pagepress

Italian Journal of Food Safety

Italian Journal of Food Safety 2026; 15:14812
doi:10.4081/ijfs.2026.14812

©Copyright: the Author(s), 2026
Licensee PAGEPress, Italy

Effects of mulberry powder on quality, texture, oxidative stability, and microbiological characteristics of white pork sausage (Moo Yor) during refrigerated storage

Viboon Pongkanpai,[1] Chatchawan Chotimarkorn,[1] Patima Permpoonpattana,[2] Saneeta Salam,[2] Umaporn Pastsart[2]

[1]Department of Food Technology, Faculty of Innovative Agriculture, Fisheries and Food, Prince of Songkla University, Surat Thani Campus; [2]Department of Agricultural Science and Technology, Faculty of Innovative Agriculture, Fisheries and Food, Prince of Songkla University, Surat Thani Campus, Thailand

Abstract

The study evaluated the effects of mulberry (*Morus alba* L.) powder as a natural antioxidant on color, texture, antioxidant activity, lipid oxidation, and microbial quality of white pork sausage (Moo Yor) during refrigerated storage. Four treatment groups were prepared. Group 1 was formulated with 0% mulberry (control), group 2 was formulated with 1% mulberry, group 3 was formulated with 2% mulberry and group 4 was formulated with butylated hydroxytoluene (BHT) 0.02%. The results showed that the 2% mulberry group had significantly lower lightness (L*) values and higher redness (a*) values ($p<0.05$). Texture profile analysis showed that none of the texture parameters were significantly affected ($p>0.05$). Although microbial counts increased during storage, no significant differences in total plate counts were observed among the treatments ($p>0.05$). Moreover, the addition of mulberry led to a higher antioxidant activity and a lower lipid oxidation ($p<0.05$). These findings indicate that mulberry powder can effectively replace BHT as a natural antioxidant, helping to protect meat products from oxidative deterioration while maintaining product quality without negatively affecting the texture or microbial quality of white pork sausage during refrigerated storage.

Key words: mulberry, white pork sausage, texture profile, antioxidant activity, oxidation.

Correspondence: Umaporn Pastsart, Department of Agricultural Science and Technology, Faculty of Innovative Agriculture, Fisheries and Food, Prince of Songkla University, Surat Thani Campus, Thailand 84000. Tel.: +66 77 278891. E-mail: umaporn.p@psu.ac.th

Introduction

The white pork sausage, Moo Yor, is a traditional meat emulsion sausage and a famous product in the northern and northeastern region of Thailand. It is made by grinding and blending meat with fat and various curing and flavoring agents, such as salt, sugar, pepper, and tapioca starch. Normally, white pork sausage has a short shelf life, which is about 2 weeks in the refrigerator (Klinjapo and Krasaekoopt, 2021). In processed meat products, one of the most important concerns is deterioration from the oxidation of fats and proteins. In particular, the oxidation of fats often results in changes in color, flavor, texture, and nutritional values (Kanner, 1994). However, processed meat products are often loaded with synthetic antioxidant and antimicrobial additives to reduce oxidation and inhibit spoilage microorganisms such as butylated hydroxytoluene (BHT). These synthetic additives used in the meat processing industry have a detrimental effect on the consumer's health. Nowadays, there is an awareness of the consumption of these substances, as they may remain and be unsafe for health (Saito *et al.*, 2003). Making processed meat products safer can be done by substituting synthetic substances that affect consumers with natural colorants and antioxidants in the products, *e.g.*, flavonoids, which help to prevent free radicals from oxidations and prolong the shelf life of products (Tsanova-Savova *et al.*, 2018).

Mulberry (*Morus alba* L.) belongs to the genus *Morus* and belongs to the family Moraceae and is widely found in Southeast Asian countries. Mulberry fruit has a delicious taste and high nutritional value as a source of polyphenols, especially flavonoids, flavanones, and anthocyanins (Khalifa *et al*., 2018). Mulberry fruits are also abundant in essential vitamins, minerals, and a diverse range of phenolic compounds such as cyanidin-3-O-glucoside, cyanidin-3-O-rutinoside, rutin, isoquercitrin, resveratrol, and caffeic acid (Xu *et al.*, 2018). These bioactive phenolic compounds play several important biological roles, including antioxidant, antimicrobial, anti-inflammatory, hepatoprotective, and anti-apoptotic activities. They can also enhance immune responses, which highlights their potential applications in the food, pharmaceutical, and cosmetic industries (Natić *et al.*, 2015). When incorporated into meat systems, mulberry can enhance antioxidant capacity and help maintain product quality during storage. Previous studies have demonstrated that the addition of mulberry to various meat products, such as minced pork, dried pork, and Cantonese sausage, effectively enhanced antioxidant potential and improved overall product stability during shelf life (Xu *et al.*, 2018; Xiang *et al.*, 2019; Cheng *et al.*, 2020). Moreover, the phenolic constituents of mulberries may interact with proteins through covalent and non-covalent mechanisms, thereby influencing structural stability, functional groups, solubility, and aggregation, which can ultimately modify protein functionalities in either beneficial or adverse ways (Cao *et al.*, 2016). Besides its antioxidant properties, mulber-

ry contains anthocyanins that may influence the color characteristics of meat products. Therefore, mulberry powder (MP) was selected as a natural source of antioxidants for white pork sausage, while its potential effects on product color were also evaluated. Therefore, the objective of this study was to investigate the effect of MP as a natural substitute for BHT in white pork sausage, focusing on its influence on appearance, texture, antioxidant activity, lipid oxidation, and microbial quality during refrigerated storage.

Materials and Methods

Preparation of mulberry fruit powder

The ripe fruits of mulberry were washed and dried at 60°C in a hot air oven to prepare dried mulberry with approximately 22.3% dry matter content. Subsequently, dried mulberry was ground to obtain fine mulberry fruit powder by passing through a 20-mesh sieve. Then mulberry fruit powder was placed in a vacuum bag and stored at -20°C until use.

Preparation of white pork sausage with the addition of mulberry fruit powder

The white pork sausage consisted of 63% lean pork, 25% pork back fat, 7% ice, 2% sugar, 1.5% salt, 1% tapioca flour, and 0.5% pepper. All ingredients were obtained from the local market in Surat Thani province, Thailand. Briefly, lean pork, all flavoring agents, and half of the ice were chopped for 1 minute, and then pork back fat and the remaining ice were added and chopped until they were homogenous. While chopping, the temperature of the mixture was controlled below 15°C. Subsequently, the batter of sausage was divided into 4 groups (3 replicates in each group) as follows: the batter was mixed with 0%, 1%, 2% MP, and 0.02% BHT (Loba Chemie, Mumbai, India). The addition was based on batter weight. All treated groups were wrapped with banana leaves in a cylindrical shape, about 100 g each cylinder, then steamed for about 30 minutes until the white pork sausage was well cooked.

Color value (CIE Lab)

Internal color of white pork sausage was measured in terms of the CIE-Lab coordinates L* (lightness), a* (redness) and b* (yellowness) with a Hunterlab Miniscan color meter (Reston, VA, USA) (D65 light source, 10° standard observer, 45°/0° geometry, 1-inch light surface, white standard) following standard procedures.

Texture profile analysis

Texture profile analysis (TPA) was performed in duplicate for each sample at day 0 of storage using a Brookfield CT3 texture analyzer (Middleboro, MA, USA) (10,000 g) equipped with a TA-SBA shear blade probe. Pork sausages were cut into pieces measuring 5 cm in length, 1 cm in width, and 1 cm in height. Samples were compressed to 75% of their original height in a double-cycle test at a speed of 2 mm/s. The TPA parameters, including hardness, adhesiveness, cohesiveness, springiness, gumminess, and chewiness, were recorded.

Antioxidant activity

Antioxidant capacity of white pork sausage was determined using the DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging assay following the procedure of Wang *et al.* (2019), with minor modifications. Analyses were performed at 0, 5, and 10 days of storage after sausages were wrapped in oxygen-permeable film and kept at 4°C under fluorescent light. DPPH dissolved in ethanol served as a free radical donor, and the decrease in absorbance at 517 nm was measured spectrophotometrically. For extraction, 2.5 g of sausage was homogenized with 7.5 mL ethanol, shaken for 10 minutes at room temperature, and centrifuged at 1,800 rpm for 10 minutes. The supernatant (0.5 mL) was then mixed with 3.5 mL of 0.1 mM DPPH solution and incubated in the dark for 20 minutes. Ethanol served as the blank. All measurements were performed in triplicate. DPPH scavenging activity (%) was calculated as [Eq. 1]:

$$\text{Scavenging activity (\%)} = (1 - As/Ac) \times 100 \quad \text{[Eq. 1]}$$

where As is the absorbance of the sample and Ac is the absorbance of the control.

Determination of lipid oxidation

Lipid oxidation of white pork sausage was evaluated on days 0, 5, and 10 of storage using the thiobarbituric acid reactive substances (TBARS) assay as described by Tarladgis *et al.* (1960). Results were expressed as µg malondialdehyde (MDA) per g of meat. Briefly, 10 g of sample was homogenized with 40 mL of distilled water and 1 mL of BHT solution using an Ultra-Turrax homogenizer at 13,000 rpm. The homogenate was mixed with 3 mL of 4 M HCl and subjected to distillation. A 5 mL portion of the distillate was combined with 1 mL of TBA reagent, heated in a boiling water bath for 35 minutes, cooled to room temperature, and the absorbance was recorded at 532 nm.

Microbiological analysis

Total plate count (TPC) of white pork sausage was determined according to standard microbiological procedures. Samples were analyzed on days 0, 5, and 10 of refrigerated storage. For analysis, 1 g of sausage sample was aseptically transferred into a sterile tube containing 9 mL of sterile 0.9% NaCl and homogenized. Serial decimal dilutions were then prepared, and appropriate dilutions were spread in duplicate on plate count agar. Plates were incubated at 37°C for 24 hours. Microbial counts were expressed as log CFU/g of sample.

Statistical analysis

Data were analyzed for the fixed effect of treatment using analysis of variance. Differences among means were compared by Duncan's multiple range test at a significance level of $p<0.05$. All statistical analyses were performed with SPSS software (IBM, Armonk, NY, USA).

Results

Color

The color values of white pork sausage during refrigerated storage (0, 5, and 10 days) are presented in Table 1. Sausages supplemented with 1% and 2% MP exhibited significantly lower L* and higher a* compared with the control and BHT treatments ($p<0.05$). In contrast, mulberry supplementation had no significant effect on b* throughout storage ($p>0.05$).

Texture profile analysis

TPA results of white pork sausage at day 0 of storage are sum-

OPEN ACCESS

marized in Table 2. MP supplementation had no significant effect on hardness, adhesiveness, springiness, cohesiveness, gumminess, or chewiness ($p>0.05$). Although group 1 (control) and group 4 (BHT) tended to show higher values for hardness, cohesiveness, gumminess, and chewiness compared with groups 2 and 3 (1% and 2% mulberry), these differences were not statistically significant ($p>0.05$).

Antioxidant activity

Figure 1 illustrates the DPPH radical scavenging activity of white pork sausage during refrigerated storage. Sausages with MP 2% had the highest antioxidant activity on day 0, followed by MP 1%, BHT, and the control ($p<0.05$). On day five, a similar pattern was seen, with MP 2% > MP 1% > BHT > control ($p<0.05$) being the order of antioxidant activity. By day 10, the antioxidant capacity of sausages supplemented with mulberries was consistently higher than that of BHT and the control ($p<0.05$). These findings suggest that MP, especially at 2%, can effectively boost the antioxidant qualities of white pork sausage and could be used as a natural substitute for artificial antioxidants in cold storage.

Lipid oxidation

The lipid oxidation values of white pork sausage during cold storage are presented in Figure 2. On day 0, sausages supplemented with 1% and 2% MP showed significantly lower values ($p<0.05$), while the control group had the highest TBARS value ($p<0.05$), followed by the BHT group. By day 5, lipid oxidation increased across all treatments, but the lowest TBARS values were consistently observed in the mulberry-supplemented groups, particularly at 2% ($p<0.05$). A similar trend was evident on day 10, where the control group had the greatest extent of lipid oxidation, followed by the BHT group, whereas sausages with 1% and 2% MP maintained significantly reduced MDA levels ($p<0.05$). Overall, the findings unequivocally show that adding MP to white

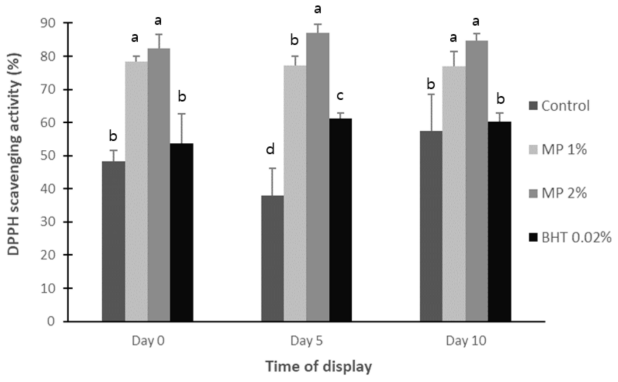


Figure 1. Mean values of DPPH radical scavenging activity (%) of white pork sausage (Moo Yor) supplemented with mulberry powder during refrigerated storage. Error bars indicate standard deviations. Different superscript letters (a-d) within the same storage day denote significant differences ($p<0.05$). MP, mulberry powder; BHT, butylated hydroxytoluene.

Table 1. Effects of mulberry (*Morus alb*a L.) powder on color of white pork sausage (Moo Yor) during refrigerated storage (mean ± standard deviation).

Item	Concentration of mulberry powder (%)			0.02% BHT	p
	0	1	2		
Day 0					
*L**	69.67±0.17^{a}	60.20±0.39^{b}	54.96±0.03^{c}	69.67±0.23^{a}	0.00
*a**	1.98±0.01^{c}	4.21±0.03^{b}	5.60±0.14^{a}	2.07±0.30^{c}	0.00
*b**	14.61±0.04	15.10±0.11	15.84±0.48	15.19±0.66	0.16
Day 5					
*L**	70.37±1.22^{a}	62.21±0.24^{b}	56.24±0.75^{c}	69.94±0.16^{a}	0.00
*a**	2.10±0.16d	3.65±0.14^{b}	5.41±0.05^{a}	2.56±0.07^{c}	0.00
*b**	15.03±0.03	14.61±0.53	15.17±0.40	14.87±0.31	0.35
Day 10					
*L**	71.25±1.33^{a}	64.60±2.09^{b}	58.62±0.43^{c}	70.85±1.05^{a}	0.00
*a**	1.80±0.19^{c}	3.00±0.43^{b}	4.68±0.19^{a}	1.86±0.41^{c}	0.00
*b**	13.99±0.49	14.44±1.52	13.16±0.47	13.98±0.25	0.37

BHT, butylated hydroxytoluene; L*, lightness; a*, redness; b*, yellowness. $^{a-c}$Within a row, mean values with different superscripts differ significantly at $p<0.05$.

Table 2. Effects of mulberry (*Morus alba* L.) powder on texture profile parameters of white pork sausage (Moo Yor) at day 0 of refrigerated storage (mean ± standard deviation).

Item	Concentration of mulberry powder (%)			0.02% BHT	p
	0	1	2		
Hardness (g)	426.00±60.95	304.50±153.29	294.20±119.43	375.80±61.53	0.09
Adhesiveness (mJ)	1.20±0.64	0.65±0.36	1.06±0.44	1.24±0.81	0.46
Springiness (mm)	19.25±10.90	10.71±5.42	10.08±1.18	19.46±12.90	0.22
Cohesiveness	0.27±0.21	0.12±0.09	0.08±0.06	0.42±0.40	0.16
Gumminess (g)	86.75±33.01	34.50±25.42	31.40±27.01	83.67±48.61	0.07
Chewiness (mJ)	15.30±11.85	3.55±3.07	9.43±6.78	9.43±5.25	0.08

BHT, butylated hydroxytoluene.

pork sausage during chilled storage successfully prevented lipid oxidation, and the antioxidative effect increased as mulberry concentration increased.

Total plate count

Changes in TPC of white pork sausage during refrigerated storage are shown in Figure 3. At day 0, all treatments exhibited low microbial counts. As storage progressed, TPC values gradually increased in all treatment groups; however, no significant differences were detected among treatments throughout the storage period ($p>0.05$).

Discussion

The inclusion of MP into white pork sausage had a notable effect on product quality during refrigerated storage. In terms of appearance, sausages supplemented with mulberry (1-2%) exhibited lower L* and higher a* compared to the control and BHT treatments. This increase in redness is mainly attributed to the presence of anthocyanins and other polyphenols in mulberry, which act as strong natural pigments (Khalifa *et al*., 2018). Mulberry extracts have been shown to considerably increase the redness and oxidative stability of Cantonese sausages and raw ground beef during refrigerated storage (Zhang *et al.*, 2016; Xiang *et al*., 2019). These findings confirm the dual role of mulberry as both a natural antioxidant and colorant, increasing the consumer appeal of meat products. Conversely, the addition of mulberry had no significant effect on the texture parameters of the sausages, including adhesiveness, springiness, cohesiveness, gumminess, and chewiness ($p>0.05$). Although small numerical differences in hardness were observed, these were not statistically significant. The lack of texture modification suggests that mulberry polyphenols at the tested levels do not interfere with protein gelation or water-binding properties of meat emulsions. This observation aligns with previous studies suggesting that, while phenolic compounds may interact with proteins (Cao *et al*., 2016), moderate inclusion levels of plant-derived antioxidants generally do not compromise textural quality of meat products (Naveena *et al*., 2008). Regarding antioxidant capacity, mulberry supplementation markedly enhanced radical scavenging activity as determined by the DPPH assay. Sausages containing mulberry, especially at 2%, showed significantly higher antioxidant capacity than the control and BHT groups. This effect can be explained by the presence of phenolic compounds and anthocyanins that effectively donate hydrogen atoms or electrons to neutralize free radicals. These findings are consistent with earlier findings demonstrating that mulberry extracts suppressed lipid and protein oxidation in meat products (Xu *et al*., 2018; Cheng *et al*., 2020). The DPPH method itself is widely accepted for evaluating radical scavenging activity in food systems (Kedare and Singh, 2011).

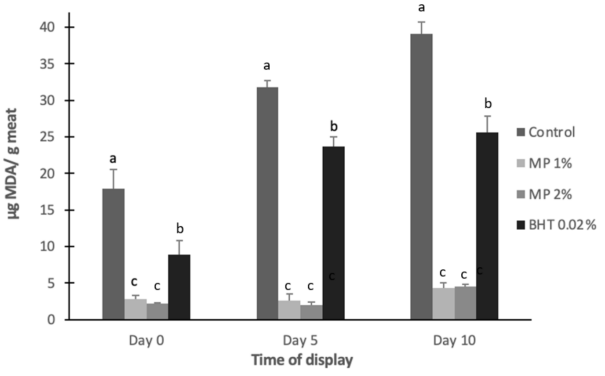


Figure 2. Mean values of lipid oxidation expressed as µg malondialdehyde per g of meat in white pork sausage (Moo Yor) supplemented with mulberry powder during refrigerated storage. Error bars indicate standard deviations. Different superscript letters (a-c) within the same storage day denote significant differences ($p<0.05$). MDA, malondialdehyde; MP, mulberry powder; BHT, butylated hydroxytoluene.

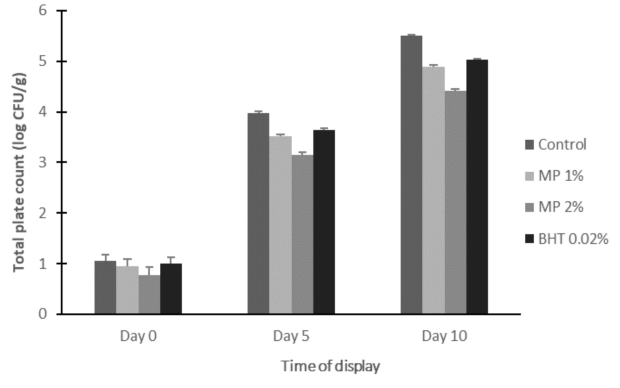


Figure 3. Mean values of total plate count (log CFU/g) of white pork sausage (Moo Yor) supplemented with mulberry powder during refrigerated storage. Error bars indicate standard deviations. MP, mulberry powder; BHT, butylated hydroxytoluene.

The TBARS analysis further confirmed the antioxidative effect of mulberry during cold storage. Sausages formulated with MP had significantly lower MDA values compared with control samples across all storage times. The antioxidative protection was dose-dependent, with 2% mulberry showing the strongest effect and, in some cases, performing better than BHT. This demonstrates the potential of mulberry to act as a natural substitute for synthetic antioxidants. These findings are in line with Xu *et al*. (2018) and Cheng *et al*. (2020), who reported that mulberry extracts effectively delayed lipid oxidation in pork products. Comparable effects of other plant-derived antioxidants such as pomegranate and grape extracts have also been reported in various meat systems (Ahn *et al*., 2002; Naveena *et al*., 2008).

The low TPC observed at day 0 in all treatments can be attributed to the steaming process, which effectively reduced the initial microbial load in white pork sausage (Jay *et al.*, 2005; ICMSF, 2011). During refrigerated storage, TPC gradually increased in all treatments, which is commonly observed in cooked meat products due to post-processing contamination and favorable conditions for microbial growth during chilled storage (Toldrá, 2006; Zhou *et al*., 2010).

No significant differences in TPC were detected among treatments throughout storage ($p>0.05$), indicating that MP supplementation did not exert a strong antimicrobial effect under the conditions tested. This finding may be explained by the fact that plant-derived phenolic compounds, while exhibiting antimicrobial activity in vitro, often show limited effectiveness in complex meat systems due to interactions with proteins and lipids (Burt, 2004; Gyawali and Ibrahim, 2014). Similar observations have been reported in studies where natural antioxidants improved oxidative

OPEN ACCESS

stability of cooked meat products without significantly altering microbial counts (Naveena *et al*., 2008).

Conclusions

Overall, this study demonstrated that MP is an effective natural additive for improving the quality and stability of white pork sausage. The addition of mulberry, particularly at 2%, enhanced product redness without negatively affecting texture, increased antioxidant activity, and significantly reduced lipid oxidation during refrigerated storage. These results suggest that mulberry can serve as a natural alternative to synthetic antioxidants such as BHT by enhancing oxidative stability and maintaining product quality during refrigerated storage. The findings also support the growing interest in using plant-based ingredients to meet consumer demand for healthier and clean-label food options.

References

Ahn J, Grün IU, Fernando LN, 2002. Antioxidant properties of natural plant extracts containing polyphenolic compounds in cooked ground beef. J Food Sci 67:1364-9.

Burt S, 2004. Essential oils: their antibacterial properties and potential applications in foods. Int J Food Microbiol 94:223-53.

Cao Y, True AD, Chen J, Xiong YL, 2016. Dual role (anti- and pro-oxidant) of gallic acid in mediating myofibrillar protein gelation and in vitro digestion. J Agric Food Chem 64:3054-61.

Cheng J, Xu L, Xiang R, Liu X, Zhu M, 2020. Effects of mulberry polyphenols on oxidation stability of sarcoplasmic and myofibrillar proteins in dried minced pork slices during processing and storage. Meat Sci 160:107973.

Gyawali R, Ibrahim SA, 2014. Natural products as antimicrobial agents. Food Control 46:412-29.

ICMSF, 2011. Microorganisms in foods 8. Use of data for assessing process control and product acceptance. Springer, New York, NY, USA.

Jay JM, Loessner MJ, Golden DA, 2005. Modern food microbiology. 7th ed. Springer, New York, NY, USA.

Kanner J, 1994. Oxidative processes in meat and meat products: Quality implications. Meat Sci 36:169-89.

Kedare SB, Singh RP, 2011. Genesis and development of DPPH method of antioxidant assay. J Food Sci Technol 48:412-22.

Khalifa I, Zhu W, Li K, Li C, 2018. Polyphenols of mulberry fruits as multifaceted compounds: Compositions, metabolism, health benefits, and stability - a structural review. J Funct Foods 40:28-43.

Klinjapo R, Krasaekoopt W, 2021. Application of microencapsulated bamboo leaf extract powder to control the rancidity in Moo Yor (Vietnamese-style sausage) during refrigerated storage. Appl Sci Eng Prog 14:13-8.

Natić M, Durović S, Papetti A, Fotirić Akšić M, Ognjanov V, Ljubojević M, Miljić U, 2015. Analysis and characterisation of phytochemicals in mulberry (Morus alba L.) fruits grown in Vojvodina, North Serbia. Food Chem 171:128-36.

Naveena BM, Sen AR, Vaithiyanathan S, Babji Y, Kondaiah N, 2008. Comparative efficacy of pomegranate juice, pomegranate rind powder extract and BHT as antioxidants in cooked chicken patties. Meat Sci 80:1304-8.

Saito M, Sakagami H, Fujisawa S, 2003. Cytotoxicity and apoptosis induction by butylated hydroxyanisole (BHA) and butylated hydroxytoluene (BHT). Anticancer Res 23:4693-701.

Tarladgis BG, Watts BM, Younathan MT, Dugan L Jr, 1960. A distillation method for the quantitative determination of malonaldehyde in rancid foods. J Am Oil Chem Soc 37:44-8.

Toldrá F, 2006. Lawrie's Meat Science. 7th ed. Woodhead Publishing, Cambridge, UK.

Tsanova-Savova S, Denev P, Ribarova F, 2018. Flavonoids in foods and their role in healthy nutrition. In: Holban AM, Grumezescu AM, eds. Handbook of food bioengineering: diet, microbiome and health. Academic Press, Cambridge, MA, USA, pp. 165-98.

Wang L, Guo H, Liu X, Jiang G, Li C, Li X, Li Y, 2019. Roles of Lentinula edodes as the pork lean meat replacer in production of sausage. Meat Sci 156:44-51.

Xiang R, Cheng J, Zhu M, Liu X, 2019. Effect of mulberry (Morus alba) polyphenols as antioxidant on physicochemical properties, oxidation and bio-safety in Cantonese sausages. LWT Food Sci Technol 116:108504.

Xu L, Zhu MJ, Liu XM, Cheng JR, 2018. Inhibitory effect of mulberry (Morus alba) polyphenol on the lipid and protein oxidation of dried minced pork slices during heat processing and storage. LWT Food Sci Technol 91:222-8.

Zhang X, Li D, Meng Q, He C, Ren L, 2016. Effect of mulberry leaf extracts on color, lipid oxidation, antioxidant enzyme activities and oxidative breakdown products of raw ground beef during refrigerated storage. J Food Qual 39:159-70.

Zhou GH, Xu XL, Liu Y, 2010. Preservation technologies for fresh meat – a review. Meat Sci 86:119-28.

Received: 4 January 2026; Accepted: 12 June 2026; Early view: 1 July 2026.

Contributions: Umaporn Pastsart: study concept and design. Chatchawan Chotimarkorn, Patima Permpoonpattana, Saneeta Salam: experiments, data analysis, and draft of the manuscript. Viboon Pongkanpai: data analysis and manuscript revision. All authors read and approved the final manuscript.

Conflict of interest: the authors declare that they have no competing interests.

Ethics approval and consent to participate: not applicable.

Availability of data and materials: the datasets are available from the corresponding author upon reasonable request.

Funding: this research was financially supported by Prince of Songkla University, Surat Thani Campus.

Declaration of generative artificial intelligence and artificial intelligence-assisted technologies in the writing process: language editing assistance was provided using an AI-assisted tool (ChatGPT, OpenAI). The authors take full responsibility for the content of the manuscript.

Publisher's note: all claims expressed in this article are solely those of the authors and do not necessarily represent those of their affiliated organizations, or those of the publisher, the editors and the reviewers. Any product that may be evaluated in this article or claim that may be made by its manufacturer is not guaranteed or endorsed by the publisher.

This work is licensed under a Creative Commons Attribution-NonCommercial 4.0 International License (CC BY-NC 4.0).

OPEN ACCESS