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EFFECT OF *IN SITU* EXOPOLYSACCHARIDE PRODUCTION ON SENSORY PROPERTIES OF TURKISH-TYPE FERMENTED SAUSAGE

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ABSTRACT

In this study, effect of *in situ* exopolysaccharide (EPS) production on sensory properties of Turkish-type fermented sausage (sucuk) was determined. Sausage mixes were prepared as four groups: (a) Control group, control sausage samples produced without LAB addition, with natural flora, (b) Strain 1 group, sausage samples produced by using Strain 1 (EPS$^+$ *Lactobacillus plantarum* 162 R strain), (c) Strain 2 group: sausage samples produced by using Strain 2 (EPS$^+$ *Leuconostoc mesenteroides* N6) and (d) Mixture group, sausage samples produced by using mixture of Strain 1 and Strain 2. These mixes were fermented at 14, 16 and 18°C for 8, 12 and 16 days. Fermentation conditions remarkably affected the sensory properties of the sausage using different EPS producing EPS. These results of this study demonstrated the importance of *in situ* EPS production on final sensory properties of sausage.
Keywords: Sausage, exopolysaccharides, sensory properties.

1. INTRODUCTION

It is known that Turkish style fermented sausage is very popular and the most consumed fermented meat product in Turkey [1]. The sausage has some critical quality characteristics such as color, texture, flavor, odor which were directly related with the fermentation conditions. Among of them are fermentation temperature, fermentation time along with use of starter LAB cultures having exopolysaccharide (EPS) production characteristics [1, 2].

Exopolysaccharides (EPS) have been reported to have unique characteristics based on differences in the sugar subunits and glycosidic linkages present in their repeating units. This explain the reason why there is a great diversity among bacterial EPS and novel EPS structures [3, 4]. EPS are well known to have some essential effects on physicochemical and textural properties of fermented food products especially dairy products as natural bio-thickening agents and *in situ* produced stabilizers [5]. Also, sensory properties are among the most important

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quality characteristics of the fermented sausages.

So far, a great number of studies have been reported to understand the functional characteristics of EPS produced by LAB strains in fermented dairy products. However, no study has appeared to uncover the role of *in situ* EPS production on sensory properties of fermented meat products such as fermented sausage. Therefore, in this study, we aimed to determine the effect of *in situ* EPS production on sensory properties of Turkish type sausage by using two different EPS producing strains and their mix. In addition, effect of different fermentation temperatures and time to understand the effects of fermentation conditions on sensory properties of sausage.

2. EXPERIMENTAL

2.1. Bacterial Strains

For preparation of sausage samples, EPS^+ strains (exopolysaccharide producing strains) were used. In this study, the sausage samples were produced and investigated as four different treatment groups, and will be referred as following throughout the manuscript:

(a) **Control group:** Control sausage samples produced without LAB addition, with natural flora.

(b) **Strain 1 group:** Sausage samples produced by using Strain 1 (EPS^+ *Lactobacillus plantarum* 162 R strain),

(c) **Strain 2 group:** Sausage samples produced by using Strain 2 (EPS^+ *Leuconostoc mesenteroides* N6),

(d) **Mixture group:** Sausage samples produced by using mixture of Strain 1 and Strain 2.

All strains were incubated in 10% reconstituted skim milk and stored at −70 ºC until further use. The stock cultures were activated in MRS medium at 37 ºC for 24 h and following another propagation in MRS medium all strains were inoculated to the sausage mix at 1% concentration for the fermentation of process.

2.2. Sausage Manufacturing

Fresh, boneless beef cuts (from middle-aged cows) with approximately 14% fat and sheep tail fats were obtained from a retail market (İstanbul, Turkey). Controlled fermentation process was achieved by using the aforementioned bacterial strains. The sausage samples were produced according to a general method used in Turkish sausage manufacturing plants. The sausage formulation and spice mixture were comprised of 90% beef, 10% tail fat, 2% salt, 1% garlic, 0.7% red pepper, 0.5% powdered black pepper, 0.9% cumin, 0.25% allspice, as outlined [6]. The physicochemical and microbiological properties of beef meat and tail fat as well as final numbers of bacterial strains in the sausage mix were shown in Table 1. The mix was separated into four groups each of which was inoculated with the respective bacterial culture mentioned above at 1% level and then each mix group was further kept for 30 h. Each mix group (control, strain 1, strain 2 and mixture groups) was separately ground through a grinder machine. Then, sausage batons were separated into 10 experimental batches and fermented according to experimental samples (S1–S10) each of which represents different processing conditions; namely, different fermentation conditions (fermentation temperature and time; Table 2). For this purpose, the batons were placed in the fermentation cabinet and ripened at respective temperature levels and for respective days.

Table 1. Physicochemical and microbiological properties of formulation components of sausage samples

	Physicochemical and microbiological properties			
Formulation components	Dry matter (%)	Protein (%)	Fat (%)	LAB number (log cfu/g)
Meat	22.35	20.06	0.96	1.42
Tail fat	85.96	2.35	83.1	–
Strain 1	–	–	–	7.5
Strain 2	–	–	–	7.8

LAB: lactic acid bacteria

Table 2. The samples fermented at different fermentation conditions

Samples	Fermentation temperature (°C)	Fermentation time (day)
S1	14	8
S2	14	12
S3	14	16
S4	16	8
S5	16	12
S6	16	12
S7	16	16
S8	18	8
S9	18	12
S10	18	16

2.3. Sensory Analysis

The sensory analysis of sausage samples were determined based on protocols described before [7]. Sensory tests were performed at 20–22 ºC in a well-ventilated room in artificial light and the temperature of the product was approximately ambient temperature. Sausage slices were 5 mm thick, cut with a knife and served at room temperature on white plastic dishes. Water and unsalted crackers were provided for panelists to rinse and clean their mouths between samples. Sensory analyses of the sausage samples were carried out by fifteen selected staff and graduate students of Food Engineering department at Yıldız Technical University, comprised of eight females and seven males. Each panelist was trained before evaluation in order to familiarize with the sensory analysis, samples and methodology. In this study, sensory analysis tests were conducted for both raw and cooked sausage samples. All coded raw sausage samples were evaluated for their exterior surface color, cross section color, typical sausage odor, appearance, texture and general acceptability while cooked sausage samples were tested for their color, typical sausage odor, appearance, taste, texture and general acceptability. The hedonic scale ranged from 1 to 9 points where 1 reflected a very low in terms of disliking and 9 a very high score in terms of liking. Panelists evaluated ten samples in two sessions (five at each session) consecutively in two days. The manner in which the treatment combinations were divided between the sessions and the order in which the samples were presented was randomized to minimize the carryover effects [8-9].

2.4. Statistical Analysis

The SPSS Statistics package (17.0; SPSS Statistics/IBM, Armonk, NY) was used to conduct an ANOVA to show the differences between experimental samples and between treatments ($P < 0.05$).

3. RESULTS AND DISCUSSION

The sensory properties of raw and cooked sausage samples produced with different EPS producer strains and under different fermentation conditions were also evaluated and results are given in Tables 3 and 4, respectively. As can be seen from the tables the sensory properties of sausage samples were found to be dependent on the conditions tested. In raw sausage samples the exterior surface and cross section color scores resulted in an increase after 12 d ripening compared to the 8 d but a further decrease was demonstrated after 16 d ripening period (Table 3). Previous knowledge also confirms this trend for the color scores of sausage samples which was related with the denaturation of color pigments at the end of ripening period [10,11]. But this trend was not observed for the sausage samples produced with EPS producer strains especially at 16°C and 18°C ripening temperature which appeared to be a positive factor for the final quality of sausage. But other than that, no fundamental effect was found in color scores of sausage samples depending on strain specific conditions. Similarly fermentation period was more effective factor than the fermentation temperature on odor, appearance, texture and general acceptability scores of sausage samples. In general these scores at 12 d fermentation period were higher than to that of 8 and 16 d fermentation periods (Table 3). Importantly although in general the texture perception of raw sausage samples produced with EPS producer strains were found to be higher than the control group but this effect was not presented as a trend as it was in the textural analysis of the sausage samples. This trend was also observed for the general acceptability of raw sausage samples (Table 3). Similarly fluctuations in the color and appearance scores of cooked sausage samples were observed and the lowest and the highest color and appearance scores of cooked sausage samples were 5.33-7.29, 4.63-7.20, respectively observed the lowest for than those of the control group and highest for EPS + group (Table 4). Generally, the odor and taste perceptions of the EPS + groups were higher than those of the control sausage samples. Similar to the raw sausage samples, the fluctuations were observed in the texture and general acceptability values of cooked samples between the treatment groups (Table 4). Overall textural properties of sausage samples somewhat were affected by the fermentation conditions as well as selection of LAB strains as a function of EPS production.

4. CONCLUSIONS

In this study we determined the effects of EPS production characteristics on sensory characteristics of sausage samples produced with different strain conditions with regards to EPS production characteristics under different fermentation conditions. The sensory analysis of the sausage samples exhibited some improvements, to some extent, in sausage texture depending on EPS production. Overall, the results of this study showed the importance of fermentation conditions on final quality of sausage and revealed the functional roles of *in situ* EPS production during the ripening of sausage.

Table 3. Differences between treatment groups for each sample with respect to sensory properties of raw sausage samples

	Treatments				**Treatments**			
	Control	**Strain 1**	**Strain 2**	**Mixture**	**Control**	**Strain 1**	**Strain 2**	**Mixture**
Samples	**Exterior surface color**				**Cross section color**			
S1	5.60^{Bg}	6.40^{Ae}	6.40^{Ad}	6.40^{Af}	5.60^{Cg}	6.40^{Ae}	6.40^{Ae}	6.20^{Bf}
S2	7.00^{Bb}	7.25^{Aa}	6.25^{Df}	6.88^{Cb}	7.13^{Ba}	7.38^{Aa}	6.00^{Dg}	6.63^{Cb}
S3	6.60^{Bd}	6.20^{Cf}	6.80^{Aa}	6.60^{Bd}	6.80^{Bd}	5.60^{Dh}	7.00^{Aa}	6.40^{Ce}
S4	5.07^{Cj}	4.67^{Dh}	5.73^{Ah}	5.27^{Bj}	5.13^{Cj}	4.87^{Di}	5.60^{Ah}	5.53^{Bi}
S5	7.08^{Aa}	6.42^{Bd}	6.17^{Cg}	6.42^{Be}	6.83^{Ac}	6.00^{Df}	6.50^{Cd}	6.58^{Bc}
S6	6.80^{Ac}	6.20^{Cf}	6.60^{Bc}	6.20^{Ch}	7.00^{Ab}	7.00^{Ab}	6.60^{Bb}	6.40^{Ce}
S7	6.13^{Ce}	7.13^{Ab}	6.63^{Bb}	6.63^{Bc}	5.63^{Cf}	7.00^{Ab}	6.50^{Bd}	6.50^{Bd}
S8	5.25^{Dh}	6.50^{Bc}	6.80^{Aa}	6.25^{Cg}	5.25^{Di}	6.75^{Ad}	6.20^{Bf}	6.00^{Cg}
S9	5.89^{Af}	5.44^{Cg}	5.44^{Ci}	5.67^{Bi}	6.22^{Ae}	5.89^{Bg}	5.44^{Di}	5.67^{Ch}
S10	5.13^{Di}	6.50^{Bc}	6.29^{Ce}	7.40^{Aa}	5.38^{Dh}	6.83^{Ac}	6.57^{Cc}	6.80^{Ba}
	Typical sausage odor				**Appearance**			
S1	5.60^{De}	6.40^{Bd}	5.80^{Cg}	6.80^{Ab}	5.20^{Ch}	6.60^{Ad}	6.40^{Bd}	6.40^{Bf}
S2	6.25^{Cd}	6.63^{Aa}	6.63^{Ad}	6.50^{Bc}	7.13^{Ab}	7.13^{Ab}	6.38^{Ce}	6.88^{Bb}
S3	6.40^{Cb}	6.00^{Df}	7.20^{Ba}	7.20^{Aa}	6.60^{Ce}	6.20^{Dg}	7.00^{Aa}	6.80^{Bc}
S4	5.27^{Cg}	5.53^{Bh}	5.80^{Ag}	5.27^{Ch}	5.33^{Bf}	5.07^{Ci}	5.67^{Ai}	5.67^{Ai}
S5	6.33^{Bc}	6.08^{Ce}	6.75^{Ac}	6.00^{Ce}	7.17^{Aa}	6.25^{Cf}	6.17^{Df}	6.42^{Be}
S6	7.40^{Aa}	6.60^{Cb}	7.00^{Bb}	6.00^{De}	6.80^{Ac}	6.40^{Be}	6.80^{Ab}	5.80^{Ch}
S7	5.50^{Cf}	6.50^{Ac}	6.25^{Bf}	6.25^{Bd}	5.25^{Cg}	6.75^{Ac}	6.50^{Bc}	6.50^{Bd}
S8	5.00^{Ci}	5.75^{Bg}	6.60^{Ae}	5.75^{Bf}	5.00^{Di}	6.75^{Bc}	7.00^{Aa}	6.00^{Cg}
S9	5.11^{Bh}	5.44^{Ai}	4.67^{Ci}	5.67^{Cg}	6.67^{Ad}	5.89^{Bh}	5.89^{Bh}	5.67^{Ci}
S10	4.38^{Dj}	6.00^{Bf}	5.57^{Ch}	6.80^{Ab}	4.88^{Dj}	7.50^{Aa}	6.14^{Cg}	7.40^{Ba}
	Texture				**General acceptability**			
S1	5.80^{Af}	5.80^{Af}	5.80^{Ag}	5.00^{Bi}	5.40^{Dg}	6.40^{Af}	6.20^{Cf}	6.40^{Be}
S2	6.75^{Cc}	7.38^{Aa}	6.25^{De}	7.00^{Bb}	6.63^{Cc}	7.25^{Aa}	6.13^{Dg}	6.75^{Bc}
S3	7.00^{Bb}	6.20^{Ce}	7.40^{Aa}	7.00^{Bb}	6.80^{Bb}	6.20^{Cg}	7.40^{Aa}	6.80^{Bb}
S4	5.33^{Bh}	5.07^{Di}	5.47^{Ah}	5.20^{Ch}	5.47^{Bf}	5.00^{Dj}	5.87^{Ah}	5.40^{Ch}
S5	6.58^{Ad}	5.42^{Dh}	6.50^{Bd}	6.42^{Cd}	7.00^{Aa}	6.08^{Dh}	6.50^{Cd}	6.58^{Bd}
S6	7.20^{Aa}	6.60^{Cb}	6.80^{Bb}	6.40^{De}	7.00^{Aa}	6.80^{Bb}	7.00^{Ab}	6.40^{Ce}
S7	5.88^{De}	6.25^{Bd}	6.00^{Cf}	6.50^{Ac}	5.75^{Ce}	6.75^{Ac}	6.50^{Bd}	6.75^{Ac}
S8	5.25^{Di}	6.25^{Bd}	6.80^{Ab}	6.00^{Cf}	5.00^{Dh}	6.50^{Be}	6.80^{Ac}	6.00^{Cf}
S9	5.56^{Bg}	5.78^{Ag}	5.00^{Ci}	5.78^{Ag}	5.89^{Ad}	5.67^{Ci}	5.78^{Bi}	5.89^{Ag}
S10	4.63^{Dj}	6.33^{Cc}	6.57^{Bc}	7.20^{Aa}	5.00^{Dh}	6.67^{Bd}	6.29^{Ce}	7.00^{Aa}

$^{A-D}$ Different uppercase superscript letters show differences between the treatments for each sample ($P < 0.05$).

$^{a-j}$ Different uppercase superscript letters show differences between the samples within the same treatment (control and strain groups) ($P < 0.05$).

Table 4. Differences between treatment groups for each sample with respect to sensory properties of cooked sausage samples

	Treatments				**Treatments**			
	Control	**Strain 1**	**Strain 2**	**Mixture**	**Control**	**Strain 1**	**Strain 2**	**Mixture**
Samples	**Color**				**Typical sausage odor**			
S1	6.20^{De}	6.40^{Cf}	7.00^{Ac}	6.60^{Bd}	6.60^{Dc}	6.80^{Cd}	7.20^{Ab}	7.00^{Bc}
S2	7.13^{Ab}	6.88^{Bb}	6.63^{Ce}	6.25^{De}	6.25^{Dd}	6.88^{Ab}	6.75^{Bd}	6.63^{Ce}
S3	6.20^{Ce}	6.60^{Be}	6.80^{Ad}	6.80^{Ab}	6.60^{Cc}	6.80^{Bd}	7.00^{Ac}	6.80^{Bd}
S4	5.33^{Ci}	5.27^{Dh}	5.87^{Aj}	5.60^{Bh}	5.53^{Df}	5.67^{Ch}	6.00^{Ah}	5.80^{Bi}
S5	7.00^{Ac}	6.17^{Dg}	6.50^{Cg}	6.67^{Bc}	6.83^{Ab}	6.17^{Df}	6.75^{Bd}	6.25^{Cg}
S6	7.20^{Aa}	6.80^{Bc}	7.20^{Ab}	6.60^{Cd}	7.40^{Aa}	7.20^{Ba}	7.40^{Aa}	7.20^{Bb}
S7	6.25^{Bd}	6.75^{Ad}	6.25^{Bh}	6.25^{Be}	5.88^{Ce}	6.75^{Ae}	6.13^{Bg}	6.13^{Bh}
S8	5.50^{Dg}	6.75^{Ad}	6.60^{Bf}	5.75^{Cg}	5.00^{Dh}	6.00^{Bg}	6.60^{Ae}	5.50^{Cj}
S9	5.56^{Cf}	5.22^{Di}	6.11^{Ai}	5.89^{Bf}	5.33^{Cg}	5.56^{Bi}	5.56^{Bi}	6.33^{Af}
S10	5.38^{Dh}	7.00^{Ca}	7.29^{Aa}	7.20^{Ba}	4.50^{Di}	6.83^{Bc}	6.57^{Cf}	7.60^{Aa}
	Appearance				**Taste**			
S1	6.00^{Be}	5.80^{Ch}	6.20^{Ag}	6.00^{Bf}	6.20^{Ce}	6.40^{Bf}	6.60^{Ag}	6.40^{Be}
S2	6.88^{Ab}	6.88^{Aa}	6.38^{Be}	5.88^{Ch}	6.75^{Bd}	7.00^{Aa}	6.63^{Cf}	6.50^{Dc}
S3	6.60^{Ad}	6.00^{Cg}	6.40^{Bd}	6.40^{Bc}	7.00^{Bc}	7.00^{Ba}	7.20^{Ab}	7.00^{Bb}
S4	5.00^{Ci}	5.00^{Cj}	5.40^{Bi}	5.53^{Aj}	5.33^{Di}	5.73^{Bg}	5.87^{Ai}	5.60^{Ch}
S5	7.00^{Aa}	6.08^{Df}	6.58^{Bc}	6.17^{Cd}	7.17^{Ab}	6.67^{Bb}	7.17^{Ac}	6.42^{Cd}
S6	6.80^{Cc}	6.80^{Cb}	7.20^{Aa}	7.00^{Bb}	7.60^{Aa}	6.60^{Dd}	7.40^{Ba}	7.00^{Cb}
S7	5.50^{Dg}	6.38^{Ae}	6.00^{Ch}	6.13^{Be}	6.13^{Cf}	6.63^{Bc}	6.75^{Ad}	5.88^{Df}
S8	5.25^{Dh}	6.50^{Ad}	6.00^{Bh}	5.75^{Ci}	5.50^{Dg}	6.50^{Ae}	6.20^{Bh}	5.75^{Cg}
S9	5.67^{Cf}	5.33^{Di}	6.33^{Af}	5.89^{Bg}	5.44^{Ah}	5.22^{Bh}	5.44^{Aj}	5.22^{Bi}
S10	4.63^{Dj}	6.67^{Cc}	6.71^{Bb}	7.20^{Aa}	4.63^{Dj}	6.67^{Cb}	6.71^{Be}	7.20^{Aa}
	Texture				**General acceptability**			
S1	5.40^{Cf}	5.40^{Ch}	5.80^{Ag}	5.60^{Bg}	6.00^{Ce}	6.20^{Bg}	6.60^{Af}	6.20^{Bf}
S2	7.13^{Ab}	6.75^{Bb}	6.50^{Cd}	6.25^{Dd}	6.75^{Bc}	6.75^{Ba}	6.88^{Ad}	6.50^{Cd}
S3	6.80^{Ac}	6.40^{Be}	6.80^{Ac}	6.80^{Ac}	6.60^{Bd}	6.60^{Bc}	7.00^{Ab}	7.00^{Ac}
S4	5.13^{Di}	5.40^{Bh}	5.20^{Ci}	5.47^{Ai}	5.13^{Dh}	5.53^{Ch}	5.80^{Aj}	5.67^{Bh}
S5	6.50^{Ad}	5.83^{Bf}	6.50^{Ad}	5.67^{Cf}	7.00^{Ab}	6.25^{Cf}	6.92^{Bc}	6.25^{Ce}
S6	7.40^{Aa}	6.80^{Da}	7.20^{Ba}	7.00^{Cb}	7.40^{Aa}	6.60^{Cc}	7.40^{Aa}	7.20^{Bb}
S7	5.38^{Cg}	6.50^{Ad}	5.75^{Bh}	5.75^{Be}	6.00^{Ce}	6.57^{Ad}	6.38^{Bh}	5.88^{Dg}
S8	6.00^{Ce}	6.75^{Ab}	6.20^{Be}	5.50^{Dh}	5.25^{Dg}	6.50^{Ae}	6.40^{Bg}	5.50^{Cj}
S9	5.33^{Ch}	5.67^{Bg}	5.89^{Af}	5.67^{Bf}	5.33^{Cf}	5.33^{Ci}	5.89^{Ai}	5.56^{Bi}
S10	3.88^{Dj}	6.67^{Cc}	7.00^{Bb}	7.40^{Aa}	4.38^{Di}	6.67^{Cb}	6.86^{Be}	7.40^{Aa}

$^{A-D}$ Different uppercase superscript letters show differences between the treatments for each sample ($P < 0.05$).
$^{a-j}$ Different uppercase superscript letters show differences between the samples within the same treatment (control and strain groups) ($P < 0.05$).

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