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<b>Author</b>	Jiye Yoon, Su Min Bae, Seung Hwa Gwak, and Jong Youn Jeong
<b>Affiliation</b>	Department of Food Science & Biotechnology, Kyungsoong University, Busan 48434, Korea
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<b>ORCID (All authors must have ORCID) <a href="https://orcid.org">https://orcid.org</a></b>	Jiye Yoon (0000-0003-4781-6552) Su Min Bae (0000-0002-9367-4594) Seung Hwa Gwak (0000-0003-4975-1641) Jong Youn Jeong (0000-0001-5284-4510)
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**CORRESPONDING AUTHOR CONTACT INFORMATION**

For the <u>corresponding</u> author (responsible for correspondence, proofreading, and reprints)	Fill in information in each box below
First name, middle initial, last name	Jong Youn Jeong
Email address – this is where your proofs will be sent	jeongjy@ks.ac.kr
Secondary Email address	nexoxen@naver.com
Postal address	Department of Food Science & Biotechnology, Kyungsoong University, Busan 48434, Korea

Cell phone number	+82-10-9533-4032
Office phone number	+82-51-663-4711
Fax number	+82-51-622-4986

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# 9 Use of green tea extract and rosemary extract in naturally cured 10 pork sausages with white kimchi powder

## 11 Abstract

12 The impact of green tea extract powder and rosemary extract powder, alone or in combination,  
13 on the quality characteristics of naturally cured pork sausages produced with white kimchi  
14 powder as a nitrate source was evaluated. Ground pork sausages were assigned to one of seven  
15 treatments: control (0.01% sodium nitrite and 0.05% sodium ascorbate), treatment 1 (0.3%  
16 white kimchi powder and 0.05% green tea extract powder), treatment 2 (0.3% white kimchi  
17 powder and 0.1% green tea extract powder), treatment 3 (0.3% white kimchi powder and 0.05%  
18 rosemary extract powder), treatment 4 (0.3% white kimchi powder and 0.1% rosemary extract  
19 powder), treatment 5 (0.3% white kimchi powder, 0.05% green tea extract powder, and 0.05%  
20 rosemary extract powder), and treatment 6 (0.3% celery juice powder, 0.05% green tea extract  
21 powder, and 0.05% rosemary extract powder). Naturally cured products had lower ( $p < 0.05$ )  
22 cooking yield and residual nitrite content than control sausages. However, compared to the  
23 control, naturally cured products with white kimchi powder (treatments 1 to 5) showed similar  
24 ( $p > 0.05$ ) the pH, oxidation-reduction potential, CIE L\* values, CIE a\* values, nitrosyl  
25 heme content, total pigment content, and curing efficiency to the control. When the  
26 amount of green tea extract powder or rosemary extract powder was increased to 0.1%  
27 (treatments 2 and 4), lipid oxidation was reduced ( $p < 0.05$ ). These results indicate that green tea  
28 extract powder, rosemary extract powder, and white kimchi powder may provide an effective  
29 solution to replace synthetic nitrite and ascorbate used in traditionally cured products.

30 **Keywords:** naturally cured meat, sodium nitrite, white kimchi powder, green tea extract powder,  
31 rosemary extract powder

## Introduction

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The demands of consumers interested in health are causing clean-label trends such as organic and preservative-free foods to emerge in the food industry (Asioli et al., 2017; Yong et al., 2020). In the meat-processing industry, nitrite is widely used as an essential food additive that develops the typical cured meat color and flavor, improves microbiological safety, and provides antioxidant activity (Honikel et al., 2008; Pegg and Shahidi, 2000; Sebranek, 2009; Sebranek and Bacus, 2007a). However, nitrite ( $\text{NaNO}_2$  or  $\text{KNO}_2$ ), which is traditionally used as a curing agent, is a chemical additive and has become a factor for consumers to avoid the consumption of processed meat products. Moreover, nitrite is difficult to replace because it has simultaneous multifunctional activity (Alahakoon et al., 2015). Thus, a new category of meat products has emerged, such as so-called “naturally cured meat products” or “indirectly cured meat products,” which replace synthetic nitrite with plant-based ingredients such as vegetable powder or concentrate (Sebranek and Bacus, 2007a; Sebranek and Bacus, 2007b; Sindelar and Houser, 2009). Meat products in this category generally use vegetables that are rich in nitrate as a natural nitrate source. In this process, a starter culture such as *Staphylococcus carnosus*, which has nitrite-reducing activity, facilitates the production of nitrite from natural nitrate (Sebranek and Bacus, 2007a). As a result, naturally cured products have similar quality characteristics to traditionally cured products (Choi et al., 2020; Jeong et al., 2020a; Jeong et al., 2020b; Krause et al., 2011; Sebranek and Bacus, 2007a; Sucu and Turp, 2018). However, most naturally cured meat products generally have a lower residual nitrite concentration and relatively large variation compared to traditionally cured meat products (Sebranek and Bacus, 2007a; Sindelar et al., 2007; Yong et al., 2021). For this reason, products containing natural nitrite/nitrate derived from vegetables may have reduced antioxidant capacity and may be less able to inhibit the growth of

56 pathogenic bacteria. Consequently, there is a need to overcome the challenge of reduced nitrate  
57 levels in meat products cured using natural ingredients rather than synthetic additives.  
58 Overcoming this challenge will improve the quality of clean-label meat products.

59 Reducing compounds such as sodium ascorbate and sodium erythorbate are widely used for  
60 meat curing. These compounds act as cure accelerators that promote the reduction of nitrite to  
61 nitric oxide during the meat-curing step (Sebranek, 2009). However, since most of these  
62 compounds are also chemical additives, it is necessary to find alternative means to produce  
63 clean-label meat products. In this respect, the application of plant extracts and ingredients may  
64 appear to be suitable solutions to replace nitrite and ascorbate in meat products (Ferysiuk and  
65 Wójciak, 2020, Kumar et al., 2015). Green tea extract is a natural antioxidant owing to the  
66 presence of polyphenols, including flavanols, flavadiols, flavonoids, and phenolic acids  
67 (Chacko et al., 2010). These polyphenolic compounds eliminate free radicals and chelate metals,  
68 thereby inhibiting lipid oxidation and extending the shelf life of meat and meat products (Shah  
69 et al., 2014). According to Choi et al. (2003), the use of green tea powder in pork sausages  
70 resulted in lower levels of thiobarbituric acid-reactive substances (TBARS) than in sausages  
71 with nitrite. Moreover, Price et al. (2013) found that green tea extract more effectively prevents  
72 lipid oxidation than sodium nitrite in cooked pork meatballs, suggesting that green tea extract  
73 could be a promising substitute to ascorbate in terms of antioxidant effects. Rosemary extract  
74 is another plant-based ingredient that contains high levels of phenolic substances such as  
75 phenolic diterpenes and phenolic acid, which confer strong antioxidant capacity (Brewer, 2011;  
76 Zhang et al., 2010). Thus, as an antioxidant and metal chelator, rosemary extract can be used to  
77 inhibit lipid oxidation and protect the flavor of meat products. Nissen et al. (2004) evaluated  
78 the antioxidative activity of rosemary, green tea, coffee, and grape skin in cooked pork patties  
79 and found that rosemary had the greatest antioxidative efficiency. Some bioactive compounds

80 in plant extracts act as reducing agents to promote the curing reaction, while others exhibit a  
81 pro-oxidant effect in meat products (Lin et al., 2011; Viuda-Martos et al., 2009; Vossen et al.,  
82 2012). Although several studies have investigated the use of plant extracts in traditionally cured  
83 meat products, the effects of green tea extract, rosemary extract, and white kimchi powder (a  
84 natural nitrite/nitrate source) on the quality characteristics of naturally cured meat products have  
85 not been evaluated. A new approach is needed to enhance the function of nitrite in naturally  
86 cured meat products with less residual nitrite content and to utilize these natural ingredients that  
87 can be applied to clean-label meat products. Therefore, the purpose of this study was to evaluate  
88 whether green tea extract, rosemary extract, and white kimchi powder can be used to cure pork  
89 sausages without adding nitrite and ascorbate.

## 92 **Materials and Methods**

### 94 **The preparation of white kimchi powder and materials**

95 Chinese cabbages and radishes grown in South Korea were purchased from five regions  
96 (Chungcheong-do, Gangwon-do, Gyeonggi-do, Gyeongsang-do, and Jeolla-do) and were  
97 randomly selected for white kimchi production. Garlic, ginger, fermented shrimp, solar salt, and  
98 refined salt were purchased at local retail markets (Busan, Korea). Fermented white kimchi was  
99 prepared using the standard recipe (Institute of Traditional Korean Food, 2008), with a slight  
100 modification. After 2 wk of fermentation at 0°C, white kimchi was powdered, as described  
101 previously (Choi et al., 2020). The fermented white kimchi was crushed and stored in a deep  
102 freezer (HKF-51, HFK, Wonju, Korea) at -80°C prior to freeze-vacuum drying using a freeze  
103 vacuum dryer (PVTFD10R, Ilshinbiobase, Yangju, Korea) at -40°C for 48 h. Thereafter, the

104 dried white kimchi was pulverized and passed through a #30 mesh sieve (Test sieve BS0600,  
105 Chunggye Sieve, Gunpo, Korea). The white kimchi powder was analyzed for pH (6.24) and  
106 nitrate content (19,634 ppm sodium nitrate), vacuum-packed, and stored in a freezer at  $-18^{\circ}\text{C}$   
107 until use as a nitrate source.

108 Celery powder (VegStable 502, Florida Food Products Inc., Eustis, FL, USA) with 35,281  
109 ppm sodium nitrate, a starter culture (CS 299, Chr. Hansen Inc., Milwaukee, WI, USA)  
110 consisting of *S. carnosus*, sodium nitrite (S2252, Sigma-Aldrich, St. Louis, MO, USA), sodium  
111 ascorbate (#35268, Acros Organics, Geel, Belgium), green tea extract powder (Green Tea  
112 Extract, Phytotech Extracts Pvt. Ltd., Bengaluru, India), and rosemary extract powder  
113 (#19100003, Plantextrakt GmbH & Co. KG, Vestenbergsgreuth, Germany) were purchased  
114 from commercial suppliers. Other ingredients and spices were obtained from an ingredient  
115 supplier (Taewon Food Industry Co. Ltd., Ansan, Korea).

116

### 117 **Ground pork sausage processing**

118 Fresh pork muscles (*M. biceps femoris*, *M. semitendinosus*, and *M. semimembranosus*) and  
119 backfat were purchased from a local meat processor (Pukyung Pig Farmers Livestock, Gimhae,  
120 Korea) at 24–48 h post-mortem. After trimming, the raw materials were cut into squares of  
121 approximately 4–5 cm and stored in a  $-18^{\circ}\text{C}$  freezer. All processing steps were performed  
122 within 1 month of storage. A batch of 35 kg per replicate was prepared for manufacturing ground  
123 pork sausages, and sausage processing was replicated three times. Before processing, frozen  
124 pork ham and backfat were completely thawed at  $2-3^{\circ}\text{C}$  and ground using a chopper (TC-22  
125 Elegant plus, Tre Spade, Torino, Italy) equipped with an 8-mm plate. The meat was chopped  
126 again using a 3-mm plate. The ground materials were randomly assigned to one of seven  
127 treatments (Table 1): control (0.01% sodium nitrite and 0.05% sodium ascorbate), treatment 1

128 (0.3% white kimchi powder and 0.05% green tea extract powder), treatment 2 (0.3% white  
129 kimchi powder and 0.1% green tea extract powder), treatment 3 (0.3% white kimchi powder  
130 and 0.05% rosemary extract powder), treatment 4 (0.3% white kimchi powder and 0.1%  
131 rosemary extract powder), treatment 5 (0.3% white kimchi powder, 0.05% green tea extract  
132 powder, and 0.05% rosemary extract powder), and treatment 6 (0.3% celery juice powder, 0.05%  
133 green tea extract powder, and 0.05% rosemary extract powder). The nitrate content in white  
134 kimchi powder and celery juice powder was calculated as 60 ppm (treatments 1 to 5) and 105.8  
135 ppm (treatment 6), respectively, for naturally cured sausages, which contained 0.03% starter  
136 culture to convert the nitrate in white kimchi powder and celery juice powder to nitrite.  
137 Phosphates were not added because the objective of this study was to evaluate meat products  
138 with a clean-label concept. The prepared pork meat and fat were mixed with 1.5% NaCl for 3  
139 min using a mixer (5KSM7990, Whirlpool Corp., St. Joseph, MI, USA). Other ingredients and  
140 ice/water were added into the mixer, and the prepared meat product was mixed for seven  
141 additional minutes depending on the treatment (Table 1). Meat mixtures from each treatment  
142 were stuffed into 24-mm cellulose casings (NOJAX Cellulose Casings, Viskase Companies,  
143 Inc., Lombard, IL, USA) using a stuffer (TRE SPADE MOD.5/V Deluxe, FACEM S.p.A,  
144 Torino, Italy). The control samples were placed in a refrigerator (C110AHB, LG Electronics  
145 Inc., Changwon, Korea) at 2–3°C for 1 h, while the naturally cured samples were placed in an  
146 incubator (C-IB4, Changshin Science, Pocheon, Korea) at 38°C for 2 h (Bae et al., 2020;  
147 Sindelar et al., 2007). All samples were cooked to reach an internal temperature of 75°C in a  
148 water bath (MaXturdy 45, Daihan Scientific, Wonju, Korea) set at 90°C. After cooking, the  
149 products were immediately cooled for 20 min in an ice slurry and stored at 2–3°C in the dark  
150 until analysis. All dependent variables were measured in duplicate.

151



152 **Cooking yield and pH measurements**

153 The sausages were weighed before and after cooking to determine the cooking yield. The  
154 cooking yield was calculated using the following equation:

$$\text{Cooking yield (\%)} = \frac{\text{Sample weight after cooking}}{\text{Sample weight before cooking}} \times 100$$

155

156 For pH measurement, 5 g of cooked sample was blended with 45 mL distilled water and  
157 homogenized for 1 min at 8,000 rpm using a homogenizer (DI 25 basic, IKA-Werke GmbH &  
158 Co. KG, Staufen, Germany). The pH of the homogenates was measured using a pH meter  
159 (Accumt AB150, Thermo Fisher Scientific Inc., Singapore).

160

161 **Oxidation-reduction potential (ORP) determination**

162 The ORP of cooked products was measured using a slight modification of the method  
163 described by Cornforth et al. (1986) and John et al. (2005). Briefly, 10 g of each sample was  
164 homogenized with 20 mL of 0.1 M sodium carbonate for 30 s at 13,000 rpm using a  
165 homogenizer (DI 25 basic, IKA-Werke GmbH & Co. KG, Staufen, Germany). Thereafter, 50  
166  $\mu\text{L}$  butylated hydroxytoluene (7.2% in ethanol) was added to minimize sample oxidation during  
167 blending. The samples were allowed to stabilize for 3 min. ORP values were determined using  
168 a platinum Ag/AgCl combination electrode (13-620-631, Thermo Fisher Scientific Inc.,  
169 Singapore) mounted on a pH meter (Accumt AB150, Thermo Fisher Scientific Inc., Singapore)  
170 set to the millivolt scale.

171

172 **Instrumental color measurements**

173 The CIE L\* (lightness), a\* (redness), and b\* (yellowness) values were measured on the cut  
174 surface of each sample using a chroma meter (CR-400, Konica Minolta Sensing Inc., Osaka,

175 Japan; 8 mm aperture, illuminant C, 2° observer angle) calibrated with a white calibration plate  
176 ( $L^* +94.90$ ,  $a^* -0.39$ ,  $b^* +3.88$ ). Each cooked sample was cut parallel to the longitudinal axis  
177 and divided into two slices. Color measurements were taken on the fresh cut surface at two  
178 random locations on each sliced sample. Four readings were obtained per treatment.

179

#### 180 **Nitrate and residual nitrite content analysis**

181 The nitrate ion ( $\text{NO}_3^-$ ) content of the prepared white kimchi powder and commercial celery  
182 powder was analyzed using the zinc reduction method (Merino, 2009). The results were  
183 converted to concentrations of sodium nitrate (ppm). The residual nitrite content of the cooked  
184 meat sausages was analyzed by the AOAC method 973.31 (AOAC, 2016). The calibration curve  
185 was prepared using sodium nitrite as a standard (S2252, Sigma-Aldrich, St. Louis, MO, USA),  
186 and the residual nitrite content was reported in ppm.

187

#### 188 **Nitrosyl hemochrome content, total pigment content, and cure efficiency determination**

189 Nitrosyl hemochrome and total pigment contents were measured using the method described  
190 by Hornsey (1956). Blended samples were mixed with 80% acetone and acidified acetone  
191 solution to measure nitrosyl hemochrome and total pigment contents, respectively. The  
192 absorbance of the filtrate was measured at wavelengths of 540 nm ( $A_{540}$ ) and 640 nm ( $A_{640}$ ),  
193 respectively, using a spectrophotometer (UV-1800, Shimadzu Corp., Kyoto, Japan). Nitrosyl  
194 hemochrome and total pigment concentrations were calculated using the following equations.  
195 Finally, curing efficiency was calculated as the ratio of nitrosyl hemochrome content to total  
196 pigment content in the meat.

197

$$\text{Nitrosyl hemochrome (ppm)} = A_{540} \times 290$$

198

$$\text{Total pigment (ppm)} = A_{640} \times 680$$

199

$$\text{Curing efficiency (\%)} = \frac{\text{Nitrosyl hemochrome}}{\text{Total pigment}} \times 100$$

200

### 201 **Thiobarbituric acid reactive substances (TBARS) determination**

202 Lipid oxidation was analyzed using the distillation method described by Tarladgis et al.  
203 (1960). After reacting malondialdehyde (MDA) released by lipid oxidation with 0.02 M 2-  
204 thiobarbituric acid (TBA) solution, the absorbance was measured at 538 nm using a  
205 spectrophotometer (UV-1800, Shimadzu Corp., Kyoto, Japan). The obtained results were  
206 multiplied by a factor of 7.8 to calculate TBARS values (mg MDA/kg cooked sample).

207

### 208 **Statistical analysis**

209 The experimental design was a completely randomized design with seven treatment groups  
210 (one control and six naturally cured treatments). All experiments used three independent  
211 replicates, with each replicate manufactured on a different processing day. The data were  
212 analyzed using the Generalized Linear Model (GLM) included in the SAS software package  
213 (SAS, 2012). When significant differences were identified, the dependent variable means were  
214 analyzed using Tukey's multiple comparisons tests ( $p < 0.05$ ).

215

216

## 217 **Results and Discussion**

### 218 **Cooking yield, pH, and ORP**

219 The cooking yield was higher in the control than in naturally cured pork sausages (treatments

220 1 to 6) ( $p < 0.05$ ) (Table 2). Recently, Choi et al. (2020) investigated the effects of using acerola  
221 juice powder on the quality of meat products indirectly cured with white kimchi powder and  
222 found that pork products cured with white kimchi powder and acerola juice powder had a lower  
223 cooking yield than nitrite-added control, similar to our results. However, in this study, there  
224 were no differences ( $p > 0.05$ ) in cooking yield for products cured with white kimchi powder or  
225 celery juice powder, regardless of addition level or combinations of green tea extract powder  
226 and rosemary extract powder. Jeong et al. (2020b) found that pork sausages cured with 0.15%  
227 to 0.35% Chinese cabbage powders had a similar cooking yield to those cured with 0.4%  
228 commercial celery juice powders. Considering the quality of meat products and industrial use,  
229 a low cooking yield may have negative impacts on both manufacturers and consumers.  
230 Sebranek and Bacus (2007a) pointed out that moisture retention in natural or organic meat  
231 products may be reduced without the addition of phosphates or water-binding agents. In this  
232 study, alkali phosphate was not intentionally added to the products. Therefore, when producing  
233 meat products using natural ingredients, appropriate ingredients that replace phosphate may  
234 improve moisture retention. There was some evidence for this. Choi et al. (2020) reported that  
235 when phosphate was not added, pork products cured with white kimchi powder had a lower  
236 cooking yield than the nitrite-added control, whereas Bae et al. (2020) found that there were no  
237 differences in cooking yield between the control with sodium nitrite and pork products cured  
238 with radish powder when the samples were prepared with adding sodium tripolyphosphate.

239 The pH of meat products cured with plant-based natural ingredients (treatments 1 to 6) did  
240 not differ ( $p > 0.05$ ) from that of the control sausages containing sodium nitrite (Table 2).  
241 Moreover, no differences ( $p > 0.05$ ) in pH were observed among the naturally cured pork  
242 sausages, suggesting that green tea extract and rosemary extract did not affect the pH of pork  
243 products cured with white kimchi powder or celery juice powder. Similar to our results, Wenjiao

244 et al. (2014) reported that the addition of tea polyphenol had no effect on the pH of pork  
245 sausages. However, Lara et al. (2011) found that the pH of cooked pork patties was significantly  
246 reduced by the addition of rosemary or lemon balm extract due to the acidic compounds present  
247 in the extracts. Jeong et al. (2020a) reported that the addition of Chinese cabbage, radish, or  
248 spinach powder to naturally cured pork products did not change the pH of cured pork products,  
249 which was in line with our results. In a preliminary test for this study, the pH values of raw pork  
250 meat, white kimchi powder, green tea extract powder, and rosemary extract powder were 5.80,  
251 6.24, 4.45, and 5.42, respectively. The equivalent pH in all samples in this study may be  
252 attributed to the relatively high pH of white kimchi powder and the buffering capacity of meat  
253 (Krause et al, 2011; Li et al., 2012).

254 A low ORP promotes the color development of meat products because it contributes to a  
255 reducing environment in meat (Claus and Jeong, 2018; Cornforth et al., 1986). In this study, the  
256 ORP values of meat products ranged from  $-141.57$  to  $-142.85$  (Table 2). Generally, ascorbate  
257 can act as a reducing agent for myoglobin under certain conditions (Holownia et al., 2003).  
258 However, in this study, control sausages containing ascorbate showed no differences ( $p>0.05$ )  
259 in ORP from the products with the natural extracts. In previous research, Wójciak et al. (2011)  
260 showed that the addition of plant extracts (green tea, red pepper, rosemary) to meat products  
261 had significantly reduced ORP values. Therefore, our results suggested that the natural extracts  
262 used in this study may have acted as reducing agents in naturally cured pork sausages. In  
263 addition, differences in pH can alter the ORP (Antonni and Brunori, 1971). Thus, it may appear  
264 that the final products with similar pH values did not result in different ORP values, regardless  
265 of nitrite/nitrate sources and the addition of green tea extract or rosemary extract powder.

266

## 267 **Instrumental color**

268 The addition of green tea extract powder, rosemary extract powder, or a combination of the  
269 two did not significantly change ( $p>0.05$ ) the CIE L\* values of white kimchi powder-treated  
270 products (treatments 1 to 5), compared to control sausages (Table 2). Jin et al. (2016) found that  
271 the use of 1% and 2% thyme or rosemary powder did not change the CIE L\* values of pork  
272 sausages at the beginning of storage. Similarly, Vossen et al. (2012) reported that the CIE L\*  
273 values between frankfurters prepared with 0.5% dog rose extract and control frankfurters  
274 containing sodium nitrite and sodium ascorbate were not different. In this study, pork sausages  
275 prepared with celery juice powder (treatment 6) as a natural nitrate source had lower ( $p<0.05$ )  
276 CIE L\* values compared to control sausages, which is inconsistent with the results of Choi et  
277 al. (2020), who reported that the CIE L\* values of products prepared with 0.4% celery powder  
278 were not different from those of nitrite-supplemented controls. This may be attributed to the  
279 difference between the manufacturers and dosages of the celery product used in these studies.

280 Redness in cured meat products is an important index that is used to determine the degree of  
281 cured meat color and is related to nitrosyl hemochrome content (AMSA, 2012). Some studies  
282 found that green tea and rosemary treatments had no effect on the redness of meat products  
283 (Bozkurt, 2006; Rojas and Brewer, 2007). In this study, the CIE a\* values of all products tested  
284 ranged from 8.40 to 8.71, and there were no differences ( $p>0.05$ ) between the control and the  
285 treatments (Table 2). From the point of view of alternative curing, these results indicate that the  
286 amount of nitrate added in the form of white kimchi powder or celery powder is sufficient to  
287 develop the cured meat color of the final products. It should be noted that treatments 1 to 5 had  
288 a low nitrate content derived from white kimchi powder (approximately 60 ppm sodium nitrate).  
289 Even if nitrate was completely reduced to nitrite, the amounts of nitrite in white kimchi powder  
290 treatments would have been lower than that in the control (100 ppm sodium nitrite), but the

291 cured color was comparable. Regarding the CIE  $a^*$  values of naturally cured products, Terns et  
292 al. (2011) showed similar results using celery juice powder and cherry powder in indirectly  
293 cured sausages. Magrinyà et al. (2016) also found that cooked cured sausages prepared with  
294 vegetable concentrate and tocopherol extract had the same redness as products prepared with  
295 sodium nitrite. Thus, white kimchi powder has the potential to replace synthetic nitrite or  
296 commercial celery powder as a natural nitrate source for the production of naturally cured meat  
297 products. Furthermore, natural ingredients such as green tea extract powder and rosemary  
298 extract powder could be effectively used as sodium ascorbate alternatives. In addition, the role  
299 of natural ingredients as replacers of nitrite/nitrate or ascorbate is supported by the results of  
300 nitrosyl hemochrome content and curing efficiency in this study (Table 3).

301 The pork sausages containing natural vegetable powders (treatments 1 to 6) showed higher  
302 ( $p < 0.05$ ) CIE  $b^*$  values than the control sausages (Table 2), probably because of the inherent  
303 pigments in the plant-derived powders (Bae et al., 2020; Jeong et al., 2020a). When the amount  
304 of green tea extract powder or rosemary extract powder increased from 0.05% to 0.1%  
305 (treatments 2 and 4) or when both were added in combination (treatments 5 and 6), the CIE  $b^*$   
306 value significantly increased ( $p < 0.05$ ). Several previous reports indicate that pigments in plant-  
307 based ingredients that replace nitrite/nitrate may affect the yellowness of the meat products (Bae  
308 et al., 2020; Horsch et al., 2014; Jeong et al., 2020b; Kim et al., 2019; Riel et al., 2017). Horsch  
309 et al. (2014) reported that yellowness in hams increased with increasing celery concentrate  
310 concentration, likely because the plant-derived concentrate includes plant pigments. Jin et al.  
311 (2016) and Nowak et al. (2016) also suggested that increased yellowness of sausages may have  
312 resulted from the color of the plant extracts. These previous findings are consistent with the  
313 results of our study.

314

### 315 **Residual nitrite, nitrosyl hemochrome, and total pigment contents and curing efficiency**

316 The residual nitrite content was the highest ( $p < 0.05$ ) in the control (23.94 ppm), and  
317 approximately 76% of the initial nitrite amount reduced after product manufacturing (Table 3),  
318 similar to the result of a previous study showing that the residual nitrite content of frankfurters  
319 decreased to approximately 75% of initial nitrite levels (Xi et al., 2012). The depletion of  
320 residual nitrite is affected by factors such as meat type, pH, initial nitrite content, cooking  
321 temperature, and reducing agents (Cassens et al, 1978; Flores and Toldrá, 2021; Honikel, 2008;  
322 Sindelar and Milkowski, 2011). In this study, a lower residual nitrite content in all naturally  
323 cured sausages (treatments 1 to 6) was observed when compared to control sausages ( $p < 0.05$ )  
324 (Table 3). Several previous studies using nitrate-rich plant substitutes as natural sources to  
325 replace synthetic nitrite in alternatively cured meat products have shown a similar trend (Bae et  
326 al., 2020; Jeong et al., 2020a; Jeong et al., 2020b; Sebranek and Bacus, 2007a; Yong et al.,  
327 2021). These results could be interpreted in two ways. First, the content of nitrate derived from  
328 white kimchi powder was relatively lower in treatments 1 to 6. Second, nitrite generated by  
329 nitrate-reducing bacteria during product processing prior to being cooked may have reacted  
330 with bioactive compounds present in natural ingredients, thereby resulting in lower residual  
331 nitrite levels in the finished products. There is some evidence that bioactive compounds play a  
332 role in residual nitrite content reduction (Lin et al., 2011; Viuda-Martos et al., 2009; Viuda-  
333 Martos et al., 2010; Zhou et al., 2020). Viuda-Martos et al. (2009) described that decreased  
334 residual nitrite content may have resulted from reactions with bioactive compounds such as  
335 polyphenols and flavonoids that are present in natural ingredients. Zhou et al. (2020) also  
336 reported decreased residual nitrite content when rosemary extract, grape seed extract, and green  
337 tea polyphenols were added to pork sausages. Nevertheless, among the products cured with  
338 white kimchi powder (treatments 1 to 5), the residual nitrite content was not affected ( $p > 0.05$ )



339 by the addition of green tea extract powder and rosemary extract powder, either alone or in  
340 combination. Treatment 6, which contained celery juice powder as a natural source of nitrate,  
341 had higher ( $p<0.05$ ) residual nitrite content than the kimchi powder counterpart (treatment 5),  
342 likely because the nitrate levels in the commercially available celery juice powder were  
343 relatively higher than those in the white kimchi powder prepared in this study, although the  
344 same concentration (0.3%) for both celery juice powder and white kimchi powder was included  
345 in the formulation.

346 In the manufacture of meat products, nitrosyl hemochrome is formed during the curing and  
347 cooking processes. It contributes to the distinctive pink color of meat products. In alternative  
348 curing methods, it has been proposed that *S. carnosus*, a starter culture with nitrate reductase  
349 activity, and high natural nitrate levels in vegetable powder, increase the rate of nitrate reduction  
350 to nitrite in the meat curing system (Alahakoon et al., 2015; Magrinyà et al., 2016; Sindelar and  
351 Houser, 2009). In particular, nitric oxide released from nitrite reacts with myoglobin to form  
352 nitrosomyoglobin. These curing reactions can be promoted by using cure accelerators such as  
353 sodium ascorbate or sodium erythorbate (Honikel, 2008; Sebranek, 2009; Sebranek et al., 2012).  
354 Although reducing agents such as sodium ascorbate were not used in the naturally cured  
355 products, the nitrosyl hemochrome content was similar ( $p>0.05$ ) in treatments 1 to 5 or higher  
356 ( $p<0.05$ ) in treatment 6 compared to the control sausages (Table 3). In addition, the nitrosyl  
357 hemochrome content was not affected ( $p>0.05$ ) by the addition of green tea extract powder,  
358 rosemary extract powder, or their combination in naturally cured sausages. In agreement with  
359 our results, Terns et al. (2011) reported that indirectly cured sausages formulated with vegetable  
360 juice powder and cherry powder without reductants showed effective cured pigment  
361 development similar to that in control sausages with sodium ascorbate and sodium erythorbate.  
362 Choi et al. (2020) obtained similar results for cooked pork products that were cured with white

363 kimchi powder and acerola juice powder. Wójciak et al. (2011) found that the use of green tea  
364 and rosemary helped to reduce nitrite to nitric oxide and form nitrosomyoglobin, thereby  
365 stabilizing the color of the cured meat products. Likewise, our results indicate that the green tea  
366 extract powder and rosemary extract powder may be involved in the curing reactions, acting as  
367 reducing agents that promote the release of nitric oxide from nitrite, thereby forming nitrosyl  
368 hemochrome in cooked sausages. This result was expected based on the results of residual  
369 nitrite content in this study.

370 In the cooked cured products tested in this study, there was no difference ( $p>0.05$ ) in total  
371 pigment content, regardless of the nitrate/nitrite source and addition of natural extract powders  
372 (Table 3). Similar results were reported when using 0.15% to 0.30% radish powder with 0.05%  
373 sodium ascorbate in alternatively cured pork products (Bae et al., 2020). However, the total  
374 pigment content found in naturally cured sausages (from 51.68 ppm to 54.29 ppm) in this study  
375 was slightly higher than that obtained in pork sausages cured with Chinese cabbage powder  
376 (from 48.62 ppm to 49.13 ppm) (Jeong et al., 2020b) and in pork products cured with radish  
377 powder (from 45.73 ppm to 45.90 ppm) (Bae et al., 2020). In this sense, the addition of green  
378 tea extract powder and rosemary extract powder may have played a role in the formation of  
379 meat pigments in the final products.

380 Treatments using white kimchi powder (treatments 1 to 5) showed a curing efficiency similar  
381 ( $p>0.05$ ) to that of control sausages (Table 3). However, treatment 6 had a higher ( $p<0.05$ )  
382 curing efficiency than that of the control group. The curing efficiency is the percentage of total  
383 pigment converted to nitroso pigment in cured meat (AMSA, 2012). Therefore, the high curing  
384 efficiency in treatment 6 may be the result of the high nitrosyl hemochrome content observed.  
385 Nevertheless, when the amount of green tea extract powder or rosemary extract powder was  
386 increased to 0.1% (treatments 2 and 4) or when the two extract powders are added together at

387 0.5% (treatment 5), the curing efficiency was not significantly different ( $p>0.05$ ) from that of  
388 treatment 6. These results suggest that the use of green tea extract powder or rosemary extract  
389 powder alone or in combination provides equivalent or higher curing efficiency compared to  
390 traditionally cured products.

391

### 392 **Thiobarbituric acid reactive substances (TBARS)**

393 Across all products tested in this study, the TBARS values were considerably lower ( $<0.15$   
394 mg MDA/kg) than the threshold range (0.5–1.0 mg MDA/kg) for rancidity perception  
395 (Tarladgis et al., 1960). However, regardless of addition of green tea extract powder and  
396 rosemary extract powder, naturally cured products prepared with white kimchi powder or celery  
397 powder (treatments 1 to 6) were found to have higher ( $p<0.05$ ) TBARS values than control  
398 sausages (Table 3). In alternative curing, vegetables that have characteristic flavors and colors  
399 are used as nitrite/nitrate alternatives (Jo et al., 2020). Because the excessive use of vegetable  
400 powders during curing may negatively affect sensory properties, the amount of vegetable  
401 powders used in alternatively cured meat products is limited, thereby resulting in a low residual  
402 nitrite content in the final products (Sebranek and Bacus, 2007a). Thus, the lower TBARS  
403 values in the control samples may result from their high residual nitrite content, which had an  
404 antioxidative effect on lipid oxidation. Previously, several studies on natural antioxidants  
405 showed that the application of green tea extract and rosemary extract inhibits lipid oxidation in  
406 meat and meat products as a function of associated phenolic compounds (Fernández-López et  
407 al., 2005; Hernández-Hernández et al., 2009; Lorenzo and Munekata, 2016; Nieto et al., 2018).  
408 In this regard, the TBARS values were expected to be lower in naturally cured sausages.  
409 However, the natural extract powders in this study did not completely compensate for the  
410 antioxidant activity in naturally cured products with low nitrite levels. A study by Lin et al.

411 (2011) confirmed that a small amount (0.009%) of sodium nitrite is more effective in preventing  
412 lipid oxidation than green tea extract at a low concentration (0.05%). Nevertheless, when the  
413 amount of green tea extract powder or rosemary extract powder was increased to 0.1%  
414 (treatments 2 and 4) or green tea extract powder, rosemary extract powder, and celery juice  
415 powder were used in combination (treatment 6), the TBARS values decreased ( $p < 0.05$ ) in the  
416 naturally cured products, suggesting that 0.1% green tea extract powder and rosemary extract  
417 powder have antioxidant effects that inhibit lipid oxidation in the sausages. Therefore, to  
418 enhance the antioxidant activity in naturally cured meat products, a substantial amount of these  
419 extracts should be added, higher than the concentration used in this study.

420

421

422

## Conclusions

423 Green tea extract powder and rosemary extract powder, alone or in combination, are potential  
424 replacements for sodium ascorbate in the production of sausages naturally cured with white  
425 kimchi powder. These naturally cured products exhibited redness, nitrosyl hemochrome content,  
426 total pigment content, and curing efficiency comparable to those of products containing nitrite.  
427 However, 0.1% green tea extract powder and rosemary extract powder inhibited lipid oxidation  
428 in naturally cured products but showed higher TBARS values than control products. These  
429 results suggest that the use of green tea extract powder and rosemary extract powder (either  
430 alone or in combination) in sausages cured with white kimchi powder may provide an effective  
431 solution to replace synthetic nitrite and ascorbate used in traditionally cured products. To  
432 supplement the antioxidant activity from these two extract powders in naturally cured meat  
433 products with low nitrite content, addition of a sufficient amount (higher than 0.1%) should be  
434 considered.

435

## Conflicts of interest

436 The authors declare no potential conflicts of interest.

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438

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440

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ACCEPTED

604 **Table 1. Formulation for pork sausages naturally cured with white kimchi powder or celery powder**

Ingredients (%)	Treatments <sup>1</sup>						
	Control	Treatment 1	Treatment 2	Treatment 3	Treatment 4	Treatment 5	Treatment 6
Pork ham	70.00	70.00	70.00	70.00	70.00	70.00	70.00
Pork back fat	15.00	15.00	15.00	15.00	15.00	15.00	15.00
Ice/water	15.00	15.00	15.00	15.00	15.00	15.00	15.00
Sub total	100.00	100.00	100.00	100.00	100.00	100.00	100.00
NaCl	1.50	1.50	1.50	1.50	1.50	1.50	1.50
Sugar	1.00	1.00	1.00	1.00	1.00	1.00	1.00
Mustard powder	0.30	0.30	0.30	0.30	0.30	0.30	0.30
Pepper	0.25	0.25	0.25	0.25	0.25	0.25	0.25
Onion powder	0.10	0.10	0.10	0.10	0.10	0.10	0.10
Garlic powder	0.10	0.10	0.10	0.10	0.10	0.10	0.10
Sodium nitrite	0.01	-	-	-	-	-	-
White kimchi powder	-	0.30	0.30	0.30	0.30	0.30	-
Celery juice powder	-	-	-	-	-	-	0.30
Starter culture	-	0.03	0.03	0.03	0.03	0.03	0.03
Sodium ascorbate	0.05	-	-	-	-	-	-
Green tea extract powder	-	0.05	0.10	-	-	0.05	0.05
Rosemary extract powder	-	-	-	0.05	0.10	0.05	0.05
Total	103.31	103.63	103.68	103.63	103.68	103.68	103.68

605 <sup>1</sup> Treatments: control, 0.01% sodium nitrite + 0.05% sodium ascorbate; treatment 1, 0.3% white kimchi powder + 0.05% green tea extract  
606 powder; treatment 2, 0.3% white kimchi powder + 0.1% green tea extract powder; treatment 3, 0.3% white kimchi powder + 0.05% rosemary  
607 extract powder; treatment 4, 0.3% white kimchi powder + 0.1% rosemary extract powder; treatment 5, 0.3% white kimchi powder + 0.05% green  
608 tea extract powder + 0.05% rosemary extract powder; and treatment 6, 0.3% celery juice powder + 0.05% green tea extract powder + 0.05%  
609 rosemary extract powder.

610

611 **Table 2. Effects of green tea extract and rosemary extract on cooking yield, pH values, ORP, and CIE color values of pork sausages**  
 612 **naturally cured with white kimchi powder or celery powder**

Treatments <sup>1</sup>	Cooking yield (%)	pH	ORP <sup>2</sup> (mV)	CIE L*	CIE a*	CIE b*
Control	94.13±0.31 <sup>A</sup>	6.07±0.04 <sup>A</sup>	-142.12±3.60 <sup>A</sup>	65.77±0.12 <sup>A</sup>	8.66±0.08 <sup>A</sup>	8.91±0.06 <sup>D</sup>
Treatment 1	90.60±0.47 <sup>B</sup>	6.00±0.04 <sup>A</sup>	-142.37±1.33 <sup>A</sup>	65.73±0.14 <sup>A</sup>	8.59±0.12 <sup>A</sup>	9.84±0.07 <sup>C</sup>
Treatment 2	90.98±0.56 <sup>B</sup>	5.97±0.05 <sup>A</sup>	-141.72±0.74 <sup>A</sup>	65.66±0.18 <sup>AB</sup>	8.71±0.13 <sup>A</sup>	10.31±0.08 <sup>B</sup>
Treatment 3	91.55±0.53 <sup>B</sup>	5.99±0.04 <sup>A</sup>	-142.85±0.43 <sup>A</sup>	65.45±0.13 <sup>AB</sup>	8.40±0.13 <sup>A</sup>	9.78±0.10 <sup>C</sup>
Treatment 4	91.41±0.43 <sup>B</sup>	5.99±0.04 <sup>A</sup>	-141.57±1.13 <sup>A</sup>	65.32±0.12 <sup>AB</sup>	8.43±0.13 <sup>A</sup>	10.32±0.06 <sup>B</sup>
Treatment 5	90.18±0.53 <sup>B</sup>	5.98±0.05 <sup>A</sup>	-142.82±1.36 <sup>A</sup>	65.45±0.10 <sup>AB</sup>	8.52±0.09 <sup>A</sup>	10.18±0.06 <sup>B</sup>
Treatment 6	90.46±0.48 <sup>B</sup>	6.01±0.02 <sup>A</sup>	-142.23±0.88 <sup>A</sup>	65.10±0.10 <sup>B</sup>	8.53±0.11 <sup>A</sup>	10.75±0.06 <sup>A</sup>

613 All values are means ± SE.

614 <sup>A-D</sup> Means within a column with different superscript letters are significantly different (p<0.05).

615 <sup>1</sup> Treatments: control, 0.01% sodium nitrite + 0.05% sodium ascorbate; treatment 1, 0.3% white kimchi powder + 0.05% green tea extract  
 616 powder; treatment 2, 0.3% white kimchi powder + 0.1% green tea extract powder; treatment 3, 0.3% white kimchi powder + 0.05% rosemary  
 617 extract powder; treatment 4, 0.3% white kimchi powder + 0.1% rosemary extract powder; treatment 5, 0.3% white kimchi powder + 0.05% green  
 618 tea extract powder + 0.05% rosemary extract powder; and treatment 6, 0.3% celery juice powder + 0.05% green tea extract powder + 0.05%  
 619 rosemary extract powder.

620 <sup>2</sup> ORP, Oxidation-reduction potential.

621

622 **Table 3. Effects of green tea extract and rosemary extract on residual nitrite, nitrosyl hemochrome, total pigment, curing efficiency, and**  
 623 **TBARS values of pork sausages naturally cured with white kimchi powder or celery powder**

Treatments <sup>1</sup>	Residual nitrite (ppm)	Nitrosyl hemochrome (ppm)	Total pigment (ppm)	Curing efficiency (%)	TBARS <sup>2</sup> (mg MDA/kg)
Control	23.94±0.33 <sup>A</sup>	38.69±0.80 <sup>B</sup>	50.66±1.00 <sup>A</sup>	76.38±0.68 <sup>B</sup>	0.094±0.002 <sup>C</sup>
Treatment 1	6.82±0.13 <sup>BC</sup>	40.41±0.98 <sup>AB</sup>	52.25±0.97 <sup>A</sup>	77.25±0.50 <sup>B</sup>	0.142±0.007 <sup>A</sup>
Treatment 2	6.43±0.17 <sup>C</sup>	42.39±0.92 <sup>AB</sup>	54.29±1.10 <sup>A</sup>	78.06±0.19 <sup>AB</sup>	0.129±0.005 <sup>B</sup>
Treatment 3	7.13±0.41 <sup>BC</sup>	40.26±0.84 <sup>AB</sup>	51.68±0.94 <sup>A</sup>	77.87±0.40 <sup>B</sup>	0.140±0.007 <sup>A</sup>
Treatment 4	6.96±0.24 <sup>BC</sup>	41.04±0.84 <sup>AB</sup>	52.59±1.23 <sup>A</sup>	78.10±0.31 <sup>AB</sup>	0.130±0.004 <sup>B</sup>
Treatment 5	6.35±0.10 <sup>C</sup>	41.83±0.86 <sup>AB</sup>	53.38±1.02 <sup>A</sup>	78.34±0.22 <sup>AB</sup>	0.138±0.006 <sup>A</sup>
Treatment 6	7.89±0.48 <sup>B</sup>	43.11±1.18 <sup>A</sup>	53.83±1.03 <sup>A</sup>	79.96±0.78 <sup>A</sup>	0.126±0.005 <sup>B</sup>

624 All values are means ± SE.

625 <sup>A-C</sup> Means within a column with different superscript letters are significantly different (p<0.05).

626 <sup>1</sup> Treatments: control, 0.01% sodium nitrite + 0.05% sodium ascorbate; treatment 1, 0.3% white kimchi powder + 0.05% green tea extract  
 627 powder; treatment 2, 0.3% white kimchi powder + 0.1% green tea extract powder; treatment 3, 0.3% white kimchi powder + 0.05% rosemary  
 628 extract powder; treatment 4, 0.3% white kimchi powder + 0.1% rosemary extract powder; treatment 5, 0.3% white kimchi powder + 0.05% green  
 629 tea extract powder + 0.05% rosemary extract powder; and treatment 6, 0.3% celery juice powder + 0.05% green tea extract powder + 0.05%  
 630 rosemary extract powder.

631 <sup>2</sup> TBARS, 2-Thiobarbituric acid reactive substances; MDA, Malondialdehyde.