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9 **Influence of Naturally Converted Nitrite and Ultrasound Marination on the Curing**  
10 **Efficiency and Residual Nitrite of Pork Loin**

11  
12 **Abstract**

13 This study used ascorbic acid and ultrasound to improve the quality of pork loin marinated  
14 with pre-converted *Angelica keiskei* (PA). Pork loins were cured without nitrite (NC); with  
15 nitrite (PC); with PA (PA); with PA and ascorbic acid (PA+A); with PA and ultrasound  
16 treatment (PA+U); and with PA, ascorbic acid, and ultrasound treatments (PA+AU). The  
17 nitrite content in the curing solution was maintained at 180 ppm, except for NC. Cured loins  
18 with PA and ascorbic acid (PA+A and PA+AU) showed identical color values, curing  
19 efficiencies, and lower residual nitrite compared with that of PC. The pH of the PA-cured  
20 loins was lower than that of PC, and the moisture content was higher than that of NC.  
21 Following ultrasound treatment, protein solubility and tenderness were enhanced, whereas the  
22 myofibril fragmentation index did not significantly differ. PA effectively inhibited lipid  
23 oxidation in cured loins. In the PA+A treatment group, there was no difference in the redness  
24 of the cured meat color compared to PC, but the residual nitrite ion scavenging ability was  
25 close to 70%. Therefore, curing pork loins with pre-converted *Angelica keiskei*, ascorbic acid,  
26 and ultrasound treatment may be useful for producing high-quality meat products.

27 **Keywords:** cured pork loin, natural converted nitrite, ultrasound, curing efficiency, residual  
28 nitrite

## 30 Introduction

31 Curing meat products is a time-honored practice dating back centuries, aimed at enhancing  
32 flavor, color, texture, and shelf-life (Jia et al., 2023). Central to this process is the use of  
33 nitrite salts, which play a pivotal role in ensuring food safety by inhibiting bacterial growth  
34 (Jo et al., 2020). However, concerns have been raised regarding the potential health risks  
35 associated with excessive nitrite consumption, including the formation of carcinogenic  
36 compounds, such as nitrosamines (Yong et al., 2021). Consequently, there has been a growing  
37 interest in exploring alternative curing methods that maintain the safety and quality of cured  
38 meats while reducing reliance on synthetic nitrite additives (Shakil et al., 2022; Zhang et al.,  
39 2023b).

40 One such avenue involves utilizing natural sources of nitrite, including those derived from  
41 the microbial conversion of nitrate present in certain food ingredients (Kim et al., 2019a;  
42 Yong et al., 2021). These naturally occurring nitrites offer promising alternatives to their  
43 synthetic counterparts, potentially mitigating concerns regarding the health effects of  
44 nitrosamines while maintaining the desirable attributes of cured meat (Choi et al., 2017; Kim  
45 et al., 2019b). By systematically analyzing the impact of naturally converted nitrite and  
46 ultrasound marination on both the curing process and residual nitrite levels, we aim to provide  
47 valuable insights into developing safer and more sustainable methods for producing cured  
48 meat products.

49 Additionally, advancements in food processing technologies, such as ultrasound, have led  
50 to the exploration of novel techniques to enhance the efficiency of curing processes (Kang et  
51 al., 2016). Ultrasound, with its ability to facilitate mass transfer and enhance the penetration  
52 of marinades into meat matrices, is an intriguing approach to improve the uniformity and  
53 effectiveness of curing treatments (Inguglia et al., 2018).

54        Thus, this study aims to contribute to the ongoing discourse surrounding the optimization  
55 of curing processes in the meat industry. By leveraging natural sources of nitrite and  
56 innovative marination techniques, we aspire to offer pragmatic solutions that prioritize food  
57 safety and consumer health without compromising the quality of cured pork loin.  
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## 59 Materials and Methods

### 60 2.1. Preparation of naturally converted nitrite

61 The natural source of nitrate, *Angelica keiskei*, was purchased from Garurang (Korea) in  
62 powdered form. *Angelica keiskei* (50 g) was mixed with distilled water (500 mL), extracted  
63 for 2 h at 100°C, and cooled to 25°C. Subsequently, *Staphylococcus carnosus* (Bactoform S-  
64 B-61, Chr. Hansen, Gainesville, FL, USA) was inoculated into the extract and incubated in a  
65 shaking incubator (JSSI-100C, JSR, Gongju, Korea) for 24 h at 37°C and 180 rpm to convert  
66 nitrate into nitrite. The fermented *Angelica keiskei* extract was centrifuged for 10 min at 3,000  
67 × g using a centrifuge (Supra R22, Hanil, Gimpo, Korea), and the supernatant was filtered  
68 through a Whatman No. 1 filter paper. The nitrite content of the fermented *Angelica keiskei*  
69 extract was diluted with a small amount of water to achieve a concentration of 180 ppm.

### 71 2.2. Curing process of pork loin

72 Fresh pork loins (*Longissimus thoracis et lumborum*) were purchased from a local butcher  
73 shop (Wanju, Korea) at 48 h postmortem. Six loins from pigs (Yorkshire × Landrace ×  
74 Durok) were cut to a thickness of 3 cm, and 36 loin chops were randomly selected for use in  
75 the experiments for the six treatments. Considering the residual nitrite content identified in a  
76 previous research, a concentration of nitrite added was selected as 180 ppm of curing solution  
77 (Honikel, 2008). The six treatments were as follows: NC, pork loin cured with 8% sodium  
78 chloride in distilled water; PC, pork loin cured with 8% sodium chloride + 180 ppm sodium  
79 nitrite in distilled water; PA, pork loin cured with 8% sodium chloride in a pre-converted  
80 *Angelica keiskei* extract (180 ppm of nitrite); PA+A, pork loin cured with 8% sodium chloride  
81 + 0.125% ascorbic acid in a pre-converted *Angelica keiskei* extract (180 ppm of nitrite);  
82 PA+U, pork loin cured with 8% sodium chloride in a pre-converted *Angelica keiskei* extract  
83 (180 ppm of nitrite) and subjected to ultrasound treatment after curing; PA+AU, pork loin

84 cured with 8% sodium chloride + 0.125% ascorbic acid in a pre-converted *Angelica keiskei*  
85 extract (180 ppm of nitrite) and subjected to ultrasound treatment after curing. Six loin chops  
86 from each treatment group were brined by immersion in 40% brine by weight and the  
87 treatments were cured for 4 days at 4°C. Ultrasound treatment was conducted on PA+U and  
88 PA+AU in an ultrasonic bath (CPX5800H-E, Emerson, St Louis, MO, USA) for 30 min at a  
89 temperature below 15°C, according to the previous research with slight modification (Leães et  
90 al., 2023). Then, all the treatments were cooked at 80°C for 1 h and cooled for 2 h at 20°C to  
91 conduct the following analysis.

92

### 93 2.3. Color measurements

94 The cooked pork loin chops were cut to a thickness of 2 cm, and the color of the cross-  
95 section was measured immediately using a colorimeter (CR-400; Minolta, Tokyo, Japan). The  
96 aperture size was 8 mm, the illuminant was D65, and the angle of observation was 2°. Color  
97 values are presented as CIE  $L^*a^*b^*$ , and the number of measurements was seven.

98

### 99 2.4. Curing efficiency

100 NO-heme (curing pigment) and total haem pigments (total pigment) in the samples were  
101 determined using the method described by Hornsey (1956). To measure the curing pigment  
102 concentration, 10 g of ground sample was mixed with 80% (v/v) acetone for 5 min under  
103 reduced light conditions. The mixture was then filtered through Whatman No. 1 filter paper  
104 (Whatman International, Maidstone, UK), and the absorbance of the filtrate was measured at  
105 540 nm using a spectrophotometer (Optizen 2120 UV plus, Mecasys Co. Ltd., Daejeon,  
106 Korea). NO-heme concentration was calculated by multiplying the absorbance by 290. For the  
107 total pigment concentration, a 10 g minced sample was mixed with acidified 80% acetone and  
108 left to react for 1 h. The resulting solution was filtered through a Whatman No. 1 filter paper,

109 and the absorbance of the filtrate was measured at 640 nm using a spectrophotometer. The  
110 total pigment concentration was determined by multiplying the absorbance at 680 nm. The  
111 curing efficiency was calculated as the percentage of total pigment converted to a curing  
112 pigment, serving as an index of the degree of cured color stability.

113

#### 114 *2.5. Residual nitrite analysis*

115 To analyze pork loins treated with naturally converted nitrite, ascorbic acid, and ultrasound  
116 marination, a sample weighing 10 g was homogenized in 150 mL of heated distilled water. The  
117 mixture was then combined with 10 mL of 0.5 N NaOH and 10 mL of 12% ammonium  
118 thiosulfate and heated at 80°C for 20 min. After heating, 20 mL of ammonium acetate buffer  
119 was added to the cooled mixture, and the volume was adjusted to 200 mL with distilled water.  
120 The solution was then incubated at 20°C for 10 min. Following incubation, the mixture was  
121 filtered through Whatman No. 1 filter paper. To 20 mL of the filtered sample, 1 mL of  
122 sulfanilamide solution, 1 mL of N-(1-naphthyl)ethylenediamine dihydrochloride reagent, and  
123 3 mL of distilled water were added. This final mixture was incubated at 20°C for 20 min, and  
124 the absorbance was measured at 540 nm, following the method described by KFDA (2016).

125

#### 126 *2.6. pH measurements*

127 The pH of raw pork loins treated with naturally converted nitrite, ascorbic acid, or ultrasound  
128 marination was determined using a pH meter (AB15; Thermo Fisher Scientific, Waltham, MA,  
129 USA). Five grams of each sample was homogenized with 20 mL of distilled water, and the pH  
130 value of each homogenate was measured using pH/ATC electrodes after calibration with buffers  
131 of pH 4, 7, and 10.

132



133 *2.7. Curing and cooking yield*

134 The curing yield of pork loins treated with naturally converted nitrite, ascorbic acid, or  
135 ultrasound marination was determined by measuring the weight differences before and after the  
136 curing process. Similarly, the cooking yield was calculated by assessing the difference in weight  
137 before and after cooking.

138

139 *2.8. Water holding capacity (WHC)*

140 The WHC of raw pork loins subjected to naturally converted nitrite, ascorbic acid, and  
141 ultrasound marination was determined using the press method described by Trout (1988). The  
142 water-holding force was assessed by positioning the sample between two Plexiglas plates and  
143 applying a pressure of 40 kg for 3 min. The meat area and released moisture were quantified  
144 using a planimeter (Planix 7; Tamaya Technics Inc., Japan). The WHC was calculated using  
145 the following formula:

146 
$$\text{WHC (\%)} = (\text{sample meat area} / \text{total moisture area}) \times 100$$

147

148 *2.9. Moisture content*

149 As per AOAC (2000), the moisture content (method 950.46B) of the cooked samples was  
150 determined by measuring the weight loss after drying the samples at 105°C for 12 h in a  
151 drying oven (SW-90D, Sang Woo Scientific Co., Bucheon, Korea).

152

153 *2.10. Differential scanning calorimetry*

154 The thermal properties of raw pork loins were analyzed using a differential scanning  
155 calorimeter (DSC-4000; PerkinElmer, Waltham, MA, USA). Approximately 30 mg of the  
156 pork sample was placed in a sealed aluminum pan, with an empty aluminum pan serving as a

157 reference. The heat flow differential between the sample and reference pans was measured.  
158 The analysis was conducted over a temperature range of 20 to 100°C, with the heating rate set  
159 to 10°C per minute.

160

### 161 *2.11. Myofibril fraction index (MFI)*

162 The MFI of pork loin samples treated with naturally converted nitrite, ascorbic acid, and  
163 ultrasound marination was measured before heating, following the method described by  
164 Culler et al. (1978). Briefly, a 4 g sample was homogenized with 40 mL of MFI buffer  
165 (comprising 100 mM potassium chloride, 20 mM potassium phosphate, 1 mM  
166 ethylenediaminetetraacetic acid, 1 mM magnesium chloride, and 1 mM sodium azide), and  
167 then centrifuged at 1,000 × g for 15 min at 4°C. The resulting pellet was resuspended in 40  
168 mL of MFI buffer and centrifuged again under the same conditions. After dissolving the pellet  
169 in 10 mL of MFI buffer, the solution was filtered through a 10-mesh steel strainer. The  
170 protein concentration was adjusted to 0.5 mg/mL, and its absorbance was measured at 540  
171 nm. To amplify the absorbance unit, a factor of 200 was applied, where a higher value  
172 indicated an increased proteolytic level of the myofibril structure.

173

### 174 *2.12. Protein solubility*

175 The protein solubility of raw pork loin samples treated with naturally converted nitrite,  
176 ascorbic acid, and ultrasound marination was assessed using BCA protein assay kits (Thermo  
177 Fisher Scientific). Buffer A was used to extract total proteins (myofibril and sarcoplasmic),  
178 whereas Buffer B was used to extract sarcoplasmic proteins, following the method of Joo et  
179 al. (1999). The difference in protein solubility between Buffer A and Buffer B was considered  
180 the myofibril solubility. Buffer A consisted of 1.1 M potassium iodide in 0.1 M potassium  
181 phosphate at pH 7.4, and Buffer B contained 0.025 M potassium phosphate at pH 7.4.

182

### 183 *2.13. Shear force*

184 Shear force measurements were performed on pork loin samples treated with naturally  
185 converted nitrite, ascorbic acid, or ultrasonic marination. Cylindrical samples with diameters  
186 of 10 mm were extracted using a hole puncher oriented vertically relative to the muscle fibers.  
187 A texture analyzer (TA-XT Plus; Stable Micro Systems Ltd., Surrey, UK) equipped with a  
188 Warner-Bratzler blade was used for measurements at a test speed of 2 mm/s.

189

### 190 *2.14. Thiobarbituric acid reactive substances (TBARS)*

191 To estimate lipid oxidation in cooked pork loins, a TBARS test was performed following  
192 the method described by Tarladgis et al. (1960). A 10 g sample was homogenized with 100  
193 mL of 0.1 M HCl. Then, the homogenate was heated to 100°C, and the distillate was  
194 subsequently collected. The distillate was reacted with 0.02 M TBA reagent at 100°C for 35  
195 min. After cooling the solution in chilled water, the absorbance was measured at 584 nm to  
196 determine the malondialdehyde content using a standard curve provided by Sigma-Aldrich  
197 (St. Louis, MO, USA).

198

### 199 *2.15. Statistical analysis*

200 Data analyses were performed using SPSS Statistics version 20 (IBM, Armonk, NY, USA).  
201 Three replicates were performed for each experiment, with at least three technical replicates.  
202 To identify significant differences among treatments, a one-way analysis of variance was  
203 employed, followed by Duncan's multiple range test to determine data with significant  
204 differences ( $P < 0.05$ ).

205

## 206 Results and discussion

### 207 3.1. Differences in color values and curing efficiency of pork loin

208 Figure 1 illustrates the effects of naturally converted nitrite, ascorbic acid, and ultrasound  
209 on the color of marinated pork loin. The lightness and yellowness values were highest for the  
210 NC treatment, and treatments with naturally converted nitrite, ascorbic acid, and ultrasound  
211 resulted in lower lightness and yellowness values than those of NC. The redness values for  
212 NC-treated pork loin were the lowest. Among the treatments, there was no significant  
213 difference between PC with nitrite and treatment with pre-converted *Angelica keiskei*, except  
214 for treatments with naturally converted nitrite and ultrasound (PA+U). Choi et al. (2017)  
215 reported that redness and yellowness of cooked meat emulsions increased with increasing  
216 fermented red beef extract, and redness increased with the addition of ascorbic acid due to its  
217 acceleration effect on NO-myoglobin. Hwang et al. (2018) found that, among pre-converted  
218 natural products, celery and spinach extracts increased the lightness, redness, and yellowness  
219 of raw meat batter. Kim et al. (2017) observed that the lightness and yellowness values of raw  
220 meat cured with fermented spinach extract were higher than those of a positive control with  
221 0.015% nitrite. The comparable color values of meat cured with naturally converted nitrite  
222 and sodium nitrite can positively influence color acceptance of consumer (Serdaroğlu et al.,  
223 2023). As inferred from the color value results, there does not seem to be a significant  
224 difference between nitrite and naturally converted nitrite and ascorbic acid, and ultrasound  
225 treatment does not appear to have a significant effect on chromaticity.

226 The effects of naturally converted nitrite, ascorbic acid, and ultrasound on the curing  
227 efficiency of marinated pork loin were significant ( $P < 0.05$ ; Figure 2). The curing efficiency  
228 of marinated pork loin was the highest in the PC treatment group and the naturally converted  
229 nitrite and ascorbic acid treatment groups (PA+A, PA+AU). It was found that ultrasound was  
230 not effective in improving the curing efficiency of marinated pork loins. Choi et al. (2020)

231 reported that the curing efficiency of meat products cured with white kimchi powder and  
232 starter culture was higher than that of the control cured with 0.01% nitrite and 0.05% ascorbic  
233 acid. Bae et al. (2020) reported that the curing efficiency of meat products cured with radish  
234 powder and starter culture, such as *Staphylococcus carnosus*, was significantly increased.  
235 Jantapirak et al. (2023) reported that the curing efficiency of chicken sausages using normal  
236 sodium nitrite levels in the formulation was much higher than that of the low-sodium nitrite  
237 treatment. The results of this study suggest that a similar salting effect can be expected when  
238 nitrite is used in combination with naturally converted nitrite and ascorbic acid.

239

### 240 3.2. Differences in residual nitrite of pork loin

241 The effects of naturally converted nitrite, ascorbic acid, and ultrasound on the residual  
242 nitrite in marinated pork loin are shown in Figure 3. In the NC group, no nitrite was detected  
243 in the residual nitrite of the marinated pork loin. The residual nitrite content of the PC with  
244 nitrite was lower than that of the other treatments with pre-converted *Angelica keiskei* and  
245 ascorbic acid ( $P < 0.05$ ). The residual nitrite in the marinated pork loin was the highest for  
246 converted nitrite and ultrasound (PA + U). The results of this study indicated that ultrasound  
247 treatment did not have a greater effect on residual nitrite than expected. According to Choi et  
248 al. (2017), the residual nitrite content decreases when ascorbic acid is added to cured pork  
249 loin containing fermented red beet extract. Kim et al. (2019) reported that pork loin cured  
250 with organic acids had a lower residual nitrite content than that cured without organic acids,  
251 and malic acid and citric acid showed the highest residual nitrite scavenging ability. King et  
252 al. (2016) reported that, in cured sausage pork, the higher the level of ascorbic acid added, the  
253 higher the nitrite depletion rate. Therefore, it is considered that adding ascorbic acid alone is  
254 effective in reducing the residual nitrous acid content even without ultrasound treatment.

255

256 *3.3. Differences in pH, curing yield, cooking yield, WHC, and moisture content of pork loin*

257 The pH values of naturally converted nitrite, ascorbic acid, and ultrasound-marinated pork  
258 loins are shown in Table 1. The pH of nitrite in sample (PC) was the highest, and the pH of  
259 NC and PC were higher than those of the other treatments with pre-conversion and ultrasound  
260 ( $P < 0.05$ ). These results are similar to those of Choi et al. (2017), who showed that the pH  
261 after treatment with fermented red beet extract was lower than that of the control without  
262 nitrite and with nitrite. Hwang et al. (2018) reported that the pH of pork sausages treated with  
263 nitrite was higher than that of sausages treated with natural pre-converted nitrite sources.  
264 These results, along with the pH of pork loin, appear to be influenced by natural nitrite  
265 fermentation products.

266 Table 1 shows the curing yield, cooking yield, WHC, and moisture content of naturally  
267 converted nitrite, ascorbic acid, and ultrasound-marinated pork loin. The curing yield,  
268 cooking yield, and WHC of naturally converted nitrite and ultrasound on marinated pork loin  
269 did not differ significantly among all treatments ( $P > 0.05$ ). Krause et al. (2011) reported that  
270 the cured meat with natural nitrite did not show a significant difference in the curing yield and  
271 cooking loss. Kim et al. (2019) indicated that the curing yield and cooking loss of cured pork  
272 loin with green nitrite showed no significant difference between the control and the treatment  
273 groups. Similar to several studies, this study also confirmed that there was no difference in the  
274 curing yield and cooking yield of meat products depending on the addition of nitrite and pre-  
275 converted natural nitrite. Similar results with Kim et al. (2019) for the curing were observed  
276 with no significant differences in cooking loss. The moisture content of the pork loin was  
277 highest when treated with nitrite (Table 1). This might be because the moisture content is  
278 affected by the pH of the marinated pork loin.

279

280

281 *3.4. Differences in thermal properties of pork loin*

282 Changes in the thermal properties of pork loins treated with naturally converted nitrite,  
283 ascorbic acid, or ultrasound marination were measured using differential scanning  
284 calorimetry, and the results are presented in Table 2. The analysis revealed endothermic  
285 transitions at peak temperatures of approximately 57.78–58.56°C (peak 1) and 75.18–77.02°C  
286 (peak 2). According to Kim et al. (2023), peak 1 corresponds to the temperature at which  
287 myosin denaturation occurs, whereas peak 2 corresponds to the denaturation of sarcoplasmic  
288 proteins and actin. Additionally, Kim et al. (2021) noted that an increase in peak temperature  
289 indicates enhanced thermal stability of proteins prone to denaturation. Delta H represents the  
290 enthalpy change, reflecting the heat energy required for protein denaturation; a lower Delta H  
291 suggests that the protein is relatively stable before being altered by heat. We hypothesized  
292 that ultrasound treatment would influence the thermal properties of pork loins; however, the  
293 results indicated no significant effects. This lack of impact is likely due to the short duration  
294 of the ultrasound treatment, which was insufficient to alter protein structures.

295  
296 *3.5. Differences in MFI, protein solubility, and shear force of pork loin*

297 The MFI, protein solubility, and shear force of naturally converted nitrite, ascorbic acid,  
298 and ultrasound in the marinated pork loin samples are shown in Table 3. MFI is a good  
299 indicator of the degree of decomposition of meat proteins and can be used to determine the  
300 degree of tenderization in meat. The MFI of the marinated pork loin samples did not differ  
301 significantly among the treatments ( $P>0.05$ ). Total protein solubility and myofibrillar protein  
302 solubility of pork loin samples were higher in treatments with ultrasound (PA+U and  
303 PA+AU) than in treatments without ultrasound. This shows that ultrasound affects  
304 myofibrillar protein solubility but does not affect muscle fragmentation. As mentioned  
305 previously, this is believed to be due to the short ultrasound processing time. The shear force

306 of PC with sodium nitrite was the highest, whereas the shear force of PA+U was the lowest  
307 ( $P<0.05$ ). Kim et al. (2019) reported that the shear force of cured pork loin did not  
308 significantly differ between nitrite and natural nitrite from fermented Swiss chard; however,  
309 the shear force may be affected by pH and moisture content. Shear force is regarded as the  
310 most effective indicator of sensorial tenderness (Holman et al., 2020), and lower shear force  
311 can be positively associated with consumer acceptance of meat (Warner et al., 2021). Looking  
312 at the shear force results, softness tended to increase when nitrite or naturally converted nitrite  
313 was added, and when ascorbic acid was combined, shear force of cured loin was increased.  
314 However, it appears that ultrasound treatment can improve the softness. This improvement in  
315 tenderness appears to be due to myofibrillar protein solubility.

316

### 317 *3.6. Differences in TBARS of pork loin*

318 The effects of naturally converted nitrite and ultrasound on the TBARS of the cured pork  
319 loin were significant (Figure 4). The NC had the highest TBARS value compared with that of  
320 the other treatments with nitrite and converted nitrite ( $P<0.05$ ), and the TBARS values did not  
321 show a significant difference among the treatments with nitrite and converted nitrite ( $P>0.05$ ).  
322 According to Kim et al. (2019), pork loin cured with nitrite from fermented spinach and  
323 organic acids had a lower TBARS than that of the control cured without nitrite. Choi et al.  
324 (2017) reported that meat emulsions with pre-converted nitrite from red beets and ascorbic  
325 acid had higher TBARS values than that those of the control. Kim et al. (2019) reported that  
326 meat products containing organic acids, including ascorbic acid, were effective against lipid  
327 rancidity. Similar to previous results, nitrite or pre-converted nitrite produced through  
328 fermentation is believed to effectively inhibit the rancidity of meat products through lipid  
329 oxidation. Ultrasound treatment appears to have no effect on rancidity or has little effect on  
330 lipid oxidation when nitrite was added. Meanwhile, difference of TBARS values among



331 treatments can be occurred during storage, as a previous research using black tea as a nitrite  
332 replacer (Zhang et al., 2023a). Regarding ascorbic acid, it can retard the lipid oxidation during  
333 storage, by reducing heme iron and hindering free radical formation (Li et al., 2013).  
334 However, ultrasound-treated meat may be more susceptible to the lipid oxidation after  
335 storage, due to the exposure of fats to free radicals formed by ultrasound treatment (Peña-  
336 González et al., 2017). Therefore, the naturally converted nitrite of the marinated pork loin  
337 used alone can effectively suppress lipid oxidation by replacing sodium nitrite, and it was  
338 observed that ultrasound does not have a significant effect on lipid oxidation of freshly  
339 cooked meat. In further studies, the effect of these additives and treatment on the cured meat  
340 during long-term storage should be determined, although previous studies have provided  
341 insights into the changes that occur during storage.

342

### 343 Conclusion

344 When curing meat, pre-converted natural nitrite sources can be used as an alternative to  
345 synthetic nitrites. We used ultrasound and ascorbic acid to improve the quality of meat cured  
346 using naturally pre-converted nitrite raw materials. Ultrasound had a positive effect on  
347 myofibrillar protein solubility and shear force but had no effect on color development, curing  
348 efficiency, or lipid acidity. However, it appears to have a negative effect on residual nitrite.  
349 Therefore, when combining a pre-converted natural nitrite source with ultrasound, ascorbic  
350 acid should be added to help improve the quality of cured meat.

351

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356 **Conflicts of Interest**

357 The authors declare no conflicts of interest.

358

359 **Declarations of interest**

360 None

361

362 **References**

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454 **Table 1. Influence of naturally converted nitrite and ultrasound marination on pH, curing yield, cooking yield, water holding capacity**  
 455 **(WHC), and moisture content of pork loin**

Traits	NC	PC	PA	PA+A	PA+U	PA+AU
pH	5.99±0.00 <sup>b</sup>	6.06±0.01 <sup>a</sup>	5.96±0.00 <sup>c</sup>	5.94±0.00 <sup>d</sup>	5.96±0.01 <sup>c</sup>	5.93±0.01 <sup>d</sup>
Curing yield (%)	111.06±7.45	110.69±0.62	107.94±0.36	108.15±0.30	110.70±1.74	107.95±0.23
Cooking yield (%)	70.10±0.79	69.66±0.68	68.02±0.40	68.73±0.45	70.17±2.26	70.16±0.94
WHC (%)	18.54±0.88	20.68±1.88	18.65±0.94	20.51±1.06	21.59±1.88	19.18±1.03
Moisture content (%)	63.41±0.12 <sup>d</sup>	64.61±0.08 <sup>a</sup>	63.79±0.05 <sup>c</sup>	62.98±0.02 <sup>e</sup>	64.03±0.05 <sup>b</sup>	63.56±0.08 <sup>cd</sup>

456 Values are presented as mean ± standard error (n ≥ 3).

457 <sup>a-d</sup>Different letters within the same row indicate significant differences between treatments ( $P < 0.05$ ).

458 NC, cured without sodium nitrite; PC, cured with sodium nitrite (180 ppm); PA, cured with a pre-converted *Angelica keiskei* extract (180 ppm of  
 459 nitrite); PA+A, cured with a pre-converted *Angelica keiskei* extract (180 ppm of nitrite) and ascorbic acid; PA+U, cured with a pre-converted  
 460 *Angelica keiskei* extract (180 ppm of nitrite) and marinated using ultrasound; PA+AU, cured with a pre-converted *Angelica keiskei* extract (180  
 461 ppm of nitrite) and ascorbic acid, and marinated using ultrasound.

462 **Table 2. Influence of naturally converted nitrite and ultrasound marination on thermal properties of pork loin**

Traits	NC	PC	PA	PA+A	PA+U	PA+AU
Peak <sub>1</sub> temperature (°C)	58.50±0.32	58.43±0.39	58.34±0.63	57.78±0.57	58.56±0.63	58.44±1.00
ΔH <sub>1</sub> (J/g)	0.13±0.01	0.14±0.02	0.17±0.03	0.13±0.01	0.14±0.01	0.14±0.02
Peak <sub>2</sub> temperature (°C)	75.90±0.24 <sup>ab</sup>	75.18±0.37 <sup>b</sup>	77.02±0.49 <sup>a</sup>	75.59±0.47 <sup>b</sup>	75.58±0.44 <sup>b</sup>	75.36±0.31 <sup>b</sup>
ΔH <sub>2</sub> (J/g)	0.55±0.05 <sup>a</sup>	0.30±0.05 <sup>c</sup>	0.30±0.03 <sup>c</sup>	0.45±0.06 <sup>ab</sup>	0.56±0.03 <sup>a</sup>	0.38±0.03 <sup>bc</sup>

463 Values are presented as mean ± standard error (n ≥ 3).

464 <sup>a-c</sup>Different letters within the same row indicate significant differences between treatments (*P*<0.05).

465 NC, cured without sodium nitrite; PC, cured with sodium nitrite (180 ppm); PA, cured with a pre-converted *Angelica keiskei* extract (180 ppm of  
 466 nitrite); PA+A, cured with a pre-converted *Angelica keiskei* extract (180 ppm of nitrite) and ascorbic acid; PA+U, cured with a pre-converted  
 467 *Angelica keiskei* extract (180 ppm of nitrite) and marinated using ultrasound; PA+AU, cured with a pre-converted *Angelica keiskei* extract (180  
 468 ppm of nitrite) and ascorbic acid, and marinated using ultrasound.

469 **Table 3. Influence of naturally converted nitrite and ultrasound marination on myofibril fragmentation index (MFI), protein solubility,**  
 470 **and shear force of pork loin**

Traits	NC	PC	PA	PA+A	PA+U	PA+AU
MFI	116.11±1.28	110.45±1.03	112.27±1.80	113.20±5.13	111.62±1.40	111.33±1.31
Total protein solubility (mg/mL)	9.46±0.04 <sup>c</sup>	9.51±0.05 <sup>c</sup>	9.83±0.05 <sup>b</sup>	9.74±0.09 <sup>b</sup>	10.04±0.00 <sup>a</sup>	10.12±0.02 <sup>a</sup>
Myofibrillar protein solubility (mg/mL)	7.25±0.06 <sup>d</sup>	7.35±0.05 <sup>cd</sup>	7.55±0.05 <sup>b</sup>	7.49±0.11 <sup>bc</sup>	7.81±0.02 <sup>a</sup>	7.87±0.01 <sup>a</sup>
Sarcoplasmic protein solubility (mg/mL)	2.21±0.03 <sup>ab</sup>	2.16±0.01 <sup>b</sup>	2.28±0.01 <sup>a</sup>	2.24±0.03 <sup>a</sup>	2.23±0.02 <sup>ab</sup>	2.25±0.03 <sup>a</sup>
Shear force (N)	44.43±1.71 <sup>d</sup>	57.40±2.67 <sup>a</sup>	51.25±2.55 <sup>bc</sup>	56.26±0.86 <sup>ab</sup>	49.33±1.90 <sup>cd</sup>	51.94±0.76 <sup>bc</sup>

471 Values are presented as mean ± standard error (n ≥ 3).

472 <sup>a-d</sup>Different letters within the same row indicate significant differences between treatments ( $P < 0.05$ ).

473 NC, cured without sodium nitrite; PC, cured with sodium nitrite (180 ppm); PA, cured with a pre-converted *Angelica keiskei* extract (180 ppm of  
 474 nitrite); PA+A, cured with a pre-converted *Angelica keiskei* extract (180 ppm of nitrite) and ascorbic acid; PA+U, cured with a pre-converted  
 475 *Angelica keiskei* extract (180 ppm of nitrite) and marinated using ultrasound; PA+AU, cured with a pre-converted *Angelica keiskei* extract (180  
 476 ppm of nitrite) and ascorbic acid, and marinated using ultrasound.



## Figure captions

**Figure 1.** Influence of naturally converted nitrite and ultrasound marination on  $L^*$  value (A),  $a^*$  value (B), and  $b^*$  value (C) of pork loin. Values are presented as mean  $\pm$  standard error ( $n \geq 3$ ). <sup>a-</sup> Different letters above the bars indicate significant differences between treatments ( $P < 0.05$ ). NC, cured without sodium nitrite; PC, cured with sodium nitrite (180 ppm); PA, cured with a pre-converted *Angelica keiskei* extract (180 ppm of nitrite); PA+A, cured with a pre-converted *Angelica keiskei* extract (180 ppm of nitrite) and ascorbic acid; PA+U, cured with a pre-converted *Angelica keiskei* extract (180 ppm of nitrite) and marinated using ultrasound; PA+AU, cured with a pre-converted *Angelica keiskei* extract (180 ppm of nitrite) and ascorbic acid, and marinated using ultrasound.

**Figure 2.** Influence of naturally converted nitrite and ultrasound marination on curing efficiency of pork loin. Values are presented as mean  $\pm$  standard error ( $n \geq 3$ ). <sup>a-c</sup> Different letters above the bars indicate significant differences between treatments ( $P < 0.05$ ). NC, cured without sodium nitrite; PC, cured with sodium nitrite (180 ppm); PA, cured with a pre-converted *Angelica keiskei* extract (180 ppm of nitrite); PA+A, cured with a pre-converted *Angelica keiskei* extract (180 ppm of nitrite) and ascorbic acid; PA+U, cured with a pre-converted *Angelica keiskei* extract (180 ppm of nitrite) and marinated using ultrasound; PA+AU, cured with a pre-converted *Angelica keiskei* extract (180 ppm of nitrite) and ascorbic acid, and marinated using ultrasound.

**Figure 3.** Influence of naturally converted nitrite and ultrasound marination on residual nitrite of pork loin. Values are presented as mean  $\pm$  standard error ( $n \geq 3$ ). <sup>a-d</sup> Different letters above the bars indicate significant differences between treatments ( $P < 0.05$ ). NC, cured without sodium nitrite; PC, cured with sodium nitrite (180 ppm); PA, cured with a pre-converted *Angelica keiskei*

extract (180 ppm of nitrite); PA+A, cured with a pre-converted *Angelica keiskei* extract (180 ppm of nitrite) and ascorbic acid; PA+U, cured with a pre-converted *Angelica keiskei* extract (180 ppm of nitrite) and marinated using ultrasound; PA+AU, cured with a pre-converted *Angelica keiskei* extract (180 ppm of nitrite) and ascorbic acid, and marinated using ultrasound.

**Figure 4.** Influence of naturally converted nitrite and ultrasound marination on thiobarbituric acid reactive substances (TBARS) of pork loin. Values are presented as mean  $\pm$  standard error ( $n \geq 3$ ). <sup>a-b</sup>Different letters above the bars indicate significant differences between treatments ( $P < 0.05$ ). NC, cured without sodium nitrite; PC, cured with sodium nitrite (180 ppm); PA, cured with a pre-converted *Angelica keiskei* extract (180 ppm of nitrite); PA+A, cured with a pre-converted *Angelica keiskei* extract (180 ppm of nitrite) and ascorbic acid; PA+U, cured with a pre-converted *Angelica keiskei* extract (180 ppm of nitrite) and marinated using ultrasound; PA+AU, cured with a pre-converted *Angelica keiskei* extract (180 ppm of nitrite) and ascorbic acid, and marinated using ultrasound.

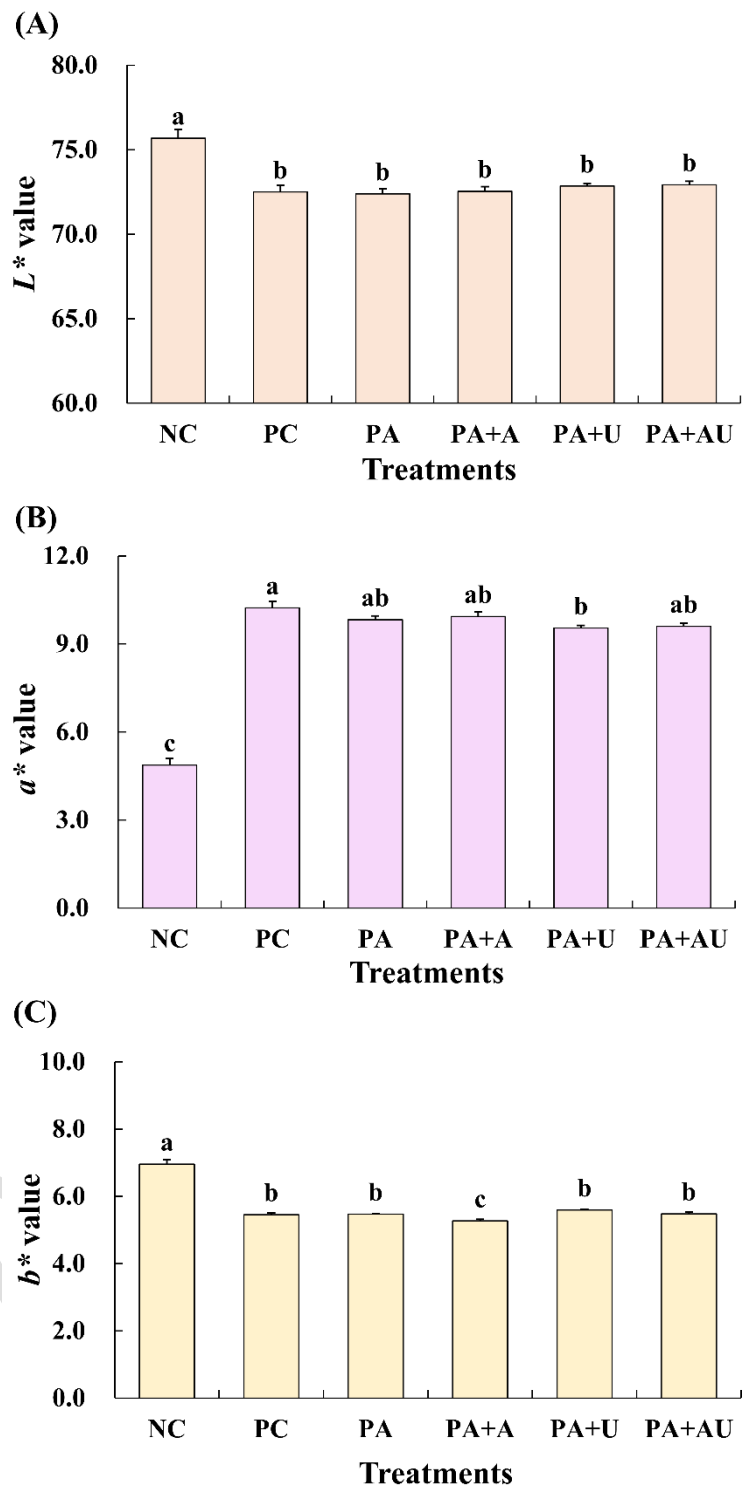


Figure 1

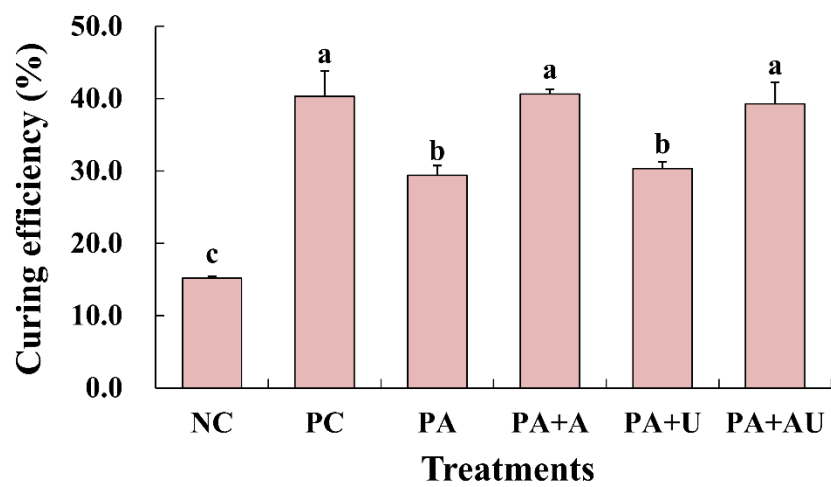


Figure 2

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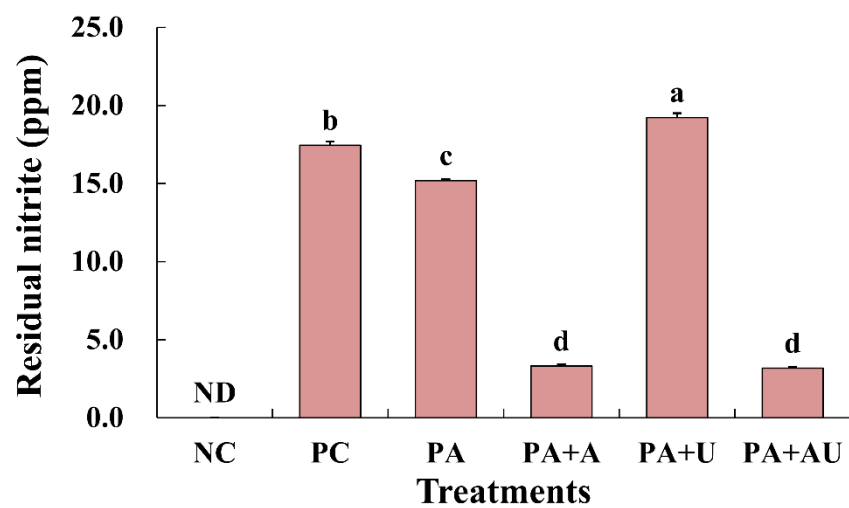


Figure 3

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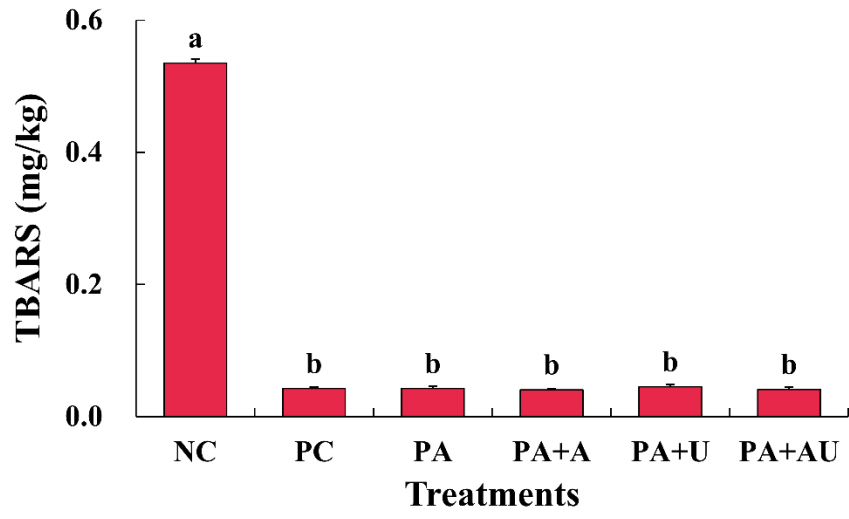


Figure 4

ACCEPTED