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Influence of Grape Seed Extract on Quality Characteristics of Vacuum Packaged Restructured Mutton Slices during Refrigerated Temperature (4±1°C)

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Abstract

The present investigation was carried out to evaluate the efficacy of grape seed extract (GSE) on quality (oxidative, microbial, textural and sensory) characteristics of vacuum packaged restructured mutton slices (RMS) during refrigerated storage (4±1°C). GSE found significantly ($P<0.05$) lower 2-TBARS values compared to control and butylated hydroxyl anisole (BHA). Free fatty acid (FFA) per cent was significantly ($P<0.05$) influenced by both antioxidants and storage period. GSE was significantly ($P<0.05$) lower FFA (0.163) compared to control (0.180) during 28 days of refrigerated storage. RMS added with GSE had significantly ($P<0.05$) reduced mean total plate counts from 3.95 to 2.70 ($\log_{10}$ cfu/g) compared to control sample. The mean *coliform* counts were 1.08, 0.84 and 0.77 ($\log_{10}$ cfu/g) in control, BHA and GSE added samples respectively. RMS added with GSE had significantly reduced mean anaerobic counts from 2.91 to 2.68 ($\log_{10}$ cfu/g). Addition of GSE had significantly ($P<0.05$) influenced various textural characteristics. As the progressing of storage period, the hardness values increased and remaining all textural characteristics are reduced gradually. RMS added with GSE had significantly higher chewiness, cohesiveness and gumminess values and lower hardness and springiness values than control and RMS added with BHA. GSE significantly ($P<0.05$) improved the color, flavor, juiciness and overall acceptability scores than control and RMS added with BHA. Based on the results, GSE is acting as better antioxidant comparing to BHA and by utilizing vacuum environment and addition of GSE at 0.1% may enhance the shelf-life of RMS up to 28 days under refrigerated storage without any significant quality deterioration.

Keywords: Restructured Mutton Slices; Grape Seed Extract; Antioxidant; Vacuum Packaging; Storage Stability.

Introduction

During restructuring, any disruption of the integrity of muscle membranes by mechanical deboning, grinding, tumbling, massaging or cooking alters the cellular compartmentalization. This facilitates the interactions of pro-oxidants with unsaturated fatty acids resulting in the generation of free radicals and the propagation of oxidative reactions. Lipid oxidation is one of the primary mechanisms of quality deterioration in restructured meat products. The changes in quality are manifested by adverse changes in flavor, color, texture and nutritive value, and in the possible production of toxic compounds (Gray *et al.*, 1996). The most common strategies for preventing lipid oxidation are the use of antioxidants and restriction of access to oxygen during storage by vacuum packaging (Tang *et al.*, 2001). Synthetic antioxidants such as butylated hydroxytoluene (BHT) and butylated hydroxyanisole (BHA) have been used to control lipid oxidation in meat products. However, the use of these synthetic antioxidants is restricted because of their toxic or carcinogenic effects. Consumer preferences for ''natural'' products have resulted an increased interest in the use of natural antioxidants from rosemary, sage, aloe vera, mustard, tea catechins, whey protein concentrate, and cottonseed meals (Rhee *et al.,* 2001; Rababah *et al.,* 2004). Several types of natural plant derived antioxidants have been studied in various raw and cooked meat systems, including extracts of grape seed, rosemary extract, sage, thyme, rice bran, white peony, red peony, sappan wood, moutan peony, rehmannia or angelica, sedge, marjoram, wild marjoram, caraway, basil extract, ginger, plum concentrates, aloe vera, mustard, tea catechins, and rosemary extract (Namaki, 1990; El Alim *et al.,* 1999; Armitage *et al,.* 2002; Tsen *et al.,* 2006; Fiorentino *et al.,* 2008).

Grape seed extract (GSE) is a byproduct derived from the grape seeds (*Vitis vinifera*) (from grape juice and wine processing) that is extracted, dried and purified to produce a polyphenolic compound rich extract (Lau and King, 2003). Standardized grape seed extracts contain 74 to 78% oligomeric proanthocyanidins and less than approximately 6% of free flavanol monomers on a dry weight basis. Proanthocyanidins in the form of monomeric phenolic compounds, such as catechin, epicatechin and epicatechin-3-O-gallate, and in dimeric, trimeric and tetrameric procyanidin forms are rich in GSE. The multiple mechanisms of their antioxidative activity are expressed in its ability of radical scavenging, metal chelation, and synergism with other antioxidants and confirmed by *b*-carotene linoleate and linoleic acid peroxidation methods (Jayaprakasha *et al.,* 2001) as well as by DPPH and phosphomolybdenum complex methods (Jayaprakasha *et al.,* 2003). The antimicrobial properties of grape seed extract against gram positive and gram negative bacteria have been reported (Jayaprakasha *et al.,* 2003).

This work was aimed at comparative evaluation of the antioxidative and antimicrobial efficacy of grape seed extract with synthetic antioxidant i.e., butylated hydroxyanisole in vacuum packaged restructured mutton slices during refrigerated storage (4±1°C).

Material and Methods

Preparation of restructured mutton slices (RMS)

The processing and preparation of RMS (C) were as follows: Thawing of frozen mutton (85 % of formulation) at 4±1°C for 16 hours then mincing the mutton portions by using 25 mm blade, add curing ingredients (salt @1.5 %, sodium tripolyphosphates @ 0.4 %, sodium nitrite @ 150 ppm, sodium ascorbate @ 500 ppm, sugar @ 1%) then cure the formulation about 12 hours at 4±1°C. Continuous vacuum tumbling of sample for about 1 hour with ice flakes (6 %) then added maida (2.5 %), spicemix (0.5 %) and onion garlic paste (2 %). Then massage the meat till tacky exudates forms in a meat massager. Then the meat batter was divided into three portions and one was control, one portion was added with butylated hydroxyl anisole (BHA) @ 0.01 % and remaining portion was added with grape seed extract (GSE) @0.1%. Fill the meat batter in round stainless steel moulds then steam cooking the samples for 45 min (core temperature of the product reached to 80±1°C). Chill the meat samples at 4±1°C about 12 hours. Cut the product into 5 mm round slices with meat slicer and vacuum packaged then stored in refrigerated temperature (4±1°C).

Analysis

The RMS samples added with antioxidants were analyzed at seven days interval for following quality characteristics namely: pH (Trout *et al.,* 1992), 2-thiobarbituric acid reactive substance (TBARS) value (Witte *et al.,* 1970), free fatty acids per cent (Koniecko, 1979), textural attributes (Bourne, 1978) and microbiological profiles (APHA, 2005). For sensory characteristics, the RMS were evaluated for appearance, flavor, chewiness, cohesiveness, juiciness, mouth coating and overall palatability using an 8-point descriptive scale (where, 8=extremely desirable, 1= extremely undesirable) as described by Keeton (1983) with slight modifications. The sensory attributes and their descriptions were adopted from Berry (1987) with some modifications. Sensory evaluation conducted between 3.30 – 4.0 pm and filtered tap water was provided to the panelists for rinsing their mouth in between evaluation of different samples.

Statistical analysis

The experiment was replicated four times and the data generated for different quality characteristics were compiled and analyzed. The data were subjected to analysis of variance (one way analysis of variance), paired t-test (Snedecor and Cochran 1995) for comparing the means to find the difference between groups, storage periods and their interaction for various parameters. The smallest difference ($D_{5\%}$) for two means to be significantly different (P<0.05) was reported.

Results and Discussion

Physico-chemical characteristics

Neither GSE nor BHA had influenced the pH values of vacuum packaged RMS during refrigerated storage (Fig.1). As progressing of refrigerated storage period, the pH values are significantly (P<0.05) increased with irrespective of treatment. This increase in pH during storage could be due to denaturation and liberation of protein metabolites, mainly amines due to bacterial activity. These results are in congruent with Brannan (2009) in ground chicken breast samples, Sasse *et al.* (2009) in cooked frozen pork patties.

There was a significant (P<0.05) difference in 2-TBARS values of vacuum packaged RMS due to antioxidants and storage period (Fig. 2). GSE found significantly (P<0.05) lower 2-TBARS values compared to control and BHA which might be due to potential antioxidative property of GSE. Initially the 2-TBARS values are in the range of 0.192-0.193 mg malonaldehyde/kg meat, but as the progressing of refrigerated storage period 2-TBARS values of control RMS had progressively increased to 1.210 mg malonaldehyde/kg meat. GSE recorded significantly lower 2-TBARS values i.e.,) 0.499 mg (28 days) malonaldehyde/kg meat which indicates that antioxidative activity of GSE started between 0-7 days of storage onwards and continues till the 28 days of refrigerated storage. The antioxidative property is mainly contributed by phenolic compounds, especially polyphenols, such as proanthocyanidins. These proanthocyanidins are dimers, trimers and oligomers of the monomeric flavan-3-ols (+)-catechin, (-)-epicatechin and (-)-epicatechin-3-O-gallate. These results are in accordance with Brannan (2009) in ground chicken breast samples.

Free fatty acid (FFA) per cent was significantly (P<0.05) influenced by both antioxidants and storage period (Fig.3). GSE was significantly (P<0.05) lower FFA (0.163) compared to control (0.180) during 28 days of refrigerated storage. The lower FFA is mainly due to delaying of oxidation of oleate fatty esters by GSE polyphenols and antimicrobial activity of GSE which causes reduction in microbial growth and subsequent microbial lipolytic activity. Storage period also significantly (P<0.05) affected the FFA per cent and the overall mean days of FFA per cent values were increased from 0.103 (0 day) to 0.264 on 28 days of refrigerated storage. The increased FFA during storage could be due to micribial lipolytic activities. This is in accordance with Sahoo and Anjaneyulu (1997) and Abdullah (2004) who reported increasing trend of free fatty acid during storage of meat products and suggested that the use of FFA percentage as an analytical indicator to judge the fat quality.

Fig. 1: Influence of GSE on pH values of vacuum packaged RMS during refrigerated storage (4±1°C)

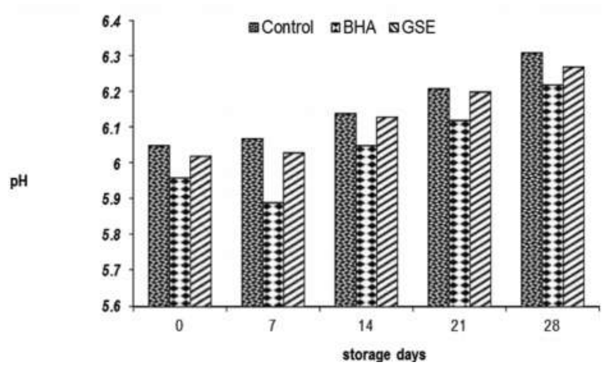


Fig. 2: Influence of GSE on TBARS values of vacuum packaged RMS during refrigerated storage (4±1°C)

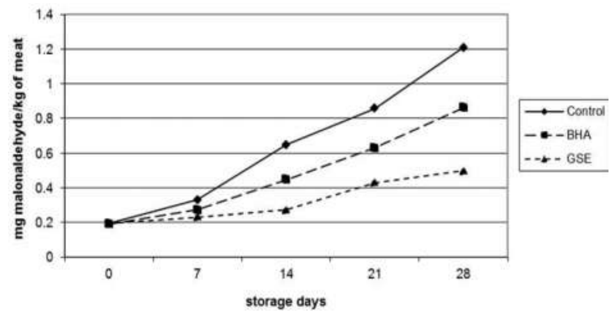
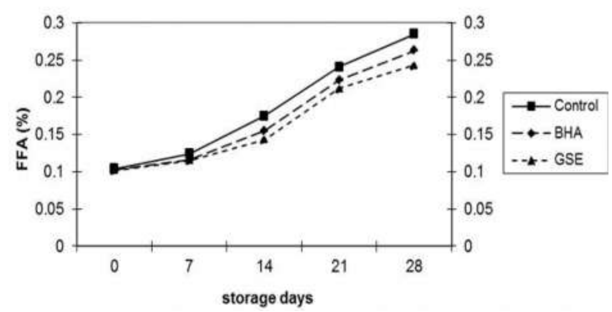


Fig. 3: Influence of GSE on free fatty acids (FFA) of vacuum packaged RMS during refrigerated storage (4±1°C)



Microbial characteristics

The antimicrobial activity of grape seed extract on various bacterial growth of vacuum packaged RMS during refrigerated storage was presented in Table 1. GSE had significantly (P<0.05) reduced all microbial population at concentration of 0.1% compared to control and BHA samples. Initial microbial load of all samples were almost same and total psychrophilic and *coliform* counts were not noted during 0 and 7 days of refrigerated storage. Total plate, *lactobacillus* and anaerobic counts were noted 0 day onwards and as the progressing of storage period their counts were also increased. In control sample total plate counts were increased from 2.14 (0 day) to 5.67 (28 day) (log_{10} cfu/g) and in BHA and GSE these counts were reached to 5.19 and 3.29 (log_{10} cfu/g) respectively at 28th day of refrigerated storage. RMS added with GSE had significantly (P<0.05) reduced mean total plate counts from 3.95 to 2.70 (log_{10} cfu/g) compared to control sample. The mean *coliform* counts were reduced from 1.08, 0.84 and 0.77 (log_{10} cfu/g) in control, BHA and GSE added samples respectively. RMS added with GSE had significantly reduced mean anaerobic counts from 2.91 to 2.68 (log_{10} cfu/g). The reduction in microbial population during refrigerated storage is mainly due to antimicrobial activity of grape polyphenols and their active components such as epigallocatechin, epigallocatechin-3-Ogallate, castalagin and prodelphinidin. As GSE is a rich source of polymers of flavan-3-ols like (+)-catechin and (")-epicatechin), its antimicrobial properties can be attributed to the general mechanisms of phenolics. Polyphenols extracted from grape seed extract have shown inhibitory effects on Gram-positive as well as Gram-negative bacteria (Gadang et *al.,* 2008).

Table 1: Influence of GSE on microbial characteristics of vacuum packaged RMS during refrigerated storage (4±1°C)

Treatment (n=8)	Storage days 0	7	14	21	28	Treatments Mean (SE)
			Total plate counts (log_{10} cfu/g)			
Control	2.14	3.11	3.89	4.92	5.67	3.95 ^{b} (0.03)
BHA	2.31	3.21	3.77	4.31	5.19	3.76 ^{b} (0.02)
GSE	2.19	2.44	2.59	2.96	3.29	2.70 ^{a} (0.05)
Days Mean (SE)	2.21 ^{a} (0.04)	2.92 ^{b} (0.07)	3.42 ^{c} (0.04)	4.06 ^{d} (0.05)	4.72 ^{e} (0.01)	
			Total psychrophilic counts (log_{10} cfu/g)			
Control	0	0	2.24	2.77	3.51	1.71 (0.02)
BHA	0	2.32	2.41	2.75	3.07	1.80 (0.06)
GSE	0	0	2.41	2.90	3.27	1.72 (0.05)
Days Mean (SE)	0	0.71 ^{a} (0.03)	2.35 ^{b} (0.01)	2.81 ^{c} (0.08)	3.42 ^{d} (0.04)	
			Coliform counts (log_{10} cfu/g)			
Control	0	1.56	1.37	1.37	1.53	1.08 ^{c} (0.13)
BHA	0	0	0	1.23	1.49	0.53 ^{a} (0.10)
GSE	0	0	1.20	1.24	1.42	0.77 ^{b} (0.09)
Days Mean (SE)	0	0.39 ^{a} (0.24)	0.86 ^{b} (0.16)	1.28 ^{c} (0.11)	1.46 ^{d} (0.05)	
			Lactobacillus counts (log_{10} cfu/g)			
Control	1.20	1.62	2.42	3.67	3.72	2.53 (0.33)
BHA	1.17	1.58	2.50	3.04	2.94	2.44 (0.28)
GSE	1.17	1.70	2.24	3.32	3.86	2.46 (0.03)
Days Mean (SE)	1.18 ^{a} (0.08)	1.63 ^{b} (0.16)	2.38 ^{c} (0.21)	3.34 ^{d} (0.05)	3.84 ^{e} (0.04)	
			Anerobic counts (log_{10} cfu/g)			
Control	1.63	2.68	2.93	3.37	3.97	2.91 ^{b} (0.33)
BHA	1.39	2.85	2.86	3.29	3.98	2.88 ^{b} (0.27)
GSE	1.52	2.78	2.64	2.92	3.52	2.68 ^{a} (0.19)
Days Mean (SE)	1.51 ^{a} (0.18)	2.77 ^{b} (0.09)	2.81 ^{b} (0.19)	3.19 ^{c} (0.25)	3.82 ^{d} (0.43)	

Means with different superscripts differ significantly (P<0.05).

Textural characteristics

Addition of natural antioxidants may delay/ prevent storage changes for certain extent thus maintains structural integrity of the products leads to increasing the shelf life of the meat products. In present investigation, addition of GSE had significantly (P<0.05) influenced various textural characteristics (Table 2). As the progressing of storage period, the hardness values increased and remaining all textural characteristics are reduced gradually. RMS added with GSE had significantly higher chewiness, cohesiveness and gumminess values and lower hardness and springiness values than control and RMS added with BHA. Initial (day 0) values of all textural parameters in all treatments did not differ. However, all textural parameters are gradually changed during refrigerated storage. But RMS added with GSE had recorded better textural characteristics compared to control and BHA added RMS. This could be due to free radicals generated in the RMS can not only cause the development of lipid oxidation but also causes protein oxidation, resulting in denaturation of protein molecules. In addition, the lipid oxidation secondary products can also cause protein denaturation, which deteriorates texture properties of meat products (Kanner, 1994). Myofibrillar proteins play crucial roles in functional properties, such as water holding properties and textural properties in various meat products. Oxidative damage of proteins may cause loss of protein solubility, aggregation, and complex formation and consequently loss of protein network (Karel *et al.*, 1975, Estevez *et al.*, 2006). These changes can result in reduced spaces between the protein molecules in the restructured slices, leading to greater loss of moisture during storage and consequently a harder texture. In addition, broken protein networks can cause loss of textural properties such as chewiness, cohesiveness and gumminess. Therefore, inclusion of grape seed extract may prevent the development of protein oxidation and maintain the textural properties of the RMS during refrigerated storage.

Table 2: Influence of GSE on textural characteristics of vacuum packaged RMS during refrigerated storage (4±1°C)

Treatment (n=12)	Storage days					Treatments Mean (SE)
	0	7	14	21	28	
			Chewiness (N mm)			
Control	33.58	30.67	27.85	27.19	23.73	28.61^{a} (0.64)
BHA	32.97	31.59	30.64	27.49	24.67	29.47^{b} (0.29)
GSE	33.99	31.47	30.68	28.74	28.11	30.60^{c} (0.59)
Days Mean (SE)	35.51^{e} (0.82)	31.24^{d} (0.58)	29.72^{c} (0.57)	27.81^{b} (0.52)	25.50^{a} (0.49)	
			Cohesiveness			
Control	0.974	0.746	0.713	0.674	0.595	0.740^{a} (0.07)
BHA	0.914	0.907	0.846	0.713	0.674	0.811^{b} (0.12)
GSE	0.939	0.927	0.897	0.807	0.721	0.858^{c} (0.24)
Days Mean (SE)	0.942^{e} (0.27)	0.860^{d} (0.28)	0.819^{c} (0.37)	0.731^{b} (0.29)	0.663^{a} (0.29)	
			Gumminess (N)			
Control	42.10	39.47	35.67	33.78	30.67	36.34^{a} (0.45)
BHA	44.31	39.69	37.61	36.47	34.17	38.45^{b} (0.28)
GSE	43.27	40.34	38.42	38.18	37.47	39.53^{c} (0.59)
Days Mean (SE)	43.23^{d} (0.59)	39.83^{c} (0.29)	37.23^{b} (0.37)	36.13^{b} (0.52)	34.10^{a} (0.43)	
			Hardness (N)			
Control	59.84	73.05	79.48	84.67	86.63	89.47^{c} (0.42)
BHA	59.61	71.18	78.14	79.87	81.44	81.10^{b} (0.49)
GSE	57.36	68.14	75.26	76.62	77.14	79.34^{a} (0.57)
Days Mean (SE)	58.94^{a} (0.55)	70.79^{b} (0.57)	77.63^{c} (0.28)	80.39^{d} (0.46)	81.74^{e} (0.29)	
			Springiness (mm)			
Control	0.854	0.799	0.746	0.642	0.634	0.735^{b} (0.13)
BHA	0.772	0.710	0.699	0.673	0.641	0.699^{a} (0.24)
GSE	0.749	0.771	0.722	0.655	0.613	0.702^{a} (0.27)
Days Mean (SE)	0.792^{d} (0.16)	0.760^{c} (0.24)	0.722^{b} (0.56)	0.657^{a} (0.34)	0.629^{a} (0.19)	

Means with different superscripts differ significantly at ($P<0.05$)

Sensory evaluation

Sensory characteristics of vacuum packaged RMS were significantly ($P<0.05$) affected by both antioxidants and storage period (Table 3). GSE significantly ($P<0.05$) improved the color scores than control and BHA during refrigerated storage of vacuum packaged RMS. This might be due to partly preventive nature of the myoglobin oxidation by GSE, which in turns delays the surface color deterioration and vacuum environment. With the progress of storage period, the mean color scores are reduced from 7.44 (0 day) to 6.12 (28 days) in vacuum packaged RMS during refrigerated storage (4±1°C). This decline in color scores encountered during storage could be due to non enzymatic browning resulted from reaction between lipid oxidation products and amino acids (Che Man *et al.*, 1995). These results are in agreed with Das *et al.* (2006) in vacuum packaged goat meat patties.

Table 3: Influence of GSE on sensory characteristics of vacuum packaged RMS during refrigerated storage (4±1°C)

Treatment (n=12)	Storage days 0	7	14	21	28	Treatments Mean (SE)
			Appearance/Colour			
Control	7.58	6.98	6.68	6.14	5.83	6.64 a (0.63)
BHA	7.33	7.02	6.79	6.39	6.27	6.76 b (0.29)
GSE	7.42	7.32	6.97	6.74	6.40	6.95 c (0.54)
Days Mean (SE)	7.44 e (0.81)	7.12 d (0.28)	6.81 c (0.19)	6.42 b (0.29)	6.12 a (0.43)	
			Flavour			
Control	7.25	6.89	6.38	6.07	5.87	6.49 a (0.28)
BHA	7.42	7.08	6.84	6.24	6.03	6.72 b (0.43)
GSE	7.46	7.03	6.91	6.33	6.12	6.77 b (0.56)
Days Mean (SE)	7.38 e (0.29)	7.00 d (0.43)	6.71 c (0.11)	6.22 b (0.53)	6.01 a (0.21)	
			Chewiness			
Control	7.00	6.91	6.93	6.42	6.37	6.73 (0.27)
BHA	6.98	6.93	6.89	6.53	6.51	6.77 (0.22)
GSE	7.03	6.97	6.99	6.51	6.43	6.79 (0.41)
Days Mean (SE)	7.00 b (0.20)	6.94 b (0.13)	6.93b (0.27)	6.49 a (0.09)	6.44 a (0.19)	
			Cohesiveness			
Control	6.98	6.74	6.61	6.54	6.51	6.68 (0.20)
BHA	6.87	6.83	6.59	6.51	6.49	6.66 (0.17)
GSE	7.01	6.94	6.72	6.57	6.55	6.76 (0.09)
Days Mean (SE)	6.92 (0.15)	6.83 (0.29)	6.64 (0.18)	6.54 (0.11)	6.52 (0.41)	
			Juiciness			
Control	7.00	6.59	6.17	6.03	5.74	6.31 a (0.76)
BHA	7.17	6.74	6.23	6.11	6.03	6.46 b (0.28)
GSE	7.19	6.87	6.31	6.09	6.05	6.51 b (0.51
Days Mean (SE)	7.12e (0.28)	6.73d (0.19)	6.24c (0.17)	6.08b (0.22)	5.94a (0.11)	
			Mouth Coating			
Control	6.25	6.27	6.13	6.19	6.03	6.19 (0.67)
BHA	6.42	6.41	6.37	6.23	6.10	6.31 (0.17)
GSE	6.45	6.39	6.34	6.27	6.14	6.32 (0.22)
Days Mean (SE)	6.37 (0.87)	6.36 (0.34)	6.28 (0.11)	6.20 (0.29)	6.09 (0.07)	
			Overall Palatability			
Control	7.13	6.74	6.43	6.18	5.92	6.48 a (0.68)
BHA	7.27	7.01	6.59	6.33	6.07	6.65 b (0.27)
GSE	7.32	7.09	6.83	6.67	6.23	6.83 c (0.19)
Days Mean (SE)	7.24 e (0.16)	6.95 d (0.28)	6.62 c (0.47)	6.39 b (0.11)	5.96 a (0.45)	

GSE showed significantly (P<0.05) higher flavor scores than control and BHA whereas, BHA was better flavor scores than control. This might be due to antioxidative effect of GSE and BHA which decreases the intensity of off-flavor generation by unknown metal chelator (Brannan and Mah, 2007). Similar to these findings Brannan (2009) reported that GSE was effective in limiting the intensity of rancid or warmed over flavor in pre-cooked meat. A significant (P<0.05) reduction was found in overall flavor scores of RMS during storage which might be due to reduction of efficacy of antioxidants and oxidation of fat as evident from TBARS values and liberation of free fatty acids (Branen, 1979) as well as increased microbial load (Sahoo and Anjaneyulu, 1997).

Antioxidants did not significantly (P>0.05) influence the chewiness scores of vacuum packaged RMS during refrigerated storage. The chewiness values of vacuum packaged RMS were significantly (P<0.05) affected by storage periods and these values were reduced from 7.00 (0 day) to 6.44 (28 days). This could attribute to more denaturation of the protein matrix as the progressing of storage, results in soft texture of the product, thus less mastication for swallowing. These results are in agreement with Reverte *et al.* (2003) in restructured beef steaks. Both antioxidants and storage period did not significantly (P>0.05) influence the cohesiveness scores of vacuum packaged RMS during 28 days of refrigerated storage. Ockerman and Organisciak (1979) reported that cohesiveness scores of restructured steak was not influenced by storage period. A significant (P<0.05) influence of antioxidants and storage period on juiciness scores of vacuum packaged RMS during refrigerated storage were noticed. The GSE showed highest juiciness scores than control and BHA. Similarly, juiciness scores of RMS added with BHA were superior over control. This could be due to antioxidant powdery substances added to product might have retained more water molecules. As the progress of storage period, a significant (P<0.05) reduction in vacuum packaged RMS was noticed. This could be due to dehydration of the product during storage period. These results are in accordance with Sahoo and Anjaneyulu (1997) in restructured buffalo meat nuggets. Neither antioxidants nor storage period significantly (P>0.05) influenced the mouth coating scores of packaged RMS during entire refrigerated storage period. Although sensory panelists recognized little differences due to both antioxidants and storage periods in mouth coating scores, they were not statistically significant. Similar findings were noted in restructured beef steaks (Schwartz and Mandigo, 1976; Coon *et al.*, 1983; Reverte *et al.*, 2003).

Overall acceptability is the reflection sensory attributes of color, flavor and juiciness. Among all treatments, GSE rated superior overall acceptability scores which might be due to favorable color, flavor and juiciness scores compared to control and BHA. Whereas, BHA scored significantly (P<0.05) higher overall acceptability than control which might be due to better scores of other related sensory attributes. With the advancement in storage period, the overall acceptability scores were significantly (P<0.05) reduced from 7.24 (0 day) to 5.96 (28 days) in vacuum packaged RMS. The possible reason for decrease in overall acceptability during storage could be due to decline in flavor, color and juiciness as a result of protein denaturation, lipid oxidation and dehydration of the meat products. These results are in accordance with Carpenter *et al.* (2007) who reported that addition of GSE significantly improve the overall acceptability of pork patties.

Based on the results, GSE is acting as excellent antioxidant comparing to BHA and concluding that by utilizing vacuum environment and addition of GSE at 0.1% enhanced the shelf-life of RMS up to 28 days under refrigerated storage (4±1ºC) without any significant quality deterioration.

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