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Impact of Creatine Supplementation on the Quality and Storage Characteristics of Emulsified Pork Sausages

Abstract

This study aimed to analyze effects of impact of creatine supplementation on the quality and storage characteristics of emulsified pork sausages. Creatine powder was added at levels of 0.5% (CS1), 1.0% (CS2), 1.5% (CS3), and 2.0% (CS4). Quality characteristics were evaluated on day 0, while storage characteristics were assessed on day 7 following storage under sealed conditions at 4℃ to monitor potential changes over time. The pH increased with higher creatine levels. The cooking yields of CS2 and CS3 were comparable to that of the control group, suggesting that creatine supplementation did not adversely affect the water-holding capacity during cooking. Hardness was highest in CS4, while gumminess and chewiness values were significantly higher in CS2, CS3, and CS4 compared to the control. Thiobarbituric acid reactive substances and total bacterial count values tended to decrease as creatine addition increased, with CS4 being the most suitable formulation for emulsified pork sausages enriched with creatine. However, sensory evaluation scores decreased with increasing creatine levels, indicating the need for further studies to improve sensory properties while maintaining benefits of creatine addition.

Keywords: Emulsified pork sausage, Creatine, Quality properties, Storage characteristics

Introduction

In modern society, as the number of nuclear families consisting of 1-2 people increases, main consumption choices in eating habits are economic efficiency and convenience (Kim et al., 2018). Accordingly, the trend of convenience food is increasing. In particular, the popularity of meat products manufactured using meat is steadily growing (Dhir and Singla, 2019). Among various meat products, emulsified sausages are products manufactured by combining meat and salt to extract salt-soluble proteins that can bind fat and water (Dickinson, 2012). Since emulsified sausages can easily use additives, they can be manufactured into various products.

Food additives are continuously being studied because they can affect physical properties or nutritional characteristics of emulsified sausages (Santhi et al., 2017; Rofers, 2001). In addition, research and development are ongoing due to the addition of various additives to the wellness trend (Ponnampalam et al., 2019). Zaini et al. (2020) have conducted a study to improve physical or nutritional characteristics by adding banana peels. One study has been conducted to improve protein content and anti-nutritional by adding pigeon peas (Gomezulu and Mongi, 2022). Accordingly, it can be seen that research is ongoing to improve nutritional and protein characteristics of products by adding additives.

Creatine (N-carbamimidoyl-N-methylglycine) is exogenously consumed and endogenously synthesized through complex interorgan processes (Clarke et al., 2021). Creatine is a substance involved in the body's energy production process and is known to be helpful for those who need protein supplementation (Antonio et al., 2021). In addition, research results have been reported that it can be used as an antioxidant directly or indirectly (Qasim and Mahmood, 2011; Sestili et al., 2011). Therefore, it is thought that creatine can be used as an additive to improve the nutritional and storage properties of the product.

Therefore, this study was conducted to determine whether pork emulsified sausages with added creatine could improve nutritional properties without changing physical properties or enhance storage by performing direct or indirect antioxidant functions. After producing creatine sausages according to the amount of addition, an experiment was conducted to analyze the appropriate amount of addition.

Materials and Methods

Preparation of emulsified pork sausage

The manufacturing process for emulsified pork sausages involved preparation of pork loin and back fat (Bosungnokdon, Seoul, Korea). Pork loin and back fat were ground using a grinder equipped with a 5 mm plate (PC-114L, Mainca, Spain). The ground pork loin was sequentially mixed with salt, CurePhos 700 (BK Giulini GMBH, Germany), nitrite, and half the amount of ice to extract salt-soluble proteins. Then, pork back fat, admixture, remaining ice and creatine (Nutricost, Vineyard, UT, USA) were added and emulsified using a silent cutter (CM-21, Mainca, Spain) at 5℃ for 1 min (Table 1). Afterwards, the emulsified mass, which did not exceed 10℃, was filled into 45ø fiber casings (Kalle GmbH, Wiesbaden, Germany) using a vacuum filler (VF-608, Handtmann, Cypress, CA, USA) and heated in a chamber (Metatek, Nonsan, Korea) at 80℃ for approximately 40 min until the core temperature reached 75℃. Cooked sausages were cooled, sealed, and used for subsequent experiments.

pH

Before and after cooking, samples were weighed and mixed with distilled water at a ratio of 1:10. The mixture was then homogenized using a stomacher (400 Circulator, Seward, Bohemia, NY, USA) for 30 seconds and the pH was measured using a pH meter (pH-200L, Istek, Seoul, Korea).

Color

Cooked emulsified sausage color was measured using a spectrocolorimeter (JX-777, Color Techno. System Co., Tokyo, Japan) standardized with a white plate (L = 94.04, a = 0.13, b = 0.51). A D_{65} white fluorescent light source was used. Results are expressed in terms of lightness (L^*), redness (a^*), and yellowness (b^*). The hue angle was determined using $\tan^{-1}(b^*/a^*)$, while chroma was calculated as $(a^{*}\times 2+b^{*}\times 2)^{1/2}$.

Water holding capacity (WHC)

The WHC was measured following the method described by Laakkonen et al. (1970). The weight of a 2 mL tube specifically designed for WHC measurement was recorded. Subsequently, 0.5 ± 0.05 g of each sample was accurately weighed and placed in a tube. Combined weight of the tube and the sample was measured before cooking the tube in a water bath at 80℃ for 20 minutes. After cooking, the tube was cooled at room temperature for 10 minutes and then centrifuged at 2,000 rpm for 10 minutes at 4℃. The weight was recorded again and the water-holding capacity was calculated using the formula provided below:

$$WHC = \frac{Total\ \ moisture\ \ (\%) - \ \ free\ \ moisture\ \ (\%)}{Moisture\ \ (\%)} \times 100$$

$$\text{Free moisture} = \frac{weight\ \ before\ \ centrifugation\ \ - \ \ weight\ \ after\ \ centrifugation}{weight\ \ of\ \ sample\ \ \times fat\ \ coefficient} \times 100$$

$$\text{Fat coefinicent} = \frac{Fat\ \ (\%)}{100}$$

Cooking yield

The emulsified pork mixture was filled into the casing and the initial weight was recorded. After heating and cooling at 80℃ for 40 minutes, the final weight was measured and the cooking yield was calculated as a percentage.

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

Texture profile analysis

To evaluate textural properties of emulsified sausages, samples were cut into cube shapes with dimensions of 1 × 1 × 1 cm. Experiments were conducted using a rheometer (Compac-100, Sun Scientific Co., Japan) equipped with a φ20 mm adapter to perform mastication and cutting tests. Experimental conditions were as follows: a head speed of 1.0 mm/s, a distance of 1.0 mm, and a force of 2.0 kg. Hardness (kg), springiness, and cohesiveness were measured directly, while gumminess (kg) and chewiness (kg) were derived from these values.

Thiobarbituric acid reactive substances (TBARS)

TBARS analysis was conducted on samples collected on the 7th day. A 10 g sample was homogenized with 15 mL of cold 10% perchloric acid and 25 mL of triple-distilled water at 10,000 rpm for 10 seconds using a homogenizer. The homogenate was filtered through a Whatman No. 2 filter paper. Subsequently, 5 mL of the filtrate was mixed with 5 mL of 0.02 M thiobarbituric acid solution. The mixture was thoroughly blended and left in a dark, cool place for 16 hours. Absorbance was then measured at 529 nm using a spectrophotometer (DU-650, Beckman, Fullerton, CA, USA). Triple-distilled water was used as a blank. In this study, the standard curve calculated with malondealdehyde was y = 0.1975 - 0.0011 (r =

0.999), and the data were calculated by inputting the absorbance value into this formula. TBA values are expressed as milligrams of malonaldehyde per kilogram of sample (mg malonaldehyde/kg).

Volatile basic nitrogen (VBN)

VBN analysis was conducted for samples collected on the 7th day. A 10 g sample was homogenized with 90 mL of distilled water at 10,000 rpm for approximately 30 seconds. The homogenate was filtered using a Whatman No. 2 filter paper and the filtrate was used for further analysis. One milliliter of the filtrate was placed in the outer chamber of a Conway unit. In the inner chamber, 1 mL of 0.01 N boric acid solution and 50 mL of an indicator solution (0.066% methyl red + 0.066% bromocresol green) were added. After applying glycerin to the lid's contact surface for sealing, the lid was then closed. Subsequently, 1 mL of 50% potassium carbonate (K_2CO_3) solution was added to the outer chamber and the unit was immediately sealed. The container was gently mixed horizontally and incubated at 37℃ for 120 minutes. After incubation, the boric acid solution in the inner chamber was titrated with 0.02 N sulfuric acid (H_2SO_4). VBN content was expressed as milligrams per 100 g of sample (mg%):

$$\text{VBN (mg\%)} = \frac{((a-b)\times F \times 14.007}{amount\ \ of\ \ sample} \times 100$$

a: Amount of sulfuric acid injected (ml)

b: Amount of sulfuric acid injected into blank (ml)

f: 0.02N H_2SO_4 standardization index

Total bacteria count (TBC)

TBC was determined using the serial dilution method for samples stored for 7 days. A 10 g sample was homogenized with 90 mL of 0.1% peptone solution in a stomacher bag for 30 seconds. The homogenized sample was serially diluted and inoculated onto plate count agar (PCA). Plates were incubated at 37℃ for 24 hours. After incubation, colonies were counted using a colony counter. Results are expressed as Log CFU/g.

Sensory evaluation

This project was approved by the Institutional Review Board (IRB) of Chungbuk National University (IRB Approval Number: CBNU-202302-HR-0017). The institution stated in advance that it would protect the rights and privacy of all participants. The panel consisted of 10 people aged 20-30 years who had experience eating emulsified sausages. The panel was trained on sufficient knowledge of samples, terminology, and evaluation criteria. Each sample was randomly presented with a 1-minute interval between tests. Trained panelists conducted sensory evaluations to assess appearance, elasticity, juiciness, off-flavor, flavor, saltiness, and overall acceptability. The assessment utilized a 9-point hedonic scale, where 1 represented "very poor" and 9 represented "excellent." Sausages from each treatment group were cut into approximately 1 cm square pieces and presented to the 10 panelists for evaluation. Each panelist rated samples based on the given criteria.

Statistical Analysis

All statistical analyses for this experiment were performed using the General Linear Model procedure in the SAS program (SAS Institute, Cary, NC, USA). Duncan's multiple range test was employed to compare treatment means, with significance set at the 5% level.

Results and Discussion

pH and Color

The pH before and after cooking and color after cooking emulsified pork sausages supplemented with creatine are presented in Table 2. The uncooked pH of CS1 was significantly lower than those of other treatments, while CS3 exhibited the highest value ($p < 0.05$). This can be attributed to the relatively high pH of the creatine powder used in this study (7.19). Cooked pH values were generally higher than uncooked values, which could be explained by denaturation of proteins during cooking. This process can disrupt internal hydrogen and soluble bonds within protein molecules, resulting in the release of alkaline groups from the protein structure (Yu et al., 2016). Cooked pH values of CS1 and CS2 were significantly ($p < 0.05$) lower than that of the control, consistent with the trend observed in uncooked pH values.

Regarding color properties, lightness showed no significant differences among treatments. However, redness exhibited an increasing trend with higher creatine levels, except in the control group. Among the treatments, CS4 displayed the highest redness value, likely due to creatine's role in stabilizing the inherent red color of meat (Kanokruangrong et al., 2025). Yellowness initially increased with creatine addition but decreased in CS4. The hue angle, a metric where 0° indicates redness and 90° indicates yellowness (Bao et al., 2020), was lowest in the control group but highest in CS3. Chroma, which measures color saturation or intensity (Tiwari et al., 2022), increased with higher levels of creatine. This trend likely reflects simultaneous increases in redness and yellowness. As noted by Pathare et al. (2012), higher chroma values correspond to greater perceived color intensity, suggesting that creatine-enhanced sausages may appeal more strongly to consumers in terms of visual quality.

WHC and cooking yield

Table 3 presents WHC and cooking yield of emulsified pork sausages supplemented with creatine. WHC refers to the ability of meat to retain water, which is highly correlated with pH and cooking yield (Warner, 2023). However, WHC showed no significant differences between control and treatment groups in this study.

Cooking yield was significantly higher in the control group than in CS1 and CS4 (both $p < 0.05$). However, CS2 and CS3 showed no significant differences in cooking yield from the control. This trend appears to be influenced by relatively higher pH values observed in CS2 and CS3. Janicki and Buzała (2013) have reported that dietary creatine supplementation can improve WHC in pork, reducing drip loss and enhancing meat quality. These findings aligned with the observation that creatine-enhanced sausages might exhibit improved textural and economic attributes, both of which are critical to consumer satisfaction and product marketability (Priya et al., 2022). Based on these results, creatine supplementation at levels of 1.0% and 1.5% appears to improve cooking yield while maintaining desirable product characteristics. These concentrations of creatine may provide practical advantages for enhancing product performance and consumer appeal.

Texture profile analysis

Texture properties of emulsified pork sausages supplemented with creatine are summarized in Table 4. Hardness was significantly higher in CS4 than in other treatments ($p < 0.05$), while springiness did not differ significantly among groups. This could be attributed to the powdered form of creatine and its role as an additive that could support muscle contraction (Tian et al., 2022). Gumminess increased with increasing creatine levels, with CS2, CS3, and CS4 showing significantly higher values than the control ($p < 0.05$). A similar trend was observed for chewiness, indicating consistency in the influence of creatine on textural properties. Cohesiveness was significantly higher in CS2 than in the control (p < 0.05). Mirsadeghi et al. (2019) have reported that creatine generated during cooking can enhance texture characteristics such as gumminess and chewiness. Additionally, Chen et al. (2017) have found that thermal degradation products, including free amino acids (e.g., glucose, creatine, creatinine), can improve cohesiveness, stickiness, and elasticity in meat products. These findings suggest that supplementing creatine at levels of 1.5% and 2.0% can enhance textural qualities of pork sausages, contributing to improved sensory properties and potentially influencing protein enrichment. This level of supplementation appears to be optimal for improving product texture while leveraging functional benefits of creatine.

VBN, TBARS, and TBC

VBN, TBARS, and TBC values of emulsified pork sausages supplemented with creatine after 7 days of storage are presented in Table 5. VBN values showed no significant differences between control and treatment groups, indicating that creatine supplementation did not lead to protein spoilage during storage. TBARS, a measure of lipid oxidation, was the highest in the control group. It exhibited a decreasing trend as creatine content increased. Lawler et al. (2002) have reported that creatine has antioxidant properties such as scavenging $ABTS^{+}$, O•−2, and $OONO^{-}$ radicals. Similarly, Lahučký et al. (2012) have demonstrated that dietary supplementation with creatine and vitamin E can increase alpha-tocopherol levels in pork, further enhancing its oxidative stability. These antioxidant effects might have contributed to reduced TBARS values in creatine-treated sausages.

TBC also decreased with increasing creatine levels. CS2, CS3, and CS4 showed significantly lower TBC values than the control group (p < 0.05). This reduction in bacterial count can also be attributed to antioxidant and potential antimicrobial effects of creatine. Marques and Wyse (2019) have suggested that creatine could exert direct and indirect antioxidant effects, further enhancing the preservation of meat products. Tsvetkova et al. (2023) reported that creatine exhibits high ABTS radical scavenging activity. Given that free radicals play a crucial role in lipid peroxidation, this radical-scavenging property suggests that creatine may contribute to the inhibition of lipid peroxidation by reducing oxidative stress. Additionally, since oxidative stress can influence microbial growth and survival, creatine's antioxidant activity may also play a role in modulating microbial populations, potentially contributing to microbial inhibition. However, further research is needed to establish a direct link between creatine and antimicrobial effects. These findings suggest that creatine supplementation can improve the storage stability of emulsified pork sausages. Optimal creatine levels for enhancing storage properties appear to be 1.5% and 2.0% as these concentrations could provide superior oxidative and microbial stability.

Sensory evaluation

Table 6 presents sensory evaluation results of emulsified pork sausages supplemented with creatine at varying levels. Appearance scores were the highest for CS2 and CS3, indicating improved visual appeal with creatine supplementation at these levels. Juiciness, flavor, off-flavor, and springiness did not show significant differences among treatment groups. These results suggest that creatine does not adversely affect sensory attributes while contributing positively to quality improvements. The overall acceptability was the highest for CS1, which could be attributed to its relatively higher scores for juiciness, flavor, and off-flavor than other treatment groups. Mora et al. (2008) have reported that higher concentrations of creatine/creatinine in products could lead to a bitter taste, which is thought to be due to the chemical structure of creatine containing nitrogen (guanidine group; Perla and Jayanty, 2013). Therefore, this may explain the decrease in overall acceptability as creatine levels increased. Based on sensory evaluation results, creatine supplementation at 1.0% and 1.5%

could enhance the appearance of sausages, while 0.5% creatine achieved the highest overall acceptability. These findings indicate that while moderate creatine supplementation can improve quality and appearance, further studies are needed to optimize sensory acceptability, particularly for higher supplementation levels.

Conclusions

This study evaluated the quality and storage characteristics of emulsified pork sausages supplemented with creatine at 0.5%, 1.0%, 1.5%, and 2.0% levels. Creatine addition elevated pH values without significantly affecting water-holding capacity, and the cooking yield improved in the 0.5% and 2.0% groups relative to the control. Texture profile analysis indicated that hardness increased with creatine treatment, while chewiness and gumminess also tended to rise, supporting previous findings on textural enhancement.

Regarding storage properties, creatine did not alter volatile basic nitrogen values, implying negligible effects on protein spoilage. In contrast, TBARS and total bacterial counts declined with higher creatine levels, highlighting its antioxidant capacity and beneficial impact on oxidative and microbial stability. Despite these improvements, sensory evaluations revealed reduced overall acceptability at higher creatine concentrations, likely due to bitterness from increased creatine/creatinine conversion.

In conclusion, while creatine supplementation can enhance the quality and storage attributes of pork sausages, addressing the associated sensory drawbacks remains essential. The study recommends 1.5% to 2.0% creatine levels, contingent upon further refinements to improve flavor.

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Table 1. Formulation and experimental design for emulsified pork sausages

Treatments	Control	CS1	CS2	CS3	CS4
Pork lean meat	69.57	69.57	69.57	69.57	69.57
Pork Fat	17.39	17.39	17.39	17.39	17.39
Water	13.04	13.04	13.04	13.04	13.04
Total	100	100	100	100	100
Salt	1.50	1.50	1.50	1.50	1.50
Curafos 700	0.15	0.15	0.15	0.15	0.15
$NaNo_2$	0.01	0.01	0.01	0.01	0.01
Sugar	0.60	0.60	0.60	0.60	0.60
Garlic powder	0.50	0.50	0.50	0.50	0.50
Vitamin-C	0.04	0.04	0.04	0.04	0.04
Creatine		0.50	1.00	1.50	2.00

Table 2. pH values of cooked and uncooked emulsified pork sausages added with creatine and color of cooked emulsified pork sausages added with creatine

Traits	Control	CS1	CS2	CS3	CS4
pH of uncooked	5.93±0.00^{c}	5.89±0.00^{d}	5.92±0.00^{c}	5.97±0.01^{a}	5.95±0.00^{b}
pH of cooked	6.06±0.02^{a}	6.00±0.00^{b}	6.03±0.02^{b}	6.07±0.00^{a}	6.07±0.00^{a}
Lightness	79.60±0.62	80.05±0.76	79.50±0.36	79.74±0.30	79.76±0.17
Redness	5.50±0.12^{bc}	5.42±0.14^{c}	5.56±0.12^{ab}	5.60±0.06^{ab}	5.66±0.0^{a}
Yellowness	8.75±0.08^{c}	8.87±0.21^{bc}	8.98±0.19^{b}	9.19±0.15^{a}	9.04±0.08^{ab}
Hue value	57.84±0.59^{c}	58.57±0.80^{ab}	58.24±0.45^{abc}	58.66±0.56^{a}	57.93±0.21^{bc}
Chroma	10.34±0.11^{c}	10.39±0.24^{bc}	10.56±0.23^{ab}	10.76±0.14^{a}	10.66±0.09^{a}

CS1: sausage addition creatine 0.5%, CS2: sausage addition creatine 1.0%, CS3: sausage addition creatine 1.5%, CS4: sausage addition creatine 2.0%.

$^{a-d}$ Means with different superscriptions within the same row differ (p<0.05).

Table 3. Water holding capacity (WHC) and cooking yield (%) of emulsified pork sausages added with creatine

Traits (%)	Control	CS1	CS2	CS3	CS4
WHC	75.89±6.65	73.10±4.76	71.67±1.65	73.19±6.50	71.83±6.43
Cooking yield	95.30±0.62^{a}	94.47±0.56^{b}	95.10±0.61^{ab}	94.90±0.23^{ab}	94.45±0.38^{b}

CS1: sausage addition creatine 0.5%, CS2: sausage addition creatine 1.0%, CS3: sausage addition creatine 1.5%, CS4: sausage addition creatine 2.0%.

$^{a-b}$ Means with different superscriptions within the same row differ significantly (p < 0.05).

Table 4. Texture profile analysis of emulsified pork sausages added with creatine

Traits	Control	CS1	CS2	CS3	CS4
Hardness (kg)	2.65±0.31^{b}	2.94±0.27^{b}	2.9±0.16^{b}	2.99±0.37^{b}	3.43±0.33^{a}
Springiness	81.85±7.13	80.27±6.02	80.53±2.99	80.11±2.32	77.48±2.96
Gumminess (kg)	1.64±0.35^{b}	2.03±0.52^{ab}	2.37±0.26^{a}	2.35±0.46^{a}	2.51±0.58^{a}
Chewingness (kg)	1.34±0.28^{b}	1.65±0.49^{ab}	1.91±0.25^{a}	1.89±0.40^{a}	1.95±0.48^{a}
Cohesiveness	62.36±13.23^{b}	68.74±14.58^{ab}	80.71±6.30^{a}	78.23±8.71^{ab}	72.76±13.34^{ab}

CS1: sausage addition creatine 0.5%, CS2: sausage addition creatine 1.0%, CS3: sausage addition creatine 1.5%, CS4: sausage addition creatine 2.0%.

$^{a\text{-}b}$ Means with different superscriptions within the same row differ significantly ($p < 0.05$).

Table 5. VBN, TBARS, and TBC of emulsified pork sausages added with creatine at 7 days after storage

Traits	Control	CS1	CS2	CS3	CS4
VBN (mg/100 g)	9.93±0.42	8.92±1.55	7.96±0.19	7.82±0.01	8.10±0.01
TBARS (mg MDA/kg)	0.06±0.01^{a}	0.04±0.01^{ab}	0.04±0.01^{ab}	0.03±0.02^{ab}	0.03±0.02^{b}
TBC (CFU/g)	4.23±0.22^{a}	4.19±0.31^{ab}	3.96±0.36^{b}	3.91±0.38^{b}	3.83±0.12^{b}

CS1: sausage addition creatine 0.5%, CS2: sausage addition creatine 1.0%, CS3: sausage addition creatine 1.5%, CS4: sausage addition creatine 2.0%.

$^{a-d}$ Means with different superscriptions within the same row differ significantly (p < 0.05).

Table 6. Sensory evaluation of emulsified pork sausages added with creatine

Traits	Control	CS1	CS2	CS3	CS4
Appearance	5.75±1.48^{ab}	6.00±1.41^{b}	7.38±1.22^{ab}	7.63±1.32^{a}	6.63±2.18^{ab}
Juiciness	5.88±1.17	6.00±1.50	5.38±2.06	5.88±1.62	6.00±1.73
Flavor	6.63±0.70	6.38±1.22	6.13±1.45	5.63±1.73	6.00±0.87
Off-flavor	3.75±1.09	3.50±1.00	3.63±1.22	4.00±1.66	3.75±1.92
Springiness	6.00±0.71	6.38±0.99	7.00±1.12	6.63±1.58	6.38±1.80
Overall preference	6.56±0.50^{ab}	6.67±1.20^{a}	6.56±1.32^{ab}	6.56±1.22^{ab}	6.11±1.12^{b}

CS1: sausage addition creatine 0.5%, CS2: sausage addition creatine 1.0%, CS3: sausage addition creatine 1.5%, CS4: sausage addition creatine 2.0%.

$^{a\text{-}b}$ Means with different superscriptions within the same row differ significantly ($p < 0.05$).