TITLE PAGE

- Korean Journal for Food Science of Animal Resources -

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ARTICLE INFORMATION	Fill in information in each box below
Article Type	Research article
Article Title	Evaluation of quality characteristics of low-nitrite pork sausages with paprika oleoresin solution during refrigerated storage
Running Title (within 10 words)	Low-nitrite pork sausages with paprika oleoresin solution
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Special remarks – if authors have additional information to inform the editorial office	
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Conflicts of interest List any present or potential conflict s of interest for all authors. (This field may be published.)	The authors declare no conflict of interest.
Acknowledgements State funding sources (grants, funding sources, equipment, and supplies). Include name and number of grant if available. (This field may be published.)	This study was not supported any funds.
Author contributions (This field may be published.)	Conceptualization: Geon Ho Kim, Koo Bok Chin Data curation: Geon Ho Kim Formal analysis: Geon Ho Kim, Koo Bok Chin Investigation: Geon Ho Kim Writing - original draft: Geon Ho Kim Writing - review & editing: Geon Ho Kim, Koo Bok Chin (This field must list all authors)
Ethics approval (IRB/IACUC) (This field may be published.)	This manuscript does not require IRB/IACUC approval because there are no human and animal participants.

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Abstract

The objective of this study was to evaluate quality characteristics of pork emulsified-sausage (ES) containing paprika oleoresin solution (POS) as a replacement for sodium nitrite ($NaNO_2$) during refrigerated storage. ESs were prepared with four treatments: 1) REF, 150 ppm $NaNO_2$; 2) CTL, 75 ppm $NaNO_2$; 3) TRT1, 75 ppm $NaNO_2$ + 0.1% POS (1% paprika oleoresin + 99% sunflower seed oil); and 4) TRT2, 75 ppm $NaNO_2$ + 0.1% POS (5% paprika oleoresin + 95% sunflower seed oil). The addition of POS into ES increased redness and yellowness but decreased lightness ($p<0.05$). TRT1 and TRT2 had higher redness and yellowness than CTL ($p<0.05$). TRT1 and TRT2 had lower total plate counts (log CFU/g) than CTL due to antimicrobial activity of POS, regardless of its levels ($p<0.05$). Residual nitrite decreased with increasing storage time for all treatments. TRT2 had lower residual nitrite, due to nitrite scavenging activity of POS ($p< 0.05$). CTL had the highest thiobarbituric acid reactant substances(TBARS) among all treatments during storage. The addition of POS into ES showed nitrite scavenging activity during refrigerated storage. In conclusion, antimicrobial and antioxidant activities of the ESs with a combination of POS and 75 ppm $NaNO_2$ were similar to those of REF(150 ppm $NaNO_2$), and improved color development of redness value. Therefore, the addition of POS could decrease the amount of nitrite in ESs, leading to healthier meat products.

Keywords: paprika oleoresin, sodium nitrite, low-nitrite, emulsified-sausage

1. Introduction

Sodium nitrite ($NaNO_2$), one of important ingredients in the manufacture of meat products, has many functions. It can react with myoglobin in meat products, resulting in the production of nitrosohemochrome, a pink pigment (Pearson and Gillett, 1996). Since meat color is an important factor for consumers to purchase the meat products, the role of sodium nitrite for the development of cured color cannot be ignored. In addition, nitrite can enhance the shelf-life by inhibiting microbial growth and toxin production of *Clostridium botulinum*. It also imparts a unique flavor of meat products (Christiansen et al., 1973; Hung et al., 2016). However, nitrite might not be completely depleted when it develops color. If it remains in a meat product during storage, N-nitrosamine, a carcinogen that can bind to amino acid and secondary amine during processing, might be generated (Hawksworth and Hill, 1971). Due to the risk of potential production of nitrosoamine from nitrites, typical cured meat products, such as ham and sausage, have been classified as first-class carcinogens designated by the World Health Organization. Consumers have a negative perception of meat products containing nitrite. Therefore, functional ingredients that can replace nitrite are needed for manufacturing meat products that health-conscious consumers would prefer. There have been a lot of studies on natural pigments, such as cactus pigment (Kang and Lee, 2008), red beet (Jeong et al., 2010), and Hongkuk (Liu et al., 2010) for the development of sausages with low levels of nitrite.

Paprika (*Capsicum annuum var. Angulosum*) contains a high amount of antioxidant substances such as carotenoid, a natural antioxidant that can prevent cancer, cell damage, and coronary artery disease (Dragsted et al., 1993). Paprika has a natural color (red, purple, or yellow). It contains a large amount of carotenoids that can be used in food, medicine, and food industry (Rascón et al., 2011). It has been reported that capsanthin increased redness value of meat products (Bázan Lugo et al., 2012). Thus, paprika is considered as a natural coloring agent to replace nitrite because it can increase the redness of meat products and prolong storage of meat products.

Paprika oleoresin that gives pigment to paprika extract is produced to improve the coloring effect of such paprika. It is processed in the form of oleoresin to preserve the color, taste, and flavor of paprika, which can be changed by heat and pH changes, after extracting paprika fruit with an organic solvent. Color stability of paprika oleoresin is higher than that of paprika fruit (Lee et al., 2002). Paprika oleoresin also contains a large amount of carotenoids. It has been widely used in the food industry, especially spices, soups, and meat products (Mínguez-Mosquera and Pérez-Gálvez, 1998; Yusop et al., 2012). The addition of paprika oleoresin to meat products might replace sodium nitrite for color development because it has higher color stability. In addition, paprika pigment is considered to be able to improve shelf-life of meat products through its antioxidant activity. However, the paprika oleoresin might evaluate to reduce sodium nitrite in pork sausage by improving both redness and shelf-life. Thus, the objective of this study was to determine quality characteristics of pork ES added with paprika oleoresin and propose appropriate levels of paprika oleoresin to be added in pork ES for developing heathier meat products.

2. Materials and Methods

2.1. Materials

Raw pork ham lean and back fat were purchased from a local meat market (Hyundai Retail Meat Market, Gwangju, Korea). Lean meat with external fat and connective tissue removed was ground with a meat chopper (M-12S, Korea Fuji Kogyo Co., Ltd., Busan, Korea) and stored frozen at -50°C until used for sausage manufacturing. Ground raw meat and back fat were stored refrigerated at 4°C for 24 h before sausages manufacturing. Paprika oleoresin was provided by Kalsec (Kalamanzoo, MI, USA). Sunflower seed oil was purchased from Sajo (Seoul, Korea) and used for preparing diluted paprika oleoresin solution (20 times and 100 times).

2.2. Experimental design

Formulation of pork sausages is listed in Table 1. In this experiment, paprika oleoresin solution was prepared by diluting paprika oleoresin and sunflower seed oil to avoid excessive increase of redness and yellowness from undiluted paprika oleoresin. Sunflower seed oil was extracted using organic solvents similar to paprika oleoresin (Salgin et al., 2006). It had no trans-fatty acids. However, it had linolenic acid, linoleic acid, unsaturated fatty acids, vitamin A, and vitamin D (Lee and Park, 2010). Pork ESs were prepared for the following four treatments: 1) REF, 150 ppm $NaNO_2$; 2) CTL, 75 ppm $NaNO_2$; 3) TRT1, 75 ppm $NaNO_2$ and 0.1% paprika oleoresin solution (1% paprika oleoresin + 99% sunflower seed oil); and 4) TRT2, with 75 ppm $NaNO_2$ and paprika oleoresin solution 0.1% (5% paprika oleoresin + 95% sunflower seed oil). Physicochemical chracteristics, textural properties, antioxidant activities, and antimicrobial activities of pork emulsified-sausages at 0, 3, 7, 14, 21, 28, and 35 days after manufacture were then determined.

2.3. Manufacture of sausages

Ground raw meat was mixed with an ice water for 30 sec using a hood mixer (Mixer, HMC-401, Hanil Electric Co., Seoul, Korea). The mixture and curing agents were then mixed with ice water for 1 min. After the addition of curing agents, back fat and paprika oleoresin solution were added. The mixture was then emulsified for 1 min. Finally, the meat batter was ground for 1 min. Stuffed meat batter in 50 mL conical tube (SPL Life Science Co., Ltd., Gyeonggi-do, Korea) was heated in a water bath (WB-22, Daihan Scientific Co., Ltd., Seoul, Korea) at 75℃ for 30 min, vacuum packaged, and refrigerated stored at 4℃ until analyzed.

2.4. pH and color determination

pH values were measured five times for the inside of each treatment using a pH meter (Model 340, Mettler-Toledo, Columbus, OH, USA). pH calibration was performed based on optimum slope measuring by pH 4.01 and 7.00 buffer.

Meat color was measured six times. Lightness (CIE L*), redness (CIE a*), and yellowness (CIE b*) of cross-sections of sausages were measured using a Minolta Color Reader (CR-10, Minolta Co., Tokyo, Japan). White color plate (CIE L*=94.8, CIE a*=1.0, CIE b*=0.1) was used as the standard.

2.5. Microbiological analysis

Homogenized sausage sample (10 g) and double-distilled(dd) water (90 mL) were mixed with a stomacher (BagMixer® 400 CC®, Interscience, Saint-Nom-la-Bretèche, France) for microbial counts. Total plate count (TPC) and violet red bile (VRB) agar plates were used for measuring numbers of total bacteria and *Enterobacteriaceae*, respectively. After spreading samples onto agar plates in petri dishes, plates were incubated at 37℃ in an incubator for 48 h.

2.6. Thiobarbituric acid reactive substances (TBARS)

Lipid oxiatation products were measured using the method of Sinnhuber and Yu (1977). Briefly, each sample was prepared by adding 1% of thiobarbituric acid solution (3 mL) to ground sausage (2 g). Then 2.5% of trichloroacetic acid solution (17 mL) was blended with the mixture in a glass tube. The tube was heated at 100 ℃ in a water bath (WB-22, Daihan Scientific Co., Ltd., Seoul, Korea) for 30 min. After heating, the supernatant of the heated mixture (5 mL) and 5 mL of chloroform were mixed together for 1 min and centrifuged at 1660 × g (Model VS-5500, Vision Science Co., Ltd., Korea) for about 5 min. The supernatant of the sample (3 mL) was blended with petroleum ether (3 mL) for 1 min. The mixture was then centrifuged at 1660 × g for 10 min. The absorbance of the reaction product was determined using a spectrophotometer (Model UV-1601, Shimadzu, Kyoto, Japan) at a wavelength of 532 nm. TBARS value was derived by calculating the measured absorbance with the following equation:

$$\text{TBARS value(mg of MDA/of sample)} = \frac{O.D.value \times 9.48}{Sample\ \ weight\ \ (g)}$$

The constant 9.48 was obatained from the reaction product of red thiobarbituric acids's sample dilution factor and its absorption coefficient (152,000 M^{-1} cm^{-1}).

2.7. Expressible moisture (EM, %)

For EM, each sample (1.5 g) was prepared by shaping into a rectangular shape. The sample was covered with three pieces of filter paper (Whatman #3, GE Healthcare, Little Chalfont, UK), placed in the bottom of a conical tube, and centrifuged at 1660 × g (VS-5500, Vision Science Co., Ltd., Gyeongsan, Korea) for 15 min. After the centrifugation was completed, EM was determined based on changed weight of the sample and the filter paper using the following formula:

$$\text{Expressible moisture (\%)} = \frac{\text{Expressible water weight of filter paper (g)}}{\text{Sample weight (g)}} \times 100$$

2.8. Proximate composition

Proximate composition was performed according to methods of the Association of Offical Analytical Chemists (AOAC, 2000). Moisture content (%) was measured with the dry oven method. Briefly, 2 g of each sample in a paper thimble was dried for 18 h in a dry oven at 102 ℃ and the weight of the samples was measured after drying. Crude fat content (%) was measured with the soxhlet extraction. Briefly, the paper thimble after moisture drying was placed in soxhlet's extractor, and fat in the samples was extracted with petroleum ether to measure the weight loss. Crude protein content (%) was determined based on the Kjeldahl method.

2.9. Cooking loss

Cooking loss was expressed in percentage (%) by calculating the weight of sausage before and after heating with the following formula.

$$\text{Cooking loss } (\%) = 100 - \left(\frac{Sample\ weight\ after\ heating\ (g)}{Sample\ weight\ before\ heating\ (g)} \times 100\right)$$

2.10. Texture profile analysis

For texture profile analysis, sausage samples with 1.3 cm in height and 1.25 cm in diameter were prepared. Texture properties such as hardness (gf), springiness (mm), gumminess, chewiness, and cohesiveness of each sample were determined with a bellow cross head compression probe (cross speed of 300 mm/min and load cell of 50 kg) using an Instron Universal Machine (Model 3344, Instron Co., Ltd., Canton, MA, USA).

2.11. Residual nitrite

Residual nitrite content was measured by a modified method of AOAC (2000). Briefly, homogenized sausage sample (5 g) was blended with double-distilled water (300 mL), and heated in a water bath (WB-22, Daihan Scientific Co., Ltd., Seoul, Korea) for 1 h at 100 ℃. After heating, filteration was performed using a funnel and a filter paper (Whatman #2, GE Healthcare, Little Chalfont, UK) to remove sausage grounds. Filtrated sample was diluted with distilled water to make a mixture of 500 mL. Sulfanilamide solution (2.5 mL) was added to 25 mL of this mixture, blended, and incubated at room temperature for 5 min for reactions to occur. Then, N-(1-Naphthyl)ethylene dihydrochloride solution (2.5 mL) was added to the reactant, blended, and incubated at room temperature for 15 min. The absorbance of the final reaction product was measured with a spectrophotometer (Model UV-1601, Shimadzu, Kyoto, Japan) at wavelength of 540 nm. A standard curve was obtained by measuring the absorbance of nitrite solution to derive residual amount of nitrite. It was used to determine residual amount of nitrite based on the measured absorbance of the sample.

2.14. Statistical analysis

All experiments for this study were carried out in triplicate for each sample of treatments. Data were represented as mean and standard deviation and were analyzed using two-way analysis of variance (ANOVA) and IBM SPSS Statistics 23 (SPSS Inc., Chicago, IL, USA) with Duncan's multiple range test and a significance level of 5% ($p<0.05$). If a significant interaction between treatments and storage time was observed, means and standard deviations were separated by treatments within each storage time and by storage time within each treatment. When the interaction was in significant ($p>0.05$), data were pooled to test the primary effect by each main factor.

3. Results and discussion

3.1. pH and color determination

Table 2 shows results of pH of pork ESs added with POS during refrigerated storage. Since no interaction between treatment and storage time was found ($p>0.05$), data were pooled by treatment within storage time and storage time within treatment. For pork ESs, their pH values ranged from 6.06 to 6.09, showing no difference among ESs ($p>0.05$). In a study of Eskandari et al. (2013), pH values of frankfurters added with 0.1 or 1.5% of paprika extract were not different from the pH values of frunfurters without adding paprika extract ($p>0.05$). Similar to results of their study, our results also show that the addition of paprika extract did not affect pH values of meat products ($p>0.05$).

As shown in Table 2, results of color determination were pooled by treatment within storage time and storage time within treatment due to no interaction between treatment and storage time was found ($p>0.05$). Lightness (L*) values of sausages were higher for the CTL than those of the treatments added with 75 ppm of nitrite alone ($p<0.05$). However, lightness (L*) values of sausages in REF and TRT2 were not different from each other ($p>0.05$), although they were lower than those of other treatments ($p<0.05$). The reason for the difference in L* values among CTL, TRT1, and TRT2 with the same amount of nitrite added might be partially due to a decrease in lightness with the addition of diluted paprika oleoresin solution. A previous study has reported that 15 or 30 g/kg of paprika powder decreased L* values of dry-cured sausages at 20 mm of meat mincing level (Fernández-López et al., 2002). Redness (a*) values of sausages in the TRT2 added with 75 ppm of nitrite and 0.1% of paprika oleoresin solution (paprika oleoresin 5% and sunflower seed oil 95%) were higher than those of the REF added with 150 ppm of nitrite alone ($p<0.05$). The addition of paprika oleoresin resulted in higher redness values ($p<0.05$). Thus, the addition of paprika oleoresin could increased the redness of pork emulsified-sausage, however, redness values showed no change during the storage time ($p>0.05$). The addition of paprika powder (0.1, 0.5, or 2%) into fresh pork sausages also increased a* values in the study of Martínez et al. (2006). Paprika has a high amount of ketocarotenoids such as capsorubin and capsanthin (Levy et al., 1995). These carotenoids, called red xanthophylls (Minguez-Mosquera et al., 1992), are specific red compounds that can increase the redenss of foods including meat products. Yellowness (b*) values of TRT2 were the highest in the TRT2 in among all treatments ($p<0.05$). The addition of diluted paprika oleoresin also increased yellowness ($p<0.05$), which showed no change during the storage time ($p>0.05$). According to a study of Jokanović et al.(2011), the addition of 1% of paprika oleoresin increased the yellowness of marinated chicken breast. Thus, meat products with paprika oleoresin increased their yellowness values, as well.

3.2. Microbiological anaylsis

As shown in Table 3, there was no interaction between treatments and storage time in results of microbiological analysis ($p>0.05$). TPC in the CTL were higher than those in other treatment groups ($p<0.05$). However, TRT1 and TRT2 were lower than those with TPC, indicating that paprika oleoresin might have an antibacterial effect. Salih (2006) reported that oil extract from paprika possessed antimicrobial activity against *Staphylococcus aureus*, *Escherichia coli*, *Pseudomonas aeruginosa*, and *Klebsiella pneumoniae* due to the presence of capsanthin from paprika. In addition, oleoresins including paprika extract at concentration above 5000 ppm had antimicrobial effects by fracturing bacterial membrane. This mechanism can inhibit the growth of foodborne pathogens such as *Listeria monocytogens*, *Staphylococcus aureus*, *Bacillus cerus*, *Salmonella*, and *Pseudomosnas aeruginosas* (Dussault et al., 2014). However, microbial counts of *Enterobacteriaceae* (VRB) were not changed during storage for any treatment ($p>0.05$). Both TPC and VRB anlaysis results increased with increasing storage time ($p<0.05$).

3.3. TBARS

Table 3 shows results of TBARS of pork emulsified-sausages added with POS during storage time. Data were pooled by treatment within storage time and storage time within treatment due to no interaction between treatment and storage time was found (p>0.05). Although no difference in the TBARS values were observed among REF, TRT1, and TRT2 (p>0.05), TBARS values of the CTL were the highest among treatments (p<0.05). These results indicated that the addition of paprika oleoresin could inhibit lipid oxidation during storage. Antioxidant activity and ilipid oxidaition inhibited the ability of paprika as reported by previous studies. *Capsicum annuum* group including paprika contained a great antioxidant activity from rich polyphenol pattern (Marin et al., 2004). When paprika extract is added to meat products, it can be expected to inhibit lipid oxidation. In fact, Shim and Chin (2013) reported that pork patties containing oven-dried paprika powder (0.5 and 1.0%) had lower TBARS values than control without paprika powder. Kim et al. (2013) have also reparted that the lipid oxidation of pork patties added with 0.323% of paprika oleoresin for nitrite replacement was lower than those of the control without adding paprika oleoresin during refrigerated storage. Carotenoids as, pigment present in paprika, have a radical scavenging ability. They can function as antioxidants by physically trapping singlet oxygen (Kiritsakis and Dugan, 1985). And, carotenoids in liposome systems reported to increase antioxidant acitivity with a decrease of oxygen content (Kiokias and Gordon, 2004). TBARS value started to increase rapdily from day 14 of storage (p<0.05). They continued to increase and tended to be the highest at day 35 of storage (p<0.05).

3.4. Expressible moisture (EM, %)

Results of EM are shown in Table 2. There was no interaction between treatments and storage time in results of EM (p>0.05). EMs of sausages in REF and CTL were not significantly different(p>0.05). However, EM values in the TRT2 were higher than those of REF and CTL (p<0.05). During storage, EM (%) showed an increase from day 21 (p<0.05). It continued to increase up to day 35 during storage. Bázan-Lugo et al. (2012) have reported that sausage batter added with more than 1% of paprika powder have higher EM values than those without adding paprika powder. These results were different from our results due to the differences in the structure of paprika powder and oleoresin type.

3.5. Proximate composition

Table 4 shows proximate composition. Moisture, fat and protein contents (%) were not different among treatments (p>0.05). The moisture content of ESs ranged from 62.2 to 62.9%, fat content ranged from 18.1 to 18.8%, and the protein content ranged from 13.9 to 14.0% . These results indicated that lower level (0.1%) of POS might not affect the proximate composition of ES. Yusop et al. (2012) have reported that the addition of 1 to 3% of paprika oleoresin does not affect moisture or fat contents of marinated chicken breasts. Therfore, the additional levels of paprika oleoresin might not be enough to change proximate composition.

3.6. Cooking loss (CL, %)

Cooking loss values of pork emulsified-sausages added with paprika oleoresin solution are shown in Table 4. There was no differences in CL among the treatments (p>0.05). Kim and Chin (2018) have also reported that cooking losses of pork low-fat sausages added with paprika powder (0.05% or 0.1%) were not different from those of the control without the addition of paprika powder. These results indicate that paprika pigment does not affect cooking loss of pork ESs.

3.7. Texture profile analaysis

Results of texture profile analysis of pork emulsified-sausages added with POS are shown in Table 4. There were no differences in all textural properties (hardness, springiness, gumminess, chewiness and cohesiveness) among treatments ($p>0.05$). Bázan-Lugo et al. (2012) have also detected that there were no differences in hardness value between sausage added with 1% of paprika powder and control (0% of paprika powder). However, treatment added with 2% of paprika powder had a higher hardness than control and 1% treatment. It indicates that the ingredients of paprika changed texture of sausage due to retain the water levels during the heating. Thus, the addition of 0.1% of POS into ESs might not be enough to change texture properties of those in our study.

3.8. Residual nitrite

Table 5 shows residual nitrite levels in pork emulsified-sausages added with paprika oleoresin solution. As expected, residual nitrite levels (ppm) in the REF were the highest among all treatments ($p<0.05$), and those in the CTL, TRT1 and TRT2 showed no differences until 21 days ($p>0.05$). Residual nitrite levels in the TRT2 at 28 and 35 days were lower than those of the CTL ($p<0.05$), indicating that the paprika oleoresin could reduce residual nitrite levels in the TRT2. Jeong et al. (2006) have reported that paprika with higher antioxidant capacity possess higher nitrite scavenge activity. Paprika contains phenolic compounds, ascorbic acid, flavonoid, and carotenoid pigments including capsanthin, capsorubin, and cryptocapsin (Baliga et al., 2003; Zhang and Hamauzu, 2003). Since they are effective in scavengering free radicals, they could inhibit oxidation during storage due to their antioxidant activity. These results indicate that antioxidants in paprika oleoresin can scavenge residual nitrite, thus decreasing residual nitrite levels.

Conclusion

The addition of 0.1% POS into ES can increase the redness and yellowness values, but decrease the lightness. Total plate counts in TRT1 and TRT2 were lower than those in the CTL. TBARS and residual nitrite values for TRT1 and TRT2 were lower than those of the CTL. Nitrite scavenging activity and antioxidant activity were observed in treatments added with POS, regardless of the concentration of paprika oleoresin. These results indicate that the addition of POS into ES can increase redness and yellowness values, inhibit lipid oxidation and growth of microorganisms, and accelerate nitrite scavenging. Therefore, combination of 75 ppm ppm nitrite level and 0.1% POS could be used to manufacture sausages healthier than those added with nitrite at 150 ppm.

Acknoledgement

This study was supported by a project (PJ01380922018) of the Rural Development Administration program.

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Table 1. The formulation for manufacturing pork emulsified-sausages added with different contents of oleoresin paprika solution

Ingredients(%)	Treatment			
	REF	CTL	TRT1	TRT2
1. Lean meat	60.0	60.0	60.0	60.0
2. Fat	20.0	20.0	20.0	20.0
3. Water	18.0	18.0	18.0	18.0
4. Non meat ingredients	1.97	1.96	2.06	2.06
1) Salt	1.50	1.50	1.50	1.50
2) Sodium tripolyphosphate	0.40	0.40	0.40	0.40
3) Sodium erythorbate	0.05	0.05	0.05	0.05
4) Sodium nitrite	0.015	0.0075	0.0075	0.0075
5) Paprika oleoresin solution	0.00	0.00	0.10	0.10
- Paprika oleoresin	0.00	0.00	0.001	0.005
- Sunflower seed oil	0.00	0.00	0.099	0.095
Total	100.0	100.0	100.1	100.1

Table 2. pH and color values of pork emulsified-sausages added with paprika oleoresin solution

Treatments[1)]	pH	CIE L*	CIE a*	CIE b*
REF	6.07±0.05[a]	73.6±0.64[c]	11.8±0.25[b]	6.10±0.29[c]
CTL	6.06±0.07[a]	75.8±0.65[a]	9.90±0.50[d]	5.53±0.35[d]
TRT1	6.09±0.03[a]	74.7±1.19[b]	11.1±0.60[c]	6.37±0.34[b]
TRT2	6.08±0.05[a]	73.9±1.06[c]	12.1±0.06[a]	6.75±0.34[a]
Storage time (days)				
0	6.08±0.03[a]	74.7±0.89[a]	11.1±1.11[a]	6.19±0.55[abc]
3	6.10±0.03[a]	74.6±1.30[a]	11.6±1.08[a]	6.00±0.50[c]
7	6.08±0.02[a]	74.6±1.50[a]	11.6±1.16[a]	6.14±0.64[bc]
14	6.09±0.03[a]	74.6±1.43[a]	11.3±1.30[a]	6.10±0.50[bc]
21	6.08±0.04[a]	74.3±1.43[a]	11.2±1.04[a]	6.15±0.62[bc]
28	6.04±0.08[a]	74.2±1.33[a]	11.4±0.87[a]	6.32±0.54[ab]
35	6.05±0.08[a]	74.5±1.19[a]	11.2±0.88[a]	6.40±0.50[a]

[a,b,c,d] Means having the same superscripts in the same column are not significantly different($p>0.05$).

[1)] Treatments: REF, Emulsified-sausage (ES) added with 150 ppm sodium nitrite ($NaNO_2$); CTL, ES added with 75 ppm $NaNO_2$; TRT1, ES added with 75 ppm NaNO2 + 0.1% paprika oleoresin solution (POS, 1% paprika oleoresin + 99% sunflower seed oil); TRT2, ES added with 75 ppm $NaNO_2$ + 0.1% POS (5% paprika oleoresin + 95% sunflower seed oil).

Table 3. Microbial counts, TBARS, and expressible moisture of emulsified-sausages added with different contents of paprika oleoresin solution

Treatments[1)]	Total plate counts	*Enterobacteriaceae* counts	TBARS[2)]	EM[3)]
REF	$<2.00^{b}$	$<2.00^{a}$	0.19 ± 0.08^{b}	20.0 ± 0.14^{a}
CTL	2.07 ± 1.18^{a}	$<2.00^{a}$	0.24 ± 0.12^{a}	20.3 ± 0.35^{a}
TRT1	$<2.00^{b}$	$<2.00^{a}$	0.18 ± 0.07^{b}	18.9 ± 0.57^{ab}
TRT2	$<2.00^{b}$	$<2.00^{a}$	0.16 ± 0.06^{b}	18.2 ± 1.06^{b}
Storage time (days)				
0	$<2.00^{e}$	$<2.00^{c}$	0.09 ± 0.01^{e}	17.3 ± 0.48^{c}
3	$<2.00^{e}$	$<2.00^{c}$	0.13 ± 0.02^{e}	18.0 ± 0.89^{c}
7	$<2.00^{e}$	$<2.00^{c}$	0.16 ± 0.03^{de}	18.5 ± 2.31^{c}
14	$<2.00^{e}$	$<2.00^{c}$	0.20 ± 0.02^{cd}	18.1 ± 1.65^{bc}
21	2.31 ± 1.27^{c}	$<2.00^{c}$	0.21 ± 0.03^{c}	19.7 ± 2.46^{b}
28	3.53 ± 0.74^{b}	2.80 ± 0.86^{b}	0.27 ± 0.07^{b}	21.5 ± 1.84^{a}
35	4.18 ± 0.81^{a}	3.72 ± 0.86^{a}	0.31 ± 0.10^{a}	22.5 ± 1.05^{a}

[a,b,c,d,e] Means having the same superscripts in the same column are not significantly different ($p>0.05$).

[1)]Treatments: REF, Emulsified-sausage (ES) added with 150 ppm sodium nitrite ($NaNO_2$); CTL, ES added with 75 ppm $NaNO_2$; TRT1, ES added with 75 ppm NaNO2 + 0.1% paprika oleoresin solution (POS, 1% paprika oleoresin + 99% sunflower seed oil); TRT2, ES added with 75 ppm $NaNO_2$ + 0.1% POS (5% paprika oleoresin + 95% sunflower seed oil).

[2)]TBARS: thiobarbituric acid reactive substances (mg/100 kg)

[3)]EM: expressible moisture

Table 4. Proximate composition and functionality of pork emulsified-sausages added with paprika oleoresin solution

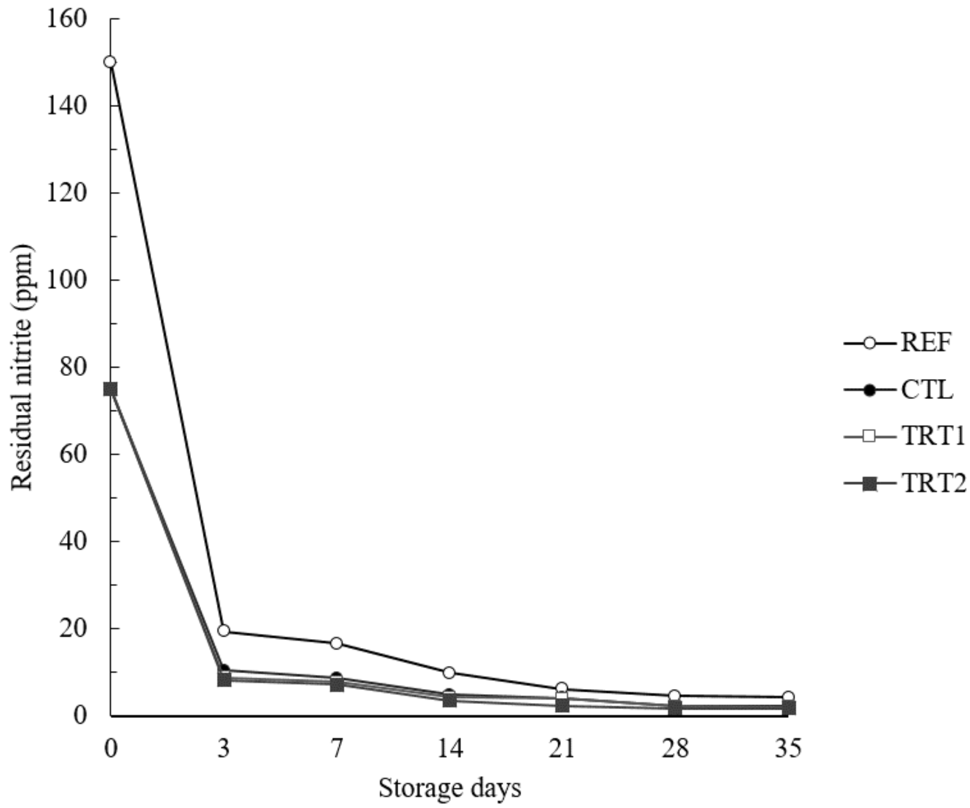
	Treatments[1)]			
	REF	CTL	TRT1	TRT2
Moisture(%)	62.2±0.66[a]	62.2±1.10[a]	62.9±1.45[a]	62.8±1.27[a]
Fat(%)	18.1±0.67[a]	18.8±0.84[a]	18.2±0.66[a]	18.5±0.79[a]
Protein(%)	13.9±0.10[a]	13.9±0.17[a]	14.0±0.13[a]	13.9±0.10[a]
Cooking Loss(%)	1.95±1.14[a]	1.93±0.59[a]	2.13±1.45[a]	1.90±0.77[a]
Hardness(gf)	2861±733[a]	2666±602[a]	2833±632[a]	3030±574[a]
Springiness (mm)	5.38±0.90[a]	5.55±0.88[a]	5.34±0.13[a]	5.15±0.20[a]
Gumminess	24.4±5.56[a]	21.0±6.01[a]	19.8±2.75[a]	22.8±3.11[a]
Chewiness	133±9.50[a]	105±19.8[a]	108±19.4[a]	119±19.1[a]
Cohesiveness	0.01±0.00[a]	0.01±0.00[a]	0.01±0.00[a]	0.01±0.00[a]

[a] Means having the same superscripts in the same column are not significantly different ($p>0.05$).
[1)] Treatments: REF, Emulsified-sausage (ES) added with 150 ppm sodium nitrite ($NaNO_2$); CTL, ES added with 75 ppm $NaNO_2$; TRT1, ES added with 75 ppm NaNO2 + 0.1% paprika oleoresin solution (POS, 1% paprika oleoresin + 99% sunflower seed oil); TRT2, ES added with 75 ppm $NaNO_2$ + 0.1% POS (5% paprika oleoresin + 95% sunflower seed oil).

Table 5. Residual nitrite of pork emulsified-sausages added with paprika oleoresin solution

Treatments[1)]	Storage time (days)						
	0	3	7	14	21	28	35
REF	22.1±2.09^{aA}	19.4±2.31^{bA}	16.7±2.10^{cA}	9.88±0.82^{dA}	6.23±2.48^{eA}	4.64±0.48^{eA}	4.25±0.46^{eA}
CTL	10.8±1.37^{aB}	10.4±1.10^{abB}	8.70±1.78^{bB}	4.91±1.27^{cB}	4.16±1.54^{cdAB}	2.29±0.51^{dB}	2.17±0.43^{dB}
TRT1	9.84±0.96^{aB}	8.89±0.65^{abB}	7.77±1.08^{bB}	4.29±0.99^{cB}	4.04±1.61^{cAB}	2.22±0.52^{dBC}	2.12±±0.55^{dBC}
TRT2	8.76±0.46^{aB}	8.24±1.09^{bB}	7.26±1.16^{bB}	3.60±0.98^{cB}	2.40±0.82^{dB}	1.84±0.58^{dC}	1.75±0.44^{dC}

^{A,B,C} Means having same superscripts in a same column are not different ($p>0.05$).
^{a,b,c,d,e} Means having same superscripts in a same row are not different ($p>0.05$).
[1)]Treatments: REF, Emulsified-sausage (ES) added with 150 ppm sodium nitrite ($NaNO_2$); CTL, ES added with 75 ppm $NaNO_2$; TRT1, ES added with 75 ppm NaNO2 + 0.1% paprika oleoresin solution (POS, 1% paprika oleoresin + 99% sunflower seed oil); TRT2, ES added with 75 ppm $NaNO_2$ + 0.1% POS (5% paprika oleoresin + 95% sunflower seed oil).



Residual nitrite (ppm)
160
140
120
100
80
60
40
20
0
0
3
7
14
21
28
35
Storage days
REF
CTL
TRT1
TRT2