

**IMPROVING PROPERTIES OF BEEF SAUSAGE BY USING
POLYSACCHARIDES
BY**

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ABSTRACT

Substitution of 3.0% starch in the control beef sausage batter with 0.1% xanthan, *Rhizobium* polysaccharide (RPS), gellan and carrageenan or a mixture of one of them with 0.5% starch resulted in higher sensory attributes. Beef sausage samples contained different tested polysaccharides recorded higher water holding capacity and plasticity than control sample at zero time and during frozen storage for 90 days. Although control sample showed higher initial cooking yield (78.11%) compared to those contained different polysaccharides (64.24 – 56.90%), a drastic loss in cooking yield was observed for control sample (60.24%) after frozen storage. Addition of the tested polysaccharides has enhanced the hardness values of the raw sausage than control sample. The maximum enhancement was achieved by addition of either 0.1% gellan or carrageenan with or without 0.5% starch. On the other hand, samples containing 0.1% of either RPS or carrageenan and a mixture of 0.1% RPS + 0.5% starch showed the lowest drip losses being 8.12%, 9.85% and 8.79%, respectively compared to 17.74% of control after 90 days of storage at -18°C. Therefore, it is recommended to use 0.1% of RPS, xanthan, carrageenan and gellan for improving properties of beef sausage.

INTRODUCTION

Polysaccharides derived from seaweed, plants and bacteria (gums) represent a group of hydrocolloids, which are effective in binding water in food (Xiong *et al.*, 1999).

Hydrocolloids especially carrageenan were often utilized in modifying both texture and sensory attributes of meat products, (Pietrasik and Duda, 2000).

In cooked meat products, several ingredients and number of proteins (soy, maize, whey protein, egg white, wheat, cotton seed) and carbohydrates (starch, pectin, cellulose gum, maltodextrins) have been studied. The results obtained were satisfactory, mainly with carbohydrates, which improve cooking yield, enhance water holding ability, reduce formulation cost and modify texture (Giese, 1996 and Akoh, 1998).

Hydrocolloids (carrageenan, starch, maltodextrins, etc.) help to improve rheological properties and stability of meat products. Incorporation of non-meat ingredients, such as gums or soy proteins, into processed meats has been shown to stabilize emulsions and increase water and fat retention (Lecomte *et al.*, 1993; Defreitas *et al.*, 1997a; Chin *et al.*, 1998 and Pietrasik and Duda, 2000).

Defreitas, *et al.* (1997c) stated that K- and L-carrageenan may be useful for increasing freeze / thaw stability of cooked meat products, particularly those made with low- pH meat . Thus carrageenan, improved moisture retention of cooked pork sausage both with and without addition of sodium tripolyphosphate. Carrageenans and the phosphate salt with NaCl provided the most effective combination for moisture retention in cooked pork sausage.

Sausage quality was determined by measuring textural, hydration and color characteristics. Addition of soy protein concentrate and K- carrageenan mix preparation favorably affected WHC and thermal stability of sausages processed regardless of the fat content. Use of an additive affects the sausage texture, but the effect depends on the amount of the additive used (Pietrasik and Duda, 2000).

Addition of gums and soy protein isolate could improve emulsion stability and water holding capacity of meat emulsions (Lin and Mei, 2000).

The I- and K- carrageenan increased ($p < 0.05$) cooking yield, hardness, and bind strength for 1% salt sausage. Sausage containing alginate, locust bean gum, and xanthan gum were softer, more deformable, crumbly and slippery ($p < 0.05$), when compared to non gum controls . An increase in pH sharply enhanced the binding strength in all gum treatments. Both the instrumental and sensory panel results suggested that I- and k-carrageenan were the only acceptable gums for use in low- or high-salt beef sausage products (Xiong *et al.*, 1999).

The aim of this work was to select and utilize some different sources of polysaccharides (*Rhizobium* polysaccharide, xanthan, carrageenan, gellan gum and guar gum) for improving the physicochemical and functional properties as well as acceptability of beef sausage.

MATERIALS AND METHODS

Bacterial strain:

Rhizobium meliloti (EMCC-10011), which previously selected by Madkour *et al.* (1997) to produce exopolysaccharides, was obtained from the Egyptian Microbial Culture Collection (EMCC), Faculty of Agriculture, Ain Shams University, Cairo, Egypt.

Media:

1. Standard Medium Broth (SMB):

Standard medium broth used by Footrakul *et al.* (1981) and modified by Madkour *et al.* (1997) was applied for cultivation and production of exopolysaccharide *R. meliloti*.

2. Yeast Mannitol Agar (YMA):

Stock culture of *R. meliloti* was grown and maintained on slopes of YMA medium (Oxoid Manual, 1979). Yeast mannitol broth was also used to activate and prepare heavy suspension of *R. meliloti*.

3. Nutrient agar:

Nutrient agar was used to enumerate total viable bacterial count of the different examined samples. The plates were incubated at 32°C for 24 hours (Oxoid Manual, 1979).

4. Bismuth sulphite agar:

Bismuth sulphite agar (Oxoid Manual, 1979) was used to enumerate *Salmonella sp.* The plates were incubated at 37°C for 24 hours.

5. Baird Parker medium:

Staphylococcus sp count was enumerated on Baird Parker agar (Oxoid Manual, 1979). 5ml of egg yolk tellurite was added to 100 ml of the base medium and mixed well. Incubation was carried out at 37°C for 24-48 hours.

6. Macconkey agar:

Enumeration of *coliform* bacteria was achieved using Macconkey agar (Oxoid Manual, 1979). The plates were incubated at 37°C for 24 hours.

Polysaccharides:

Polysaccharides from different sources including xanthan (bacterial source), carrageenan (seaweed source), guar gum (plant source) and gellan gum (bacterial source) as well as of those produced from *R. meliloti* are used in this study. All polysaccharides, other than RPS, were obtained from Sigma Chemical. Co. (st. louis, Mo), USA.

Production and recovery of *Rhizobium* polysaccharide:

Bacterial strain of *Rhizobium meliloti* (EMCC-10011) was used to produce exopolysaccharides as applied by Madkour *et al.* (1997).

Beef sausage ingredients:

Meat used in sausage manufacture was purchased immediately after slaughtering from the round cuts of hindquarter of cow carcass. Fat from the same carcass was also obtained. Commercially dried and ground spices were purchased from local market, Cairo, Egypt and starch was obtained from Egyptian, Company of Starch and Glucose, Mustorod, Cairo, Egypt. Dry spices mixture (Zaika *et al.*, 1978) of 1.2% was mixed with sausage ingredient (lean meat 70.0%, fat tissues 12.0%, sodium chloride 2.3%, water (as ice) 9.295%, starch 3.0%, sodium nitrite 0.005%, garlic 1.0%, onion 1.2%) for the manufacture of sausage.

Beef sausage manufacture:

Meat and fat tissues were cut into pieces of about egg size and frozen at -18°C for 24h. The frozen meat and fat were ground to particles of a rice size. Sausage was prepared by blending spices with sausage formula.

mixtures were stuffed by hand into mutton casings, the casings were then closed and chipped (Shehata, 1989). Natural mutton casings were obtained from the slaughtered animal and prepared according to El-Deep (1987). To evaluate the effects of the investigated polysaccharides on sausage quality, substitution of starch in the basic formula of sausage with a suitable level of the selected polysaccharides was investigated. Different sausage samples were packaged in polyethylene pages and stored under freezing at -18°C for 180 days until analysis.

Microbial analysis:

For microbiological analysis of sausage samples, eleven gram of each sample was weighed and added to 89 ml of sterilized physiological solution and the mixture was blended for 30 sec. Samples were examined for total viable bacterial count as well as *Salmonella sp.*, *Staphylococcus sp* and *coliform* (Rao *et al.*, 1984).

Physicochemical analysis:

Moisture, lipid, ash, carbohydrate content and total nitrogen of each sample was determined according to the A. O. A. C. (1996). Crude protein was calculated by multiplying total nitrogen percentage by 5.87 for according to Paulo *et al.* (1994) and Ceirwyn (1995). Total volatile nitrogen value was performed according to the method of the Harold., *et al.* (1987). The results were calculated as mg TVN/ 100g. The thiobarabitic acid value (TBA) was colorimetrically determined in minced sausage samples as mentioned by Harold *et al.* (1987). The pH values were measured using a pH meter (HANNA- Instrument, USA) according to Defreitas *et al.* (1997c). Water holding capacity (WHC) and plasticity were measured according to the method described by Grau and Hamm (1957) and its modification by Voloviskaya and Kelmen (1962). Change of sausage diameter and length was determined on cooked samples as mentioned by George and Berry (2000). Cooking yield was determined on cooked sausage by calculating weight difference of sausage before and after cooking in boiling water for 10min according to George and Berry (2000). Hardness of samples was determined according to Sanderson *et al.* (1988) by measuring Tension Compression (TC^2). The weight loss after thawing of frozen sausage to 2°C for 48h was recorded as thaw drip based on initial frozen weight. The percentage of weight loss was then calculated according to Defreitas *et al.* (1997c).

Sensory analysis:

Cooked sausage samples were assessed for their quality attributes by ten panelists according to Klein and Bardy (1984).

Statistical analysis:

Analysis of variance followed by multiple comparisons (LSD) was applied on data of sensory evaluation according to user's Guide of Statistical Analysis System (SAS, 1996) at the computer center of Faculty of Agricultur, Ain Shams University.

RESULTS AND DISCUSSION

Selection of kind and concentration of polysaccharides:

The effect of substitution of 3% starch with different level of RPS, xanthan, gellan gum, carrageenan and guar gum on sensory properties of beef sausage was tested to choose the best kind and concentration of polysaccharide suitable for production of high quality beef sausage.

Analysis of variance followed by multiple comparisons of panelist scores for sensory properties of different cooked sausage samples was applied to choose the best kind of polysaccharides (data not showed). No significant differences were obtained for sensory properties of sausage samples prepared with either 3.0% starch (control) or 0.1% of different tested polysaccharides. However, sausage contained higher than 0.1% of different polysaccharides received lower scores than control sample for all sensory properties. Comparison of sensory properties of cooked beef sausage samples containing 0.1% of different polysaccharides is given in Table (1). Results show that sausage samples prepared with RPS, xanthan, gellan and carrageenan recorded higher scores compared with guar gum. Therefore, 0.1% of these polysaccharides were selected to produce high quality beef sausage in the further experiments.

Combination of hydrocolloids has not been fully examined in many meat products. Therefore, many trials were designed to study the effect of the combined use of 0.1% of different tested polysaccharides and different concentrations of starch on sensory properties of beef sausage (data not showed). It was found that 0.1% of RPS and 0.5% starch produced significant improve in appearance and color of cooked beef sausage compared with the 3% starch. However, no significant difference was observed in tenderness, juiciness, aroma and overall acceptability between sausages contained 0.1% RPS +0.5%starch and those contained 3.0%starch. Combination of 0.1%RPS and starch higher than 0.5% significantly reduced all sensory properties of the resultant beef sausage. Similar trends were obtained for the other tested polysaccharides.

Proximate chemical analysis of raw beef sausage containing different Types and concentrations of polysaccharides are listed in Table (2).

Physicochemical evaluation of beef sausage during storage:

pH:

Changes in pH value of beef sausage prepared using different polysaccharides during storage at -18°C for 90 days were followed up (data not showed). The pH-values of all samples ranged from 5.66 to 5.70 at zero time and from 5.79 to 5.82 after 90 days of storage at -18°C indicating slightly increase during storage. These means that neither level nor kind of polysaccharide affected pH value. This result is similar to that obtained by Lin and Keeton (1998) and Shand (2000).

Table (1): Sensory evaluation of cooked beef sausage containing 0.1% of different polysaccharides .

Polysaccharides	Means of scores of sensory properties					Overall acceptability
	Tenderness	Juiciness	Appearance	Aroma	Color	
RPS	8.4 ^{ab}	8.4 ^{ab}	8.4 ^a	8.6 ^a	8.4 ^b	8.7 ^{ab}
Xanthan	8.5 ^{ab}	8.4 ^{ab}	9.0 ^a	8.5 ^a	8.7 ^{ab}	8.7 ^{ab}
Carrageenan	8.3 ^{ab}	8.5 ^{ab}	8.8 ^a	8.6 ^a	8.6 ^{ab}	8.5 ^{ab}
Gellan	8.8 ^a	8.7 ^a	8.8 ^a	8.7 ^a	9.0 ^a	8.9 ^a
Guar	7.9 ^b	7.6 ^b	8.4 ^a	8.6 ^a	8.6 ^{ab}	8.2 ^b
L.S.D	0.791	0.851	0.604	0.644	0.559	0.658

a, b : Means in a columns showing the same letter are not significantly different ($p > 0.05$)
 L.S.D : Least significant difference ($\alpha = 0.05$). RPS : *Rhizobium* polysaccharide

Table (2): Proximate chemical analysis of raw beef sausage containing different kind and concentrations of polysaccharides (fresh weight basis).

Constituents (%)	3% starch (control)	Beef sausage with 0.1% of				Beef sausage with 0.5% starch + 0.1% of			
		RPS ^a	Xanthan	Carrageenan	Gellan	RPS ^a	Xanthan	Carrageenan	Gellan
Moisture	62.23	65.26	65.80	65.47	64.83	64.56	64.20	64.85	64.81
Protein	15.80	15.38	14.97	15.80	15.09	15.30	15.50	15.50	15.40
Fat	14.31	13.85	13.93	14.22	14.78	14.59	14.65	14.05	14.06
Ash	2.44	2.54	2.41	2.42	2.50	2.40	2.50	2.56	2.52
* Total carbohydrates	5.22	2.97	2.89	2.09	2.80	3.15	3.15	3.04	3.21

RPS : *Rhizobium* polysaccharide.

* : Total carbohydrates calculated by difference.

Water holding capacity and plasticity:

Water holding capacity (WHC) is the ratio of moisture retained in sample to the initial moisture content, so higher percentage indicates release of less moisture (Pietrasik and Duda, 2000; Lin and Mei, 2000). Water holding capacity (WHC) and plasticity of beef sausage samples containing different polysaccharides were followed during storage at -18°C for 90 days and the results are given in Tables (3 and 4).

Raw sausage contained either 0.1% of different polysaccharides alone or in combination with 0.5%starch showed higher (WHC) (except those containing 0.1% gellan + 0.5%starch) compared with control sample containing 3.0% starch at zero time. All samples showed a decreasing trend in bound water percentage with increasing storage period at -18°C . Control beef sausage that contained 3% starch had the lowest WHC after 90 days of storage at -18°C when compared to other samples containing polysaccharides. This is because starch in raw sausage samples is uncooked, ungelatinized and in granule form. Therefore starch will not be able to hold higher amount of moisture unless it is heated to

rupture the granules causing swelling. Sausage samples containing RPS and xanthan showed the highest stability of WHC during storage, that the breakdown rate values were $4.17E-2$ and $3.97E-2$, respectively. Using of a mixture of 0.1% polysaccharides and 0.5% starch did not enhance the WHC or stability of water holding capacity of sausage samples during storage except for that sample containing a mixture of 0.1% gellan and 0.5% starch. These results agree with those reported by Defreitas *et al.* (1997a). They found that carrageenan improved moisture retention of low fat cooked pork sausage both with and without addition of sodium salts. They attributed such behaviour to physical entrapment of protein and water by the carrageenan with no obvious molecular interaction involving meat proteins.

Plasticity values showed a pattern similar to that of WHC. Control samples containing 3% starch showed lower plasticity than those sausage samples containing 0.1% polysaccharides or a combination of 0.1% polysaccharide + 0.5% starch. The breakdown rate values of plasticity "b" during frozen storage followed the same pattern as that of the loss in WHC for most of the samples. As seen from Table (4), higher losses during frozen storage were observed for control samples as well as samples containing gellan or carrageenan, since the "b" values were $-8.39E-1$, $-6.87E-1$ and $-5.75E-1$, respectively.

The data in the above mentioned Tables obviously showed that addition of polysaccharides alone or in combination with starch enhance both WHC and plasticity of beef sausage samples, especially when sausage samples stored at -18°C for 90 days.

Thaw drip:

Results in Table (5) show that sausage containing 3% starch (control) had higher thaw drip than those with other tested polysaccharides. Also, addition of either RPS or carrageenan reduced thaw drip to 5.35% and 3.34% respectively compared to 15.28% for control sample after 30 days of storage. Drip losses were increased with increasing storage period for all sausage samples. However, the loss of drip after 90 days at -18°C in beef sausage containing 3% starch (control) was higher than those containing other polysaccharides. This behaviour agrees with the low cooking yield observed for the same sample after 90 days of frozen storage (Table 6). It could be suggested that starch granules loss large amount of bound water upon freezing and thawing causing the higher drip losses and the lower cooking yield. Similar trend was observed with sausage samples containing xanthan or mixture of xanthan and starch, which showed drip losses of 16.75 and 16.14% as well as increasing rate of thaw drip "b" values of $1.05E-1$ and $1.54E-1$, respectively after 90 days of frozen storage. On contrary, samples containing 0.1% of either RPS or carrageenan and a mixture of RPS and starch showed the lowest drip losses being 8.12, 8.79 and 9.85% as well as "b" values of $4.62E-02$, $1.55E-02$ and $1.09E-01$, respectively after 90 days of frozen storage. These results are agreement with those obtained by Defreitas *et al.* (1997c) who found that addition of Kappa and Iota -carrageenan increased moisture retention and decreased thaw drip of sausage.

Table (3): Water holding capacity (WHC) * of beef sausage containing different polysaccharides during storage at -18°C .

Storage period (days)	3% starch (control)	Beef sausage with 0.1% of				Beef sausage with 0.5% starch+ 0.1% of			
		RPS	Xanthan	Carrageenan	Gellan	RPS	Xanthan	Carrageenan	Gellan
Zero	94.46	96.84	95.20	94.79	95.02	96.45	96.51	95.16	93.61
	± 0.01	± 0.42	± 0.38	± 0.04	± 0.18	± 0.25	± 0.13	± 0.38	± 0.06
30	94.10	96.20	95.05	94.68	93.95	95.96	96.46	95.01	93.28
	± 0.08	± 0.06	± 0.05	± 0.17	± 0.15	± 0.17	± 0.11	± 0.22	± 0.06
60	88.39	93.68	93.11	91.67	89.56	93.28	93.46	93.72	93.27
	± 0.14	± 0.09	± 0.17	± 0.18	± 0.21	± 0.03	± 0.25	± 0.17	± 0.0
90	86.63	93.51	91.88	90.56	88.56	92.93	92.69	91.18	91.61
	± 0.01	± 0.24	± 0.31	± 0.12	± 0.29	± 0.01	± 0.23	± 0.33	± 0.09
(b)	-9.73E-02	-4.17E-02	-3.97E-02	-5.23E-02	-7.92E-02	-4.41E-02	-4.82E-02	-4.41E-02	-2.00E-02
(R ²)	0.899	0.892	0.921	0.898	0.926	0.895	0.877	0.860	0.739

Remark: WHC of raw meat = 80.27 ± 0.07 RPS : *Rhizobium* polysaccharide.* : Expressed as percent bound water (mean of 3 replicates \pm SD). b: is a parameter representing breakdown rate of WHC /day.R² : Coefficient of determinationTable (4): Plasticity (g/mm^3) * of beef sausage containing different polysaccharides during storage at -18°C .

Storage period (days)	3%starch (control)	Beef sausage with 0.1%of				Beef sausage with 0.5% of starch+ 0.1%of			
		RPS	Xanthan	Carrageenan	Gellan	RPS	Xanthan	Carrageenan	Gellan
Zero	484.40	506.82	542.94	508.98	563.85	525.44	524.98	502.53	522.00
	± 3.3	± 2.2	± 1.7	± 1.8	± 1.7	± 3.3	± 0.8	± 2.5	± 2.0
30	455.54	490.78	519.29	460.86	512.00	506.58	488.91	483.58	479.00
	± 0.2	± 0.85	± 1.2	± 1.8	± 1.7	± 2.5	± 1.0	± 2.0	± 1.0
60	434.94	487.67	507.35	427.77	470.43	502.75	483.68	481.21	471.99
	± 3.9	± 1.1	± 2.6	± 1.9	± 1.9	± 2.7	± 2.0	± 2.4	± 1.8
90	407.37	480.3	474.14	418.66	412.68	487.57	475.20	445.81	455.59
	± 2.5	± 1.2	± 2.3	± 1.4	± 1.6	± 2.0	± 1.9	± 1.1	± 1.8
(b)	-8.39E-01	-2.76E-01	-7.28E-01	-1.01E+00	-1.65E+00	-3.91E-01	-5.15E-01	-5.75E-01	-6.87E-01
(R ²)	0.996	0.911	0.969	0.923	0.996	0.948	0.828	0.886	0.884

Remark: Plasticity of raw meat = 167.97 ± 2.8 .RPS : *Rhizobium* polysaccharide.* : Expressed as g/mm^2 (mean of 3 replicates \pm SD).

b : is a parameter representing breakdown rate of plasticity /day.

R² : Coefficient of determination

Table (5): Percentage of thaw drip of raw beef sausage containing different polysaccharides during storage at -18°C .

Storage period (days)	3%starch (control)	Beef sausage with 0.1% of				Beef sausage with 0.5% starch + 0.1 % of			
		RPS	Xanthan	Carrageenan	Gellan	RPS	Xanthan	Carrageenan	Gellan
30	15.28	5.35	10.45	3.34	9.60	7.86	6.88	10.84	8.35
60	16.08	6.18	14.71	6.33	9.89	7.94	7.56	14.95	9.08
90	17.74	8.12	16.75	9.85	11.66	8.79	16.14	15.02	11.22
(b)	4.10E-02	4.62E-02	1.05E-01	1.09E-01	3.43E-02	1.55E-02	1.54E-01	6.97E-02	4.78E-02
(R ²)	0.961	0.949	0.960	0.998	0.853	0.814	0.805	0.763	0.926

RPS : *Rhizobium* polysaccharide.

b : is a parameter representing increasing rate of thaw drip/day.

R² : Coefficient of determinationTable (6): Percentage of cooking yield of beef sausage containing different polysaccharides during storage at -18°C .

Storage period (days)	3%starch (control)	Beef sausage with 0.1% of				Beef sausage with 0.5% starch+ 0.1% of			
		RPS	Xanthan	Carrageenan	Gellan	RPS	Xanthan	Carrageenan	Gellan
Zero	78.11	57.22	62.99	64.23	58.38	56.90	56.59	60.09	62.35
30	77.58	57.06	63.38	64.29	60.54	60.00	62.25	61.70	63.30
60	64.81	61.30	68.27	64.49	61.20	62.96	62.70	62.56	65.32
90	60.24	62.80	68.45	64.99	67.45	63.01	62.76	62.93	65.60
(b)	-2.21E-01	6.99E-02	7.09E-02	8.27E-03	9.29E-02	7.10E-02	6.32E-02	3.13E-02	3.92E-02
(R ²)	0.898	0.872	0.842	0.861	0.853	0.893	0.666	0.919	0.932

RPS : *Rhizobium* polysaccharide.

b : is a parameter representing change rate of cooking yield /day.

R² : Coefficient of determination

Cooking yield:

Cooking yield of beef sausage samples containing 3% starch and 0.1% of different polysaccharides, with or without 0.5% starch as affected by frozen storage are listed in Table (6). The cooking yield of sausage samples containing 3 % starch (control) at zero time of storage achieved a level of 78.11%, which is closed to the cooking yield value achieved by Berry (1997) for beef patties prepared by the addition of 2.7% tapioca starch. Starch granules are well known to absorb and bind large amount of water upon heating and swelling, hence the high cooking yield value of sausage containing 3% starch. Replacement of starch with 0.1% of different polysaccharides or with a mixture of 0.1% polysaccharide + 0.5% starch did not achieve the same cooking yield level obtained with starch alone as seen in Table (6). Since, 3% starch– beef sausage showed higher initial cooking yield, a drastic losses in cooking yield was observed during frozen storage. Such a behaviour could be explained by the damage in starch granules present in sausage batters upon freezing and thawing, which reduce the capability of starch to absorb high amount of water and swelling during cooking, causing higher losses of moisture to the heating media. Such hypothesis could be confirmed by the results of Berry (1997), who reported that more of the added water was lost during cooking of low fat beef patties if the ratio of the added water to the added starch was changed in the formula and agree also with the observation of Shand (2000) for low fat pork bologna with waxy starch. On the other side, beef sausage samples containing 0.1% of polysaccharides with or without 0.5% starch showed an improvement in cooking yield values after different period of frozen storage up to 90 days. The best improvement (67.45% with change rate of cooking yield “b” value of $9.29E-2$) was achieved in samples containing 0.1% gellan (Table 6). However, carrageenan-containing sausage showed a constant cooking yields during frozen storage. Similar trend was observed for samples containing a mixture of polysaccharide and starch. The enhancement of cooking yield of those samples could be referred to the batters dissolving and distribution of the polysaccharide in the sausage samples. Storage caused higher values for water binding capacity inside the cooked sausage matrix and increased cooking yield. Similar results were obtained by Bigner–George and Berry (2000). They found that incorporation of soy protein, oat bran or carrageenan in the meat batter bind more water and increased the cooking yield compared with all beef fatty patties.

Moisture content:

Results given in Table (7) show that the highest losses in moisture content were observed between the 60 and 90 days of frozen storage for cooked sausage samples containing 3% starch (control) because of the losses in salvation capacity of the starch granules upon freezing and the free water molecules build ice crystals inside the sausage matrix. Vapor pressure of ice crystals in sausage matrix is always higher than the vapor pressure in the surrounding air of the freezing cabinet; so continuous migrations of water vapor take place from the ice crystal's surface to the surrounding air. Moisture content of the raw sausage samples containing 0.1% polysaccharide with or without 0.5% starch was almost stable during frozen storage because of the capability of such polysaccharide to

build higher molecules volume entrapping large amount of water bounded to the molecule.

In conclusion, raw or cooked beef sausage prepared with 0.1% polysaccharide with or without 0.5 % starch showed higher moisture content (change rate of moisture content "b" values were $-1.71E-2$ to $3.83E-3$ and $-1.27E-2$ to $-5.23E-3$, respectively for raw samples as well as $2.39E-2$ to $4.67E-2$ and $1.14E-2$ to $4.44E-2$, respectively for cooked samples) than control samples prepared with 3.0%starch ("b" values of raw and cooked control samples were $-2.00E-2$ and $-8.18E-2$, respectively) after 90 days of frozen storage. These results agree with the results obtained for WHC.

Hardness:

Hardness is an important physical and sensorial characteristic of processed meat products such as sausage and patties. It is an indicator for the force needed by the teeth jaws to chews the meat product. Table (8) shows that addition of 0.1%polysaccharides alone or in combination with 0.5% starch has enhanced the hardness values of all the raw sausage samples except those contained RPS. The maximum enhancement was achieved by addition of gellan followed by carrageenans or xanthan with or without starch.

Cooking of raw sausage in boiling water for 10 min has radically increased the hardness of the cooked sausage. As seen from the same Table, the maximum hardness value was achieved by addition of 0.1% carrageenan followed by RPS. These results show that carrageenan is the suitable polysaccharide for the hardness of low fat sausage. The obtained results are in agreement with those achieved by Defreitas *et al.* (1997b) who found that carrageenan dissolves throughout meat during thermal processing and forms gel during cooling enhancing texture properties of the product and improving product juiciness. Xanthan and gellan did not achieve the hardness value obtained by addition of carrageenan because of the low solubility of these polysaccharides compared with that of carrageenan. Foegeding and Ramsey (1986) stated that addition of xanthan gum was shown to decrease the hardness and force-to-fraction compared with the effect achieved by addition Kappa-carrageenan to sausage sample.

Frozen storage has, generally, decreased the hardness value of both raw and cooked sausage samples. The highest decrease of hardness was found in sample containing mixture of 0.1% xanthan and 0.5% starch (change rate value of hardness was $-9.67E-1$). However, sausage sample prepared by addition of 3% starch (control) showed an increasing of hardness during frozen storage. This is may be referred to the increasing number of starch granules ruptured upon freezing and thawing, absorbing higher amount of water upon heating.

Addition of 0.5% starch to the sausage containing 0.1% of gellan or xanthan improved the hardness value of raw and cooked sausage during storage than those contained the same polysaccharide only. This is due to the fact that starch granules uptake considerable amount of moisture during swelling and cooking so that other polysaccharides could be concentrated in every decreasing spaces between starch granules (Berry, 1997).

Table (7): Change in moisture content (mean of 3 replicates \pm SD) of raw and cooked beef sausage containing different polysaccharides during storage at -18°C .

Storage period (days)	Treatment	3% starch (control)	Beef sausage with 0.1% of				Beef sausage with 0.5% starch + 0.1% of			
			RPS	Xanthan	Carrageenan	Gellan	RPS	Xanthan	Carrageenan	Gellan
Zero	Raw	62.23 \pm 0.04	65.26 \pm 0.04	65.80 \pm 0.10	65.47 \pm 0.11	64.83 \pm 0.09	64.56 \pm 0.12	64.20 \pm 0.21	64.85 \pm 0.14	64.81 \pm 0.02
	Cooked	58.22 \pm 0.16	52.22 \pm 0.24	53.42 \pm 0.20	53.78 \pm 0.05	49.50 \pm 0.08	53.17 \pm 0.07	50.23 \pm 0.14	51.68 \pm 0.19	52.31 \pm 0.14
30	Raw	61.19 \pm 0.06	65.02 \pm 0.09	65.78 \pm 0.07	65.11 \pm 0.11	64.80 \pm 0.07	64.56 \pm 0.06	63.92 \pm 0.11	64.82 \pm 0.10	64.70 \pm 0.08
	Cooked	57.22 \pm 0.16	53.09 \pm 0.12	53.33 \pm 0.10	52.29 \pm 0.14	52.93 \pm 0.09	53.87 \pm 0.09	54.43 \pm 0.10	53.96 \pm 0.11	54.26 \pm 0.11
60	Raw	60.90 \pm 0.08	64.88 \pm 0.15	65.71 \pm 0.10	64.69 \pm 0.10	64.79 \pm 0.15	64.45 \pm 0.10	63.72 \pm 0.12	64.32 \pm 0.15	64.67 \pm 0.09
	Cooked	56.35 \pm 0.15	53.20 \pm 0.15	54.17 \pm 0.09	52.39 \pm 0.10	52.94 \pm 0.17	56.49 \pm 0.10	54.62 \pm 0.09	55.23 \pm 0.10	54.42 \pm 0.09
90	Raw	60.33 \pm 0.03	64.88 \pm 0.11	65.30 \pm 0.10	64.34 \pm 0.11	64.44 \pm 0.12	64.21 \pm 0.12	64.65 \pm 0.12	63.31 \pm 0.11	63.94 \pm 0.10
	Cooked	50.33 \pm 0.10	53.32 \pm 0.12	54.71 \pm 0.12	52.45 \pm 0.16	53.94 \pm 0.11	56.65 \pm 0.12	54.84 \pm 0.10	55.45 \pm 0.11	54.65 \pm 0.16
(b)	Raw	-2.00E-02	-4.27E-03	-5.23E-03	-1.27E-02	-3.93E-03	-3.87E-03	3.83E-03	-1.71E-02	-8.80E-03
(R ²)	Raw	0.943	0.850	0.745	0.999	0.685	0.824	0.136	0.845	0.734
(b)	Cooked	-8.18E-02	1.14E-02	1.57E-02	-1.30E-02	4.44E-02	4.35E-02	4.67E-02	4.19E-02	2.39E-02
(R ²)	Cooked	0.796	0.773	0.864	0.508	0.784	0.892	0.673	0.882	0.739

RPS : *Rhizobium* polysaccharide b : is a parameter representing change rate of moisture content /day.
R² : Coefficient of determination

Table (8): Degree of hardness (g/cm²) of raw and cooked beef sausage containing different polysaccharides during storage at -18°C .

Storage period (days)	Treatments	3%starch (control)	Beef sausage with 0.1%of				Beef sausage with 0.5%starch + 0.1% of			
			RPS*	Xanthan	Carrageena n	Gellan	RPS*	Xanthan	Carrageena n	Gellan
Zero	Raw	230	220	310	310	330	210	300	320	380
	Cooked	850	1100	950	1290	980	1000	1170	1180	1120
30	Raw	230	210	300	300	330	190	270	310	360
	Cooked	860	920	870	1260	960	990	1110	1140	1110
60	Raw	240	180	280	290	320	180	250	270	350
	Cooked	890	910	850	1250	940	970	1060	1130	1060
90	Raw	250	170	270	230	310	170	210	250	320
	Cooked	960	820	850	1220	920	930	900	1010	1050
(b)	Raw	2.33E-01	-6.00E-01	-4.67E-01	-8.33E-01	-2.33E-01	-4.33E-01	-9.67E-01	-8.33E-01	-6.33E-01
(R ²)	Raw	0.891	0.953	0.980	0.806	0.891	0.966	0.984	0.954	0.963
(b)	Cooked	1.20E+00	-2.83E+00	-1.07E+00	-7.33E-01	-6.67E-01	-7.67E-01	-2.87E+00	-1.73E+00	-8.67E-01
(R ²)	Cooked	0.876	0.875	0.753	0.968	1.000	0.920	0.920	0.840	0.914

RPS : *Rhizobium* polysaccharide. b : is a parameter representing change rate of hardness /day
R² : Coefficient of determination

Diameter and length:

Reduction percentages in diameter and length of cooked beef sausage samples compared to the raw samples are given in Table (9). Reduction in both length and diameter was observed as a result of cooking of different beef sausage. Samples containing 0.1% RPS with or without 0.5% starch showed the highest reduction in length, whereas those with 0.1% carrageenan with or without 0.5% starch recorded the highest reduction in diameter at zero time. A considerable reduction in diameter was observed with increasing storage period under frozen condition for different cooked samples. The highest reduction in diameter was calculated for samples containing 3.0% starch followed by 0.1% carrageenan and a mixture of 0.1% gellan and 0.5% starch after 90 days of frozen storage that the change rate of diameter values were $1.01E-1$, $7.06E-2$ and $9.89E-2$, respectively. On the other hand, reduction in length of cooked sample containing 3.0 % starch was increased from 14.32% to 22.22% (change rate of length "b" value was $8.13E-2$) after 90 days of frozen storage. Oppositely, samples containing 0.1% polysaccharides with or without 0.5% starch showed a decreasing pattern in length reduction during storage that "b" values ranged from $-7.23E-2$ to $-1.51E-2$.

Generally, sausage containing 0.1% of polysaccharides or their mixture with 0.5% starch showed less reduction in diameter and length during storage at -18°C for 90 days. The obtained results agree with those reported by Bigner-George and Berry (2000). They reported a reduction of 14.52% to 19.50% in diameter of beef patties prepared by addition of different polysaccharides and protein concentrate.

Thiobarabitic acid (TBA):

The thiobarabitic acid (TBA) test has been widely used to estimate the extent of lipid oxidation in meat and meat product (Wu *et al.*, 2000). TBA values (OD) of beef sausage containing different polysaccharides were measured during frozen storage of different samples. Data in Table (10) show that sausage samples had similar TBA value at zero time of storage. During storage, TBA values tended to increase and control samples recorded the highest TBA values after 90 days of storage.

Total volatile nitrogen (TVN):

Total volatile nitrogen values of beef sausage samples containing different polysaccharides were measured during storage of different samples at -18°C for 90 days and are used as an indicator for protein hydrolysis (Table 10). Results showed that all sausage samples had similar TVN values at zero time of storage (12.6 mg TVN /100 g sample). A slight increase in TVN was observed during storage of different samples (15.4 to 16.53 mg TVN / 100 g sample).

Microbiological evaluation of beef sausage during storage:

Different samples of raw beef sausage were microbiologically evaluated, for total viable bacterial count, total coliform count, and viable count of *Staphylococcus sp.*, immediately after processing as well as during storage for 90 days at -18°C and the results are given in Table (11). The initial total viable bacterial count, total coliform count and *Staphylococcus sp* count were ranged

Table (9): Percentage change in diameter and length of cooked beef sausage containing different polysaccharides during storage at -18°C .

Storage period (days)	3%starch (control)	Beef sausage with 0.1% of				Beef sausage with 0.5% starch +0.1% of			
		RPS	Xanthan	Carrageenan	Gellan	RPS	Xanthan	Carrageenan	Gellan
Change in diameter (%)									
Zero	7.33	6.67	6.06	8.82	7.89	7.14	5.92	8.82	5.08
30	11.59	8.27	9.68	9.38	10.71	8.33	6.67	10.71	11.11
60	12.12	8.82	9.38	12.90	11.76	9.68	8.97	12.90	13.16
90	17.24	9.68	9.68	14.71	12.50	9.68	11.11	13.79	14.29
(b)	1.01E-01	3.19E-02	3.52E-02	7.06E-02	4.96E-02	2.99E-02	5.96E-02	5.70E-02	9.89E-02
(R ²)	0.926	0.952	0.596	0.938	0.903	0.896	0.962	0.975	0.872
Change in length (%)									
Zero	14.32	15.79	13.30	13.38	11.11	16.24	14.86	12.50	12.24
30	18.60	15.38	12.50	8.79	10.84	14.81	10.84	12.00	10.57
60	19.28	14.79	11.00	7.33	10.40	11.36	9.33	11.31	8.72
90	22.22	11.86	10.00	6.71	9.15	10.16	8.33	11.22	6.76
(b)	8.13E-02	-4.13E-02	-3.80E-02	-7.16E-02	-2.11E-02	-7.23E-02	-7.03E-02	-1.51E-02	-6.10E-02
(R ²)	0.932	0.808	0.988	0.846	0.885	0.962	0.900	0.933	0.999

RPS : *Rhizobium* polysaccharide. b : is a parameter representing change rate of diameter or length/day.

R² : correlation coefficient.

Table (10): Thiobarbituric acid (TBA) and total volatile nitrogen of raw beef sausage containing different polysaccharides during storage at -18°C .

Storage Period (days)	(3%starch) control	Beef sausage with 0.1% of				Beef sausage with 0.5% starch+0.1% of			
		RPS*	Xanthan	Carrageenan	Gellan	RPS*	Xanthan	Carrageenan	Gellan
TBA (OD)									
Zero	0.038	0.035	0.035	0.035	0.034	0.034	0.034	0.035	0.036
30	0.09	0.051	0.068	0.067	0.060	0.057	0.052	0.065	0.072
60	0.312	0.205	0.197	0.209	0.197	0.178	0.189	0.188	0.277
90	0.384	0.287	0.214	0.224	0.227	0.198	0.197	0.201	0.280
TVN mg / 100g sample of sausage									
Zero	12.60	12.6	12.60	12.60	12.60	12.60	12.60	12.60	12.04
30	13.44	13.16	13.02	13.16	13.44	12.88	13.02	13.16	13.72
60	15.96	15.40	15.40	15.68	15.12	15.40	15.68	14.84	14.56
90	16.53	15.68	15.96	15.96	16.24	15.96	15.68	15.40	15.40

RPS: *Rhizobium* polysaccharide.

Table (11): Total viable bacterial count, total *coliform* count and viable count of *Staphylococcus* sp (cfu/g) of raw beef sausage containing different polysaccharides during storage at -18°C

Storage period (days)	Control (3%starch)	Beef sausage with 0.1% of				Beef sausage with 0.5 % starch+ 0.1% of			
		RPS	Xanthan	Carrageenan	Gellan	RPS	Xanthan	Carrageenan	Gellan
Total viable count (cfu/g)									
Zero	1.64×10^5	5.45×10^4	2.54×10^5	2.62×10^5	4.0×10^5	4.8×10^5	2.9×10^5	2.1×10^5	3.40×10^5
30	3.6×10^4	3.3×10^4	4.85×10^4	2.75×10^4	3.5×10^4	2.15×10^4	1.0×10^4	2.2×10^4	4.05×10^4
60	1.15×10^4	1.05×10^4	9.0×10^3	9.0×10^3	1.25×10^4	5.5×10^3	9.5×10^3	9.5×10^3	3.5×10^3
90	3.0×10^3	4.5×10^3	1.0×10^3	4.5×10^3	1.0×10^3	3.5×10^3	7.0×10^3	4.5×10^3	2.5×10^3
Total coliform count (cfu/g)									
Zero	7.0×10^3	3.0×10^3	1.0×10^3	6.0×10^3	4.5×10^3	6.0×10^3	4.0×10^3	1.05×10^3	1.20×10^3
30	2.4×10^2	7.25×10^2	4.2×10^2	1.0×10^3	1.7×10^2	1.45×10^2	2.45×10^2	2.55×10^2	1.95×10^2
60	1.5×10^2	3.15×10^2	1.8×10^2	1.9×10^2	1.2×10^2	5.0×10^1	1.7×10^2	2.05×10^2	1.15×10^2
90	6.0×10^1	8.0×10^1	5.5×10^1	3.5×10^1	1.5×10^1	2.0×10^1	2.5×10^1	1.5×10^1	1.0×10^1
Viable count (cfu/g)									
Zero	4.05×10^2	4.85×10^2	6.35×10^2	4.9×10^2	7.45×10^2	5.35×10^2	6.95×10^2	6.25×10^2	7.35×10^2
30	1.15×10^2	1.55×10^2	1.5×10^2	1.5×10^2	1.6×10^2	1.0×10^2	1.15×10^2	1.05×10^2	7.0×10^2
60	9.0×10^1	5.5×10^1	8.5×10^1	1.2×10^2	6.0×10^1	6.5×10^1	2.5×10^1	4.0×10^1	4.5×10^1
90	3.5×10^1	4.0×10^1	5.5×10^1	8.5×10^1	5.0×10^1	4.5×10^1	2.0×10^1	2.0×10^1	3.0×10^1

RPS: *Rhizobium* polysaccharide.

from 5.45×10^4 to 4.8×10^5 , 1.0 to 7.0×10^3 and 4.05 to 7.45×10^2 cfu/g of tested sausage samples, respectively. Storage of different samples at -18°C for 90 days considerably reduced the aforementioned microorganism counts to 1.0 to 7.0×10^3 , 1.0 to 8.0×10^1 and 2.0 to 8.5×10^1 cfu/g of the tested sausage samples, respectively. These records were within the range allowed by the Egyptian Organization of Standardization (EOSQC, 1999). These results agree with the results of Rao *et al.* (1984).

Sensory evaluation of beef sausage during storage:

Effect of substitution of 3.0% starch with 0.1% of different polysaccharides or mixture of 0.5% starch and 0.1% of different used polysaccharides on sensory properties of beef sausage was followed up during frozen storage of different samples for 90 days and the scores were statistically analyzed (data not showed). Sausage containing carrageenan and gellan gum were given similar or higher ratings than the control for most attributes at zero time of storage. However, the sensory panel assigned lower scores to samples containing xanthan and RPS, when compared to those containing carrageenan and gellan. Storage of beef sausages containing 3.0% starch or 0.1% of different polysaccharides at -18°C for 90 days significantly reduced most of the sensory attributes of cooked samples. Also, the rate of change in sensory properties was less pronounced for sausage samples containing 0.1% of different polysaccharides than control sample during frozen storage. However after 90 days of frozen storage the sensory panel assigned higher tenderness, juiciness, appearance, aroma, color and overall acceptability scores to samples containing 0.1% of gellan or carrageenan when compared to those containing xanthan, RPS and 3.0% starch (control). In conclusion, substitution of 3.0% starch with 0.1% gellan or carrageenan gums in beef sausage manufacture produced sausage with high acceptability, especially when samples stored under frozen conditions.

Sensory attributes of cooked beef sausages containing 0.5% starch and 0.1% polysaccharides showed similar trend with those contained only 0.1% of different polysaccharides during frozen storage. However, the sensory panel assigned higher appearance, aroma, color and overall acceptability scores to samples contained 0.1% gellan when compared to those containing a mixture of 0.1% gellan and 0.5% starch after 90 days of storage at -18°C .

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تحسين صفات السجق البقري باستخدام السكريات العديدة

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وجد أن استبدال الـ ٣% نشأ الموجودة في خلطة السجق البقري الكنترول بـ ٠,١% زانثان أو RPS (*Rhizobium polysaccharide*) أو جيلان مع ٠,٥% نشأ أعطي قيم حسية أعلى. سجلت عينات السجق البقري المحتوية على السكريات العديدة المختبرة قيم أعلى للمقدرة على الاحتفاظ بالماء والبلاستكية من العينة المقارنه عند بداية التجربة وأثناء التخزين المجمد (-١٨°م). على الرغم من كون عينة الكنترول سجلت أعلى حصيله طهي ابتدائي (٧٨,١١%) مقارنة بالعينات المحتوية على السكريات العديدة (٦٤,٥٤% - ٥٦,٩%) إلا أنه لوحظ إنخفاض قوي في حصيله الطهي وصل إلى ٦٠,٢٤% بعد التخزين المجمد. أدت إضافة السكريات العديدة المختبرة إلى تحسين قيم الصلابه للسجق الخام عن عينة الكنترول. وقد تم الوصول لأعلى تحسين لتقسيم الصلابه عند إضافة ٠,١% من كل من الجيلان والكارجينان في وجود أو عدم وجود ٠,٥% نشأ. ومن الناحية الأخرى فإن العينات المحتوية على أي من الـ RPS والكارجينان أو خليط من ٠,١% RPS و ٠,٥% نشأ أظهرت أقل فقد في السائل الناضح drip losses (٨,١٢% و ٩,٨٥% و ٨,٧٩% على التتابع) مقارنة بعينة الكنترول (٧,١٧%) وذلك بعد التخزين لمدة ٩٠ يوم على -١٨°م. ولهذا ينصح باستخدام ٠,١% من الـ RPS والزانثان والكارجينان والجيلان لتحسين صفات السجق البقري.