found significant in the RUG and RAG along the storage period (p≤0.05).

The protein was affected by vacuuming and rosemary application significantly (p<0.05). The protein value had shown differences statistically significant (p $\leq$ 0.05) for the RUG and RAG on the 15<sup>th</sup>, 30<sup>th</sup>, and 40<sup>th</sup> day (Table 3).

Significant differences statistically ( $p \le 0.05$ ) observed in the total lipid values for intragroup and among the groups (Table 4). But, a significant statistical difference (p > 0.05) was not seem between RUG and RAG.

The rosemary treatment affected the pH until to the 20<sup>th</sup> day (Table 5). Along the storage progresses, this effect was not observed. In the generally, the pH value of RAG had higher than RUG. The pH values of RAG had shown statistically significant change (p $\leq$ 0.05) in the intragroup.

The application of rosemary and than vacuuming has shown decrease effect in the initial value of TVB-N. The TVB-N value of RUG had higher than the fresh samples in the first day. Statistically significant difference (p $\leq$ 0.05) was seem between groups TVB-N value of both RUG and RAG (Table 6). But, application rosemary has statistically significant (p $\leq$ 0.05) effect on the TVB-N value on the 0<sup>th</sup> day and 30<sup>th</sup> day.

In the initially, the TBARS value was close the each other for all groups. However, the changes in the TBARS value was found to be statistically no significant (p>0.05) in 0<sup>th</sup> day. Significant difference statistically (p $\leq$ 0.05) had seen in the TBARS value of the intragroups and among the groups (Table 7).

The RAG group had preferred mostly for the color criteria. The smell criteria score was affective to taken in to the decision of spoilage of samples. The RUG achieved the highest taste score. When the sample had spoiled, the taste point has approached to the spoilage point, (Table 8).

Groups Days	FF	VFF	RUG	RAG	Sig. t test
0	1.63±0.11 <sup>a</sup>	1.62±0.02 Aa	1.86±0.45 Aa	1.73±0.23 Aa	-
5		1.57±0.02 <sup>A</sup>	1.85±0.03 <sup>A</sup>	1.82±0.12 A	-
10		1.65±0.03 <sup>A</sup>	$1.55 \pm 0.03$ <sup>A</sup>	1.61±0.13 <sup>A</sup>	-
15			$1.55 \pm 0.04$ <sup>A</sup>	1.94±0.05 <sup>A</sup>	+
20			1.78±0.13 A	1.93±0.14 A	-
25			$1.83 \pm 0.09$ <sup>A</sup>	$1.84{\pm}0.06$ <sup>A</sup>	-
30			1.75±0.03 <sup>A</sup>	$1.86{\pm}0.07$ <sup>A</sup>	-
35			1.85±0.01 <sup>A</sup>	$1.74{\pm}0.09^{\text{ A}}$	-
40			$1.74{\pm}0.06$ <sup>A</sup>	1.66±0.08 <sup>A</sup>	-
45				1.64±0.06 <sup>A</sup>	NT

Table 1. Ash contents of groups for storage days (%)

\* The same lowercase for treatment and the same capital letter for the storage period are statistically insignificant (p > 0.05). The effects of rosemary treatment were marked as + (significant), and - (insignificant). \*NT (Not Tested)

Table 2. Moisture contents of groups for storage days (%)

Groups Days	FF	VFF	RUG	RAG	Sig. t test
0	78.36±0.18 a	78.80±0.40 Aa	78.83±0.23 Aa	78.41±0.06 ABa	-
5		78.52±0.48 A	$76.69 \pm 0.82$ BE	79.60±0.10 <sup>A</sup>	-
10		78.69±0.16 <sup>A</sup>	$78.58 \pm 0.10^{\text{A}}$	$77.84{\pm}0.67^{\text{ B}}$	-
15			77.51±0.38 AB	78.39±1.14 AB	+
20			78.66±0.25 <sup>A</sup>	78.49±0.17 AB	-
25			78.56±0.25 <sup>A</sup>	79.72±0.02 <sup>A</sup>	-
30			$78.13\pm0.89^{\text{AB}}$	79.26±0.39 AB	-
35			76.55±0.89 <sup>B</sup>	79.79±0.34 <sup>A</sup>	-
40			78.33±0.31 AB	79.31±0.24 AB	-
45				78.86±0.21 AB	NT

\* The same lowercase for treatment and the same capital letter for the storage period are statistically insignificant (p > 0.05). The effects of rosemary treatment were marked as + (significant), and - (insignificant). \*NT (Not Tested)

Gr Days	roups FF	VFF	RUG	RAG	Sig. t test
0	16.49±0.20 <sup>ab</sup>	16.89±0.07 Aa	16.39±0.23 Aab	16.40±0.10 Ab	-
5		16.86±0.26 <sup>A</sup>	16.92±0.13 AB	16.54±0.13 AB	-
10		16.44±0.09 <sup>A</sup>	16.82±0.16 AB	16.95±0.28 <sup>BC</sup>	-
15			16.95±0.10 AB	16.59±0.06 AB	+
20			16.58±0.35 AB	16.93±0.37 <sup>BC</sup>	-
25			16.54±0.06 AB	16.53±0.05 AB	-
30			16.83±0.15 AB	17.72±0.11 <sup>D</sup>	+
35			17.03±0.05 <sup>в</sup>	16.91±0.19 <sup>BC</sup>	-
40			16.88±0.06 AB	17.35±0.09 <sup>CD</sup>	+
45				17.11±0.09 <sup>BC</sup>	NT

Table 3. Protein contents of groups for storage days (%)

\* The same lowercase for treatment and the same capital letter for the storage period are statistically insignificant (p > 0.05). The effects of rosemary treatment were marked as + (significant), and - (insignificant). \*NT (Not Tested)

Groups Days	FF	VFF	RUG	RAG	Sig. t test
0	3.37±0.30 <sup>a</sup>	3.31±0.21 Aa	3.29±0.30 <sup>Ba</sup>	3.39±0.17 Da	-
5		$3.42 \pm 0.82$ <sup>A</sup>	2.65±0.00 AB	$2.42\pm0.19^{\text{AB}}$	-
10		$3.48 \pm 0.18$ <sup>A</sup>	$3.05\pm0.57$ AB	$2.82 \pm 0.08$ <sup>BC</sup>	-
15			$2.59\pm0.09^{\circ}$ C	2.46±0.14 <sup>E</sup>	-
20			2.39±0.08 AB	2.99±0.25 <sup>CD</sup>	-
25			2.58±0.27 AB	1.97±0.17 <sup>A</sup>	-
30			$2.37 \pm 0.40$ AB	1.95±0.17 <sup>A</sup>	-
35			2.42±0.23 AB	$2.38 \pm 0.02$ AB	-
40			2.14±0.35 AB	1.95±0.17 <sup>A</sup>	-
45				2 69+0 07 <sup>BC</sup>	NT

 Table 4. Total lipid contents of groups for storage days (%)

\* The same lowercase for treatment and the same capital letter for the storage period are statistically insignificant (p > 0.05). The effects of rosemary treatment were marked as + (significant), and – (insignificant). \*NT (Not Tested)

Groups	FF	VFF	RUG	RAG	Sig. t test
0	6.29±0.01 <sup>a</sup>	6.30±0.27 <sup>Ba</sup>	6.46±0.01 Ab	6.54±0.02 <sup>Cc</sup>	+
5		6.39±0.01 <sup>C</sup>	6.61±0.01 <sup>B</sup>	6.13±0.07 <sup>A</sup>	+
10		6.05±0.01 <sup>A</sup>	6.76±0.01 <sup>D</sup>	6.84±0.01 <sup>G</sup>	+
15			$6.91 \pm 0.00^{E}$	$6.93 \pm 0.00$ H	+
20			6.71±0.01 <sup>C</sup>	6.73±0.01 <sup>F</sup>	-
25			6.70±0.01 <sup>C</sup>	6.69±0.01 EF	-
30			6.62±0.01 <sup>B</sup>	6.65±0.01 DE	-
35			6.61±0.01 <sup>B</sup>	$6.59 \pm 0.00$ CD	-
40			6.63±0.01 <sup>B</sup>	$6.59 \pm 0.00$ CD	+
45				6.46±0.01 <sup>B</sup>	NT

Table 5. The pH values of groups for storage days

\* The same lowercase for treatment and the same capital letter for the storage period are statistically insignificant (p > 0.05). The effects of rosemary treatment were marked as + (significant), and - (insignificant). \*NT (Not Tested)

	Table 6. The	TVB-N va	alues of group	os for storage	days (mg/100g)
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Groups Days	FF	VFF	RUG	RAG	Sig. t test
0	14.71±0.17 <sup>a</sup>	11.68±0.47 Ac	15.55±0.55 <sup>Aa</sup>	13.36±0.00 ABb	+
5		14.04±0.61 <sup>A</sup>	14.20±0.34 AB	13.36±0.67 AB	-
10		18.32±1.98 <sup>B</sup>	12.69±0.34 A	14.04±0.44 <sup>B</sup>	-
15			13.45±0.22 <sup>A</sup>	12.86±0.15 <sup>A</sup>	-
20			13.70±0.08 A	13.95±0.22 <sup>B</sup>	-
25			16.14±0.52 <sup>CD</sup>	16.56±0.22 <sup>D</sup>	-
30			17.31±0.83 <sup>D</sup>	13.70±0.08 AB	+
35			16.39±0.44 <sup>CD</sup>	15.72±0.30 <sup>CD</sup>	-
40			15.80±0.69 <sup>CD</sup>	15.13±0.15 <sup>C</sup>	-
45				21.94±0.29 <sup>E</sup>	NT

\* The same lowercase for treatment and the same capital letter for the storage period are statistically insignificant (p > 0.05). The effects of rosemary treatment were marked as + (significant), and - (insignificant). \*NT (Not Tested)

Days	Groups	FF	VFF	RUG	RAG	Sig. t test
	0	0.25±0.03 <sup>a</sup>	0.32±0.03 Aab	$0.35 \pm 0.04$ Ab	0.27±0.00 Aab	-
	5		$0.43\pm0.04$ A	0.31±0.08 <sup>A</sup>	0.11±0.00 A	-
	10		1.29±0.10 <sup>B</sup>	$0.43{\pm}0.07$ <sup>A</sup>	$0.55 \pm 0.03$ <sup>CD</sup>	-
	15			0.80±0.03 <sup>B</sup>	$0.36{\pm}0.07^{\text{BC}}$	+
	20			$0.92{\pm}0.05^{\text{BC}}$	$0.65 \pm 0.06$ DEF	+
	25			$1.00\pm0.05^{\circ}$ C	$0.78 \pm 0.05$ EF	+
	30			$0.93 \pm 0.04$ <sup>BC</sup>	$0.59{\pm}0.03$ DE	+
	35			$1.38\pm0.05^{\text{ D}}$	0.80±0.03 <sup>F</sup>	+
	40			1.01±0.04 <sup>C</sup>	$0.65 \pm 0.03$ DEF	+
	45				$0.75\pm0.17^{\text{ DEF}}$	NT

Table 7. The TBARS values of groups for storage days (µg MDA/g)

\* The same lowercase for treatment and the same capital letter for the storage period are statistically insignificant (p > 0.05). The effects of rosemary treatment were marked as + (significant), and - (insignificant). \*NT (Not Tested)

The texture score of VFF was decreased rapidly on 10<sup>th</sup> day. The same decrease was not seem in the other groups. The used technology was effective to the texture score of samples. The highest appearance score was obtained for RUG, and RAG on the  $15^{\text{th}}$  day.

The appearance score of RUG had higher than RAG's on the 20<sup>th</sup>, 25<sup>th</sup> and 30<sup>th</sup> days (Table 8). The significant differences ( $p \le 0.05$ ) had seen in the juiciness score. The highest score was obtained on the 5<sup>th</sup> day. General appreciation score, which

reflected the consumer pleasure, was decrease by the elongation of the storage period.

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Parameters	Groups	$0^{\mathrm{th}}$ days	5 <sup>th</sup> days	10 <sup>th</sup> days	15 <sup>th</sup> days	20 <sup>th</sup> days	25 <sup>th</sup> days	30 <sup>th</sup> days	35 <sup>th</sup> days	40 <sup>th</sup> days	45 <sup>th</sup> days
	VFF	7.30±0.37 <sup>b</sup>	$8.10{\pm}0.48$ <sup>a</sup>	2.00±0.15 °							
Color	RUG	$6.30{\pm}0.54$ <sup>ab</sup>	7.40±0.45 <sup>a</sup>	$6.90{\pm}0.38^{a}$	7.75±0.63 °	7.20±0.47 <sup>a</sup>	7.00±0.56 <sup>a</sup>	$6.40{\pm}0.31~^{\rm ab}$	$6.30{\pm}0.52~^{\rm ab}$	$4.90{\pm}0.28^{\rm b}$	
	RAG	$7.70{\pm}0.40^{\text{ ab}}$	8.30±0.45 <sup>a</sup>	7.60±0.43 <sup>ab</sup>	$8.00{\pm}0.00$ <sup>ab</sup>	6.90±0.41 abc	$6.00{\pm}0.52{ m cd}$	$6.80{\pm}0.49~{}^{ m bc}$	6.60±0.37 bcd	5.20±0.57 <sup>d</sup>	5.20±0.29 <sup>d</sup>
	VFF	7.10±0.59 <sup>b</sup>	8.50±0.22 <sup>a</sup>	$1.70{\pm}0.34^{\circ}$							
Smell	RUG	$6.40{\pm}0.40$ bc	8.00±0.26 <sup>a</sup>	$5.90{\pm}0.43$ bcd	7.25±0.48 <sup>ab</sup>	$6.50{\pm}0.52$ bc	$6.10{\pm}0.67{}^{bcd}$	4.80±0.65 <sup>d</sup>	5.70±0.63 cd	1.90±0.23 <sup>e</sup>	
	RAG	$8.00{\pm}0.37$ <sup>ab</sup>	8.80±0.29 ª	7.70±0.40 <sup>abc</sup>	7.25±0.25 bc	7.00±0.47 <sup>bcd</sup>	$6.70{\pm}0.54$ bcd	$6.60{\pm}0.50\mathrm{cd}$	$6.50{\pm}0.60$ cd	5.70±0.42 <sup>d</sup>	1.90±0.18 °
	VFF	•									
Taste	RUG	$6.00\pm0.47$ bc	8.00±0.39 <sup>a</sup>	$6.10\pm0.46$ bc	$7.75\pm0.48^{a}$	$6.90{\pm}0.50^{ m ab}$	5.80±0.59 <sup>bc</sup>	$5.10\pm0.60^{\circ}$	5.30±0.37°	2.30±0.21 <sup>d</sup>	
	RAG	$8.20{\pm}0.29$ <sup>ab</sup>	8.80±0.39 ª	7.90±0.53 <sup>abc</sup>	7.75±0.25 abcd	$6.50{\pm}0.64~^{\mathrm{cde}}$	6.50±0.43 <sup>cde</sup>	7.10±0.55 bcde	6.30±0.73 de	6.00±0.26 °	$2.20{\pm}0.20$ <sup>f</sup>
	VFF	7.50±0.37 <sup>a</sup>	8.40±0.56 <sup>a</sup>	2.90±0.53 <sup>b</sup>							
Texture	RUG	$6.90{\pm}0.64$ <sup>a</sup>	8.00±0.45 <sup>a</sup>	6.90±0.59 ª	$8.00{\pm}0.41$ <sup>a</sup>	7.10±0.50 <sup>a</sup>	6.80±0.55 <sup>a</sup>	$7.30{\pm}0.30^{\rm a}$	$6.30{\pm}0.37~^{\rm ab}$	$5.00{\pm}0.49^{\rm b}$	
	RAG	7.80±0.29 <sup>ab</sup>	$8.50{\pm}0.48^{a}$	7.90±0.50 <sup>ab</sup>	7.75±0.48 <sup>ab</sup>	6.80±0.49 <sup>bc</sup>	6.00±0.33 cd	$6.70{\pm}0.40^{\rm  bc}$	$6.50{\pm}0.48$ bc	5.70±0.40 <sup>cd</sup>	4.90±0.35 <sup>d</sup>
	VFF	7.40±0.54 <sup>a</sup>	8.30±0.47 <sup>a</sup>	3.20±0.61 <sup>b</sup>							
Appearance	RUG	6.80±0.59 <sup>ab</sup>	7.70±0.40 <sup>ab</sup>	$6.20{\pm}0.61^{ m b}$	8.25±0.75 <sup>a</sup>	7.40±0.40 <sup>ab</sup>	7.30±0.50 <sup>ab</sup>	7.30±0.26 <sup>ab</sup>	$6.40{\pm}0.78$ <sup>b</sup>	3.00±0.26 °	
	RAG	7.90±0.31 <sup>abc</sup>	$8.10{\pm}0.48$ <sup>abc</sup>	7.60±0.43 <sup>abc</sup>	8.50±0.50 <sup>a</sup>	7.30±0.40 <sup>abcd</sup>	6.40±0.43 <sup>cde</sup>	6.50±0.50 <sup>cd</sup>	6.80±0.39 bcd	5.00±0.68 °	5.90±0.38 <sup>ed</sup>
	VFF	7.50±0.45 ª	8.70±0.37 <sup>a</sup>	$3.33\pm0.80^{\rm b}$							
Juiciness	RUG	7.20±0.61 abc	8.20±0.42 <sup>a</sup>	6.30±0.67 <sup>bc</sup>	7.50±0.96 <sup>ab</sup>	7.30±0.54 <sup>abc</sup>	$5.50\pm0.48$ °	6.90±0.38 abc	6.70±0.65 <sup>abc</sup>	$3.70{\pm}0.40^{d}$	
	RAG	$8.30{\pm}0.21$ ba	8.50±0.45 <sup>a</sup>	7.20±0.47 <sup>abc</sup>	7.75±0.63 <sup>ab</sup>	$6.78{\pm}0.36\mathrm{bcd}$	$6.30{\pm}0.47$ bcd	$6.40{\pm}0.60$ bcd	7.20±0.55 <sup>abc</sup>	5.70±0.45 <sup>cd</sup>	5.60±0.31 <sup>d</sup>
	VFF	7.00±0.58 <sup>a</sup>	8.50±0.40 <sup>b</sup>	1.90±0.23 °							
General	RUG	$6.60{\pm}0.54$ <sup>bcd</sup>	8.30±0.45 <sup>a</sup>	$6.10{\pm}0.50{ m cd}$	7.75±0.63 <sup>ab</sup>	7.40±0.45 <sup>abc</sup>	$6.00{\pm}0.45$ <sup>cd</sup>	$6.50{\pm}0.34$ bcd	$5.60{\pm}0.37^{\rm d}$	2.70±0.50 °	
mannardan	RAG	$8.20{\pm}0.20$ <sup>abc</sup>	8.80±0.39ª	7.90±0.48 abcd	8.25±0.48 <sup>ab</sup>	$6.90{\pm}0.46$ bod	$6.40{\pm}0.37~^{\rm d}$	6.80±0.57 bcd	6.70±0.70 <sup>cd</sup>	4.80±0.49 <sup>e</sup>	$2.40{\pm}0.16^{\rm f}$

Values in the same line the with same lowercase are statistically insignificant (p > 0.05)

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