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Effects of partial replacement of nitrite with different fruit and vegetable powder on physicochemical and sensory aspects of fried beef meatballs

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Abstract

Fruit and vegetable powders can replace sodium nitrite in the meat-processing industry. However, basic information on the physicochemical and sensory aspects of fried beef meatballs is limited. In the present work, Chinese cabbage, celery, and cranberry powders were used to assess the effects of different addition levels and marinating times on the characteristics of fried beef meatballs. By combining 10 g/kg cranberry powder with 60 mg/kg NaNO₂, the highest a* value (13.34) and the lowest cooking loss (28.05%) were obtained. Results from low-field NMR indicated that the relaxation time T_{2b} in three fruit and vegetable powder-added groups was significantly lower than that of the control. T_{2b} values and the pH both exhibited a decreasing trend as the amount of fruit and vegetable powder additions increased. Concerning different marinating times by using cranberry powder on the properties of fried beef meatballs, marinating for 12 h displayed the best effect. The present work provided a potential solution for nitrite substitution in fried beef meatballs.

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Introduction

The colour of meat and meat products is essential in consumers' purchasing decisions (Xu et al., 2019). Nitrite is generally considered to be an essential additive in meat products. Nitrite contributes to colour formation by adding nitric oxide (NO) to the haem-iron of myoglobin, thus preventing myoglobin oxidation, and stabilising the characteristic red colour of meat products (Posthuma et al., 2018). It can also be used as a powerful antibacterial agent to protect against various foodborne microorganisms, notably Clostridium botulinum (Sindelar and Milkowski, 2012). Furthermore, nitrite effectively prevents oxidative rancidity, and imparts a unique flavour to the product (Ruiz-Capillas et al., 2015). Despite its numerous advantages, nitrite has raised concerns because it can be converted into nitrosating agents linked with forming N-nitrous compounds, particularly N-nitrosamines (NAs) (Dutra et al., 2017). NAs are a group of compounds well-known for their potential toxicity, carcinogenicity, and mutagenicity (Deng et al., 2021). As a result, nitrite

intake should be reduced as much as possible to reduce the risk of NAs formation, and thus, potential health risks.

Reducing nitrite intake can be met by substituting nitrite in meat products with fruit and vegetable powders. Many studies have replaced synthetic nitrite with vegetable powders with high nitrate content such as celery, Chinese cabbage, and spinach (Li et al., 2013). According to Sindelar et al. (2007), treatment combinations containing 0.2 - 0.4% celery powder were comparable to the 156 mg/kg sodium nitrite-added control for colour and lipid oxidation. Choi et al. (2020) reported that adding 0.25 and 0.35% Chinese cabbage effectively produced meat products with higher CIE a* values, and significantly lower residual nitrite contents. Jeong et al. (2020b) investigated that Chinese cabbage and radish powders can replace sodium nitrite in cured meat products. There have also been studies showing that cranberry powder can replace nitrite. The extracted cranberry powder was bright red, and polyphenolic contained abundant compounds. Cranberry powder contains various bioactive

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compounds and has antimicrobial effects against many pathogenic bacteria (Qiu and Wu, 2007; Guo *et al.*, 2020). Therefore, cranberry extract powder has been proposed as an additive to meat products to inhibit lipid changes, and avoid a decrease in muscle pigment (Stobnicka and Gniewosz, 2018; Tamkutė *et al.*, 2019).

Customers worldwide value beef because it is rich in high-quality proteins, minerals, and other highly digestible and absorbable nutrients (Duan et al., 2020). The resultant pleasing colour and unique flavour of fried beef products are the main attributes that attract consumers (Wang et al., 2018). Recently, lower sodium nitrite has been a major concern in fried beef meatballs due to the increasing demand for natural and safe products. In this regard, many vegetable extracts have been investigated to replace nitrite in meat processing (Ozaki et al., 2021). Therefore, the present work aimed to investigate the effects of different addition levels and marinating times of celery, Chinese cabbage, and cranberry powders as alternatives for reducing sodium nitrite addition on the quality attributes of fried beef meatballs.

Materials and methods

Materials

Celery, Chinese cabbage, and cranberry powders were purchased from Fufeng Sinuote Biotechnology Co. Ltd. (Shanxi, China). Food grade sodium nitrite (NaNO₂), fresh beef, soybean oil, salt, and starch were obtained from a local Chinese supermarket. All other chemicals and reagents used were of analytical grade.

Fired beef meatball manufacturing

Raw beef from cattle back legs was pre-treated to remove visible connective tissue and intermuscular fat. Beef was minced using a mincing machine after being cut into small pieces. Next, 10% starch and 20% available brine containing 1% salt were added and thoroughly mixed to aid the formation of meatballs. The formulation of the meatballs was modified according to Wang *et al.* (2023). The ground beef was allowed to stand for 10 min to achieve a certain level of adhesion before being shaped into a ball. Each meatball weighed approximately 10 g, and measured approximately 4 cm in diameter. Considering that the addition of 60 mg/kg NaNO₂ was the lowest limiting factor for

achieving the minimum bacteriostatic standards (Merino et al., 2016), 60 mg/kg NaNO2 was chosen to be added. According to previous reports, the addition levels of different fruit and vegetable powders have been adopted (Jeong et al., 2020a). Various amounts of Chinese cabbage powder (4 - 12 g/kg), celery powder (4 - 12 g/kg), or cranberry powder (6 - 14 g/kg) were combined and injected into the samples with needle injectors. Thus, in the present work, three treatment groups were set as Chinese cabbage powder + 60 mg/kg NaNO2 group, celery powder + 60 mg/kg NaNO2 group, and cranberry powder + 60 mg/kg NaNO₂ group, respectively. Various control groups were also set: no NaNO2 addition was regarded as the negative control (C1), 120 mg/kg NaNO2 addition was regarded as the positive control (C2) (Zarringhalami et al., 2009), 60 mg/kg NaNO2 addition was regarded as the control group (C3). Samples were marinated at 4°C for 12 h after injection. To investigate the effect of different marinating times by using cranberry powder combined with NaNO2 on physicochemical and sensory aspects of fried beef meatballs, a time interval of 2 - 16 h was set for evaluation.

After marination, the beef meatball samples were fried using commercial soybean oil in a deep fryer (AUX Group Co., LTD, Zhejiang, China) at 170°C for 3 min, as previously reported (Porto-Fett *et al.*, 2016; Kwon *et al.*, 2023). A thermometer was used to monitor the cooking temperature in real time. Following frying, all samples were allowed to cool to room temperature. For each group, at least 50 fried beef meatballs were prepared, and three samples were randomly selected from each group, and used for subsequent tests.

Colour

Colour values of fried samples were measured using a closed cone colorimeter (Minolta CR400®, Minolta Camera Co. Ltd., Osaka, Japan) (Riel $et\ al.$, 2017). L^* (lightness), a^* (redness), and b^* (yellowness) values were obtained using an instrument with illuminant D65, a measuring aperture of 8 mm diameter, and a standard observer of 10° . Before analysis, the instrument was calibrated using standard black and white tiles. Following the method described, the colour of the freshly cut interior of each fired sample was determined using the Commission International de l' Eclairage (CIE) L^* (lightness), a^* (redness), and b^* values (yellowness) system (Jo $et\ al.$, 2001).

Cooking loss

Cooking loss combines liquid and soluble matter excluded from the meatball during frying. Meat weight was measured before frying, and 5 min after frying to calculate the percentage of weight loss following a previously reported methods (Clausen and Ovesen, 2005).

pH

A homogeniser (Shanghai HUXI Industrial Co. Ltd., Shanghai, China) was used to homogenise approximately 2 g of fried beef meatball samples blended with 20 mL of distilled water for 1 min. The pH of the muscle homogenates was measured using a pH meter (Shanghai Kunquan Biotechnology Co. Ltd., China) following a previously reported method (Liu *et al.*, 2010). The pH meter probe was calibrated with standard pH buffers at room temperature before use.

Moisture content

Moisture content was calculated using the standard procedure outlined in the official AOAC methods (Elansari and Hobani, 2009). In brief, 2 g sample was dried in a 105°C drying oven for 2 h, and then weighed after cooling for 0.5 h. The procedure was repeated until the mass difference between the two-time intervals was no more than 2 mg.

Moisture mobility and muscle redistribution

A minispec mq20 NMR spectrometer (Bruker Optik GmbH, Ettlingen, Germany) was used to measure moisture mobility and redistribution in fried beef meatballs at 20 MHz frequency, and field strength of 0.47 T. Fried beef meatballs (2 g) were placed in 18 mm nuclear magnetic resonance (NMR) tubes. The tubes were then inserted into NMR at room temperature (20°C). Parameters of relaxation time (T_2) were 0.5 ms (time between 90° and 180° pulse), 1200 echoes at eight scan repetitions, and interval scans for 2 s at 25°C were set according to Han et al. (2019). The mean apparent relaxation time (T_2) and corresponding peak area (A₂) were recorded for each sample using the CONTIN algorithm normalisation of the raw data.

Residual nitrite

Sample preparation and nitrite determination were performed following the methods described in GB5009.33-2003, with slight modifications (Xu *et*

al., 2013). In brief, each sample (5 g) was prepared by adding ultra-pure water at 1:9 ratio, and homogenising for 3 min. Next, 10 mL homogenate was added to 50 g/L saturated borax solution, and heated for 15 min at 70°C. After cooling to room temperature, 106 g/L potassium ferrocyanide (5 mL) and 220 g/L zinc acetate (5 mL) were added to the solution to precipitate protein. The solution was filtered after 30 min. Taking 4 mL of filtrate and adding 1 mL of sulphanilamide and N-1-naphthyl ethylenediamine, and reacted for 20 min, the resulting red azo compound was photometrically determined at 538 nm using a TECAN Infinite® 200 PRO Full wavelength multifunctional microplate reader (Tecan Group Ltd., Switzerland).

Sensory analysis

The sensory test of the fried beef meatballs was performed according to Wang et al. (2018). The fried beef meatballs were cooled to room temperature before evaluation. The sensory test was conducted in a sensory evaluation laboratory (25°C) equipped with individual booths under white lighting. Twenty-three groups of fried beef meatball samples with different amounts of fruit and vegetable powders and marinating times were evaluated by 15 trained panellists (seven males and eight females aged 20 to 30 years). Each panel member was asked to rate the colour, odour, taste intensity, and overall acceptance. Approximately 10 min elapsed before evaluating the next samples. The panellists were asked to cleanse their oral palate before each sample with plain crackers and mineral water to eliminate the residual taste among samples. All testing samples were used at each evaluation, and repeated three times by the same panel (Deng et al., 2021). The following sensory characteristics were evaluated using a 1- to 7point rating scale (7-like extremely, 1-dislike extremely). The samples were served on white plates, and labelled with random numbers. The average score determined by each panel member was used to calculate the score for each sample (including each of the four attributes).

Statistical analysis

Three samples were analysed per group, and the measurements were repeated at least three times. The experimental data were analysed by using SPSS statistical program (SPSS, version 20.00 software, 2021) as a completely randomised design. One-way ANOVA was performed to assess the significance level of p < 0.05. The results were expressed as mean \pm standard error.

Results and discussion

Effect of different fruit and vegetable powders on physicochemical and sensory aspects of fried beef meatballs

Colour

L* values

Table 1 shows the changes in the colour parameters of lightness (L^*) , redness (a^*) , and yellowness (b^*) of fried beef meatballs at different fruit and vegetable powder concentrations. In general, the Chinese cabbage and cranberry powder groups had significantly lower L^* values than the positive control C3 (57.86), regardless of the addition level (p < 0.05). The L* value was highest (57.00) in the 4 g/kg celery powder and 60 mg/kg NaNO2 groups. However, Sindelar et al. (2007) reported that the addition of celery powder to sausage, regardless of the addition level, did not significantly change (p >0.05) the L^* values as compared to those from the sodium nitrite control (156 mg/kg). This could be because the sausages were cooked in impermeable casings without being fried. In the Chinese cabbage powder group, adding 8 g/kg yielded the lowest L^* value. And other Chinese cabbage powder groups had no differences in L^* values. These trends were inconsistent with the findings of Choi et al. (2020) who found no differences (p > 0.05) between the levels of white kimchi powder added to cooked ground pork products. The 10 g/kg cranberry powder treatment had the lowest L^* value (51.77) as compared to the other treatments (p < 0.05). The reason for the difference in the L^* values may be the discrepancy in moisture content. There was a substantial loss of moisture and water evaporation in the fried samples (Zhang et al., 2020; Choi et al., 2020).

a* values

The a^* value, which is characterised by red colour and colour stability, is the most sensitive parameter for colour measurement (Deng *et al.*, 2021). The a^* value of the unadded nitrite fried beef sample (C1) was found to be 6.17 after frying, while

the positive control of 120 mg/kg NaNO₂ group (C2) had significantly higher a^* values (12.64). For the only added 60 mg/kg NaNO₂ group sample (C3), a^* value of 10.40 was observed.

Adding celery powder at a rate of 8 g/kg resulted in the highest a^* value (12.68) among the celery powder groups. As the addition increased above 8 g/kg, the a^* values gradually decreased. The a* value was the highest in 6 g/kg Chinese cabbage powder (12.81). This result could be due to the inability of celery and Chinese cabbage powders to produce more nitrosochromogens metmyoglobin. As for the cranberry powder addition group, the highest a^* values were found with the 10 g/kg addition (13.34), which was significantly higher than that of control C2 with added 120 mg/kg NaNO₂ (12.64), and the group with added celery and Chinese cabbage powders.

The a^* values impact was probably due to the purple-red colour of the cranberry powder caused by its addition to fried beef. Cranberry powder is a bright red colour, but when mixed with beef, the colour of the anthocyanin pigments of cranberry partially shifts from red to blue, resulting in a purple-red colour in the meat (Sullivan et al., 2012; Xi et al., 2012). In general, the combination of nitric oxide and myoglobin (NO-Mb) causes redness in meat products. These findings could be attributed to more NO-haemachrome being formed progressively from NO-Mb during marinating with fruit and vegetable powders. Therefore, the results indicated that the cranberry powder effectively increased redness in fried beef meatballs, and could potentially substitute synthetic nitrite and reduce their amount.

b* values

There was no significant difference in b^* values between samples with celery and Chinese cabbage powders (p > 0.05). As compared to the added fruit and vegetable powders and NaNO₂, the control group (C1) had the highest b^* values (13.41). This result indicated that NaNO₂ and fruit and vegetable powders affected the surface colour of fried beef meatballs. These results may be attributed to the plant pigments found in the vegetable powders (Horsch *et al.*, 2014). This finding agreed with Krause *et al.* (2011) who discovered that hams with vegetable powder addition influenced the b^* values.

Table 1. Effects of different addition level of fruit and vegetable powder on CIE colour of fried beef meatballs.

_	Ingredie	ent addition			
Treatment	NaNO ₂ (mg/kg)	Natural ingredient (g/kg)	L^*	a*	<i>b</i> *
Control (-) C1			55.20 ± 0.59^{CD}	6.17 ± 0.15^{J}	13.41 ± 0.23^{A}
Control (+) C2	120		55.50 ± 0.54^{CD}	12.64 ± 0.10^{CD}	$11.45\pm0.21^{\rm HI}$
Control (+) C3	60		57.86 ± 0.44^{A}	10.40 ± 0.09^{I}	11.74 ± 0.33^{GH}
	60	4	57.00 ± 0.34^{AB}	$12.65\pm0.08~^{\mathrm{CD}}$	12.56 ± 0.09^{DE}
Celery powder +	60	6	56.43 ± 0.69^{BC}	11.76 ± 0.05^{GH}	11.71 ± 0.34^{GH}
$NaNO_2$	60	8	54.17 ± 0.12^{EF}	12.68 ± 0.24^{BC}	11.92 ± 0.35^{FG}
Group	60	10	55.68 ± 0.60^{CD}	12.04 ± 0.13^{FG}	11.73 ± 0.06^{GH}
	60	12	51.98 ± 0.89^{G}	11.50 ± 0.09^{HI}	11.15 ± 0.09^{H}
	60	4	52.94 ± 0.39^{G}	$12.26\pm0.31^{\mathrm{EF}}$	12.11 ± 0.18^{EF}
Chinese cabbage	60	6	52.61 ± 0.71^{FG}	12.81 ± 0.25^{AB}	12.54 ± 0.18^{DE}
$powder + NaNO_2$	60	8	51.66 ± 1.02^{G}	$12.50\pm0.38^{\mathrm{DE}}$	11.32 ± 0.10^{GH}
Group	60	10	$47.04 \pm 0.81^{\rm H}$	$11.15\pm0.32^{\mathrm{HI}}$	11.77 ± 0.21^{GH}
	60	12	53.19 ± 0.41^{FG}	12.75 ± 0.21^{AB}	12.70 ± 0.24^{CD}
	60	6	54.60 ± 0.19^{DE}	12.16 ± 0.12^{FG}	12.95 ± 0.15^{BC}
Cranberry	60	8	55.36 ± 0.29^{CD}	11.98 ± 0.03^{FG}	12.80 ± 0.11^{CD}
$powder + NaNO_2$	60	10	51.77 ± 0.02^{G}	13.34 ± 0.20^{A}	12.48 ± 0.10^{DE}
Group	60	12	55.63 ± 0.17^{CD}	$11.54\pm0.17^{\mathrm{HI}}$	12.29 ± 0.10^{EF}
-	60	14	54.82 ± 0.17^{DE}	11.92 ± 0.09^{FG}	12.60 ± 0.22^{DE}

Values are mean \pm standard error. Means followed by different uppercase superscripts in the same column for treatment are significantly different (p < 0.05). Control C1 (-): no sodium nitrite. Control (+) C2: added 120 mg/kg sodium nitrite. Control (+) C3: added 60 mg/kg sodium nitrite.

Cooking loss

Cooking loss is an important product economy evaluation index primarily attributed to water evaporation and fat loss during cooking. Table 2 summarises the cooking losses of all the fried beef meatball samples. Cooking loss was significantly lower for the products containing celery and Chinese cabbage powders than for the negative control (C1, 32.64%) and the nitrite-added control (C2, 31.83%; C3, 31.64%) (p<0.05). These results differed slightly from those of Choi *et al.* (2020) who found that cured products containing celery powder had a higher cooking loss than the control. However, Lee *et al.* (2008) found that incorporating kimchi powder at 1-2% into sausages resulted in lower cooking loss than

the control without kimchi powder. The 10 g/kg cranberry powder had the lowest cooking loss, 28.05%, which significantly lower than the control (*p* < 0.05). Similar studies have found that vegetable powder contained dietary fibre, and adding soy fibre, plasma protein, and other fibres increased cooking yields in meat products (Anderson and Berry, 2000). Dietary fibres can be combined with other ingredients in meat products to increase cooking yields, and further improve the binding capacity of water and fat (Serdaroglu, 2006). Therefore, fibres from vegetable extract powder can improve the properties of fried beef meatballs. However, excessive fruit and vegetable powder decreased binding ability and increased cooking loss.

Table 2. Effects of different fruit and vegetable powder addition levels on cooking loss, moisture content, pH values, and residual nitrite of fried beef meatballs.

	Ingred	ient addition	_	B.# * 4		D '1 1
Treatment	NaNO ₂ (mg/kg)	Natural ingredient (g/kg)	Cooking loss (%)	Moisture contents (%)	рН	Residual nitrite (mg/kg)
Control (-) C1			32.64 ± 0.16^{B}	21.08 ± 0.08^{DE}	5.80 ± 0.00^{BC}	3.92 ± 0.44^{I}
Control (+) C2	120		31.83 ± 0.29^{BC}	19.09 ± 0.86^{FG}	5.80 ± 0.02^{BC}	20.27 ± 0.78^{A}
Control (+) C3	60		31.64 ± 1.59^{BC}	$23.75 \pm 2.50^{\text{CD}}$	5.86 ± 0.00^A	14.30 ± 0.21^{GH}
	60	4	27.59 ± 0.59^{G}	13.31 ± 0.86^{G}	5.81 ± 0.01^B	15.85 ± 0.08^{DE}
Celery powder +	60	6	28.98 ± 0.30^{FG}	16.55 ± 0.70^{FG}	5.78 ± 0.01^{CD}	15.25 ± 0.21^{FG}
$NaNO_2$	60	8	29.68 ± 0.45^{FG}	25.06 ± 0.47^{BC}	5.79 ± 0.00^{BC}	17.12 ± 0.44^{B}
Group	60	10	31.14 ± 0.22^{CD}	22.30 ± 1.49^{DE}	5.78 ± 0.00^{CD}	16.71 ± 0.14^{BC}
	60	12	30.05 ± 0.90^{EF}	19.65 ± 2.50^{EF}	5.75 ± 0.00^{E}	15.62 ± 0.35^{DE}
	60	4	29.80 ± 0.50^{EF}	23.44 ± 0.47^{DE}	5.79 ± 0.00^{BC}	19.64 ± 0.16^{A}
Chinese cabbage	60	6	29.32 ± 0.95^{FG}	21.70 ± 0.38^{DE}	5.79 ± 0.00^{BC}	19.99 ± 0.27^{A}
$powder + NaNO_2$	60	8	31.15 ± 0.41^{CD}	20.86 ± 0.54^{DE}	5.78 ± 0.00^{CD}	14.10 ± 0.09^{GH}
Group	60	10	29.19 ± 0.30^{FG}	23.65 ± 0.78^{CD}	5.77 ± 0.00^{CD}	$13.97 \pm 0.05^{\mathrm{H}}$
	60	12	30.43 ± 0.89^{EF}	20.80 ± 0.54^{DE}	5.75 ± 0.01^{E}	16.67 ± 0.06^{CD}
	60	6	31.09 ± 0.92^{DE}	27.09 ± 1.41^{AB}	5.81 ± 0.01^B	15.83 ± 0.26^{DE}
Cranberry powder +	60	8	30.98 ± 0.26^{DE}	22.10 ± 1.62^{DE}	5.78 ± 0.00^{CD}	19.68 ± 0.22^{A}
$NaNO_2$	60	10	28.05 ± 0.58^{FG}	25.07 ± 0.75^{BC}	5.79 ± 0.00^{BC}	16.69 ± 0.56^{CD}
Group	60	12	$31.25 \pm 0.69^{\text{CD}}$	19.99 ± 1.30^{EF}	5.78 ± 0.00^{CD}	15.49 ± 0.85^{EF}
	60	14	34.09 ± 0.35^{A}	21.07 ± 0.87^{DE}	5.75 ± 0.00^E	15.94 ± 0.23^{DE}

Values are mean \pm standard error. Means followed by different uppercase superscripts in the same column for treatment are significantly different (p < 0.05). Control C1 (-): no sodium nitrite. Control (+) C2: added 120 mg/kg sodium nitrite. Control (+) C3: added 60 mg/kg sodium nitrite.

Moisture content

The moisture content of the samples was determined to provide information on water loss while observing the effect of fruit and vegetable powders on fried beef meatballs. Table 2 shows the changes in the moisture content of all samples. The celery powder group samples had a water content of 25.06% when tested at 8 g/kg, which was significantly higher than that control group C2 (19.09%). However, the Chinese cabbage powder groups demonstrated no significant differences in concentration. The moisture content of cranberry powder samples with additional amounts of 6 and 10 g/kg was much higher (27.09 and 25.07%) than the control groups C2 and C3 (23.75%). These changes could be attributed to the fact that cranberry powder

can affect the water-holding capacity of fried meat, thus resulting in a slight difference in moisture content between samples (Ozaki *et al.*, 2021). Our findings revealed that fried beef meatballs prepared with cranberry powder had higher moisture content. This result was consistent with the findings of Ham *et al.* (2016) who discovered higher weight loss in fermented dry sausages, thus indicating that fibres could retain water in their structure in cooked meat products.

pH

Fried beef meatballs with fruit and vegetable powders had lower pH values (p < 0.05) than the nitrite-added control C3 (5.86) (Table 2). Moreover, as the powder level increased, the pH decreased

further. These decreases in pH might be directly related to the fruit and vegetable powders because they had lower pH values. These findings were consistent with those of Jeong *et al.* (2020b) who reported that the pH values of cured products decreased with increasing concentrations of Chinese cabbage powder. The cranberry powder results were consistent with the findings of Xi *et al.* (2012) who found that adding cranberry powder with a pH of 2.2 can cause pH values in cured frankfurters to drop. Furthermore, Pennisi *et al.* (2020) discovered that a lower pH could also contribute to colour formation.

Residual nitrite

Several vegetables with high nitrate content, such as celery, spinach, and Swiss chard, are qualified as nitrate sources in powder or juice forms (Sebranek and Bacus, 2007; Sindelar *et al.*, 2007). Therefore, meat products generally showed lower residual nitrite levels than typical nitrite-cured products (Sindelar and Milkowski, 2012).

Residual nitrite concentrations were significantly different between the treatments, as shown in Table 2. As expected, the negative control treatment (C1) had little residual nitrite (3.92 mg/kg), and was significantly less than that of the other samples (p < 0.05). The initial residual nitrite in the control group may have been due to nitrogen compounds in the muscle matrix, which converted other forms of nitrogen into nitrite (Hospital et al., 2015). In the present work, the traditionally cured control, C2, had the highest residual nitrite concentrations (20.27)mg/kg). The demonstrated that the fruit and vegetable powder groups had lower residual nitrite contents than the positive control C2, but slightly higher residual nitrite contents than the control C3 (14.3 mg/kg). The result could be that nitrate from Chinese cabbage and celery reformation occurred during powder marination, and was reduced to nitrite (Sullivan et al., 2012).

The residual nitrite values increased and then decreased with increasing content of the same powder. As compared to the other samples from the cranberry powder groups, the 8 g/kg cranberry powder sample had the highest residual nitrite values (19.68 mg/kg). There was less residual nitrite in the meat system when the cranberry powder concentration was increased. This could be because the cranberry powder possessed inherent ascorbic acid which aided in converting nitrite to nitric oxide

(Li *et al.*, 2018). Sullivan *et al.* (2012) reported that ascorbic acid had a reductive action that can promote the formation of nitric oxide, which accelerated the reduction in residual nitrite content. However, lower levels of cranberry powder did not prevent elevated nitrite levels from binding to other protein components. Therefore, the residual nitrite content in the present work was associated with the level of natural ingredient powder added.

Water distribution

Low-field NMR (LF-NMR) has the potential to provide a precise evaluation of the water state of fried beef. Table 3 shows the relaxation time (T₂) of hydrogen protons and the corresponding peak area (A₂) for the three distinct water conditions. As a fast and non-destructive testing method, LF-NMR can analyse changes in water characteristics caused by thermal processing. By analysing the proton state, three components were connected to protein structures and the myofibrillar network (Han et al., 2019). T₂ represents the lateral relaxation time, which can be used to express various fried beef indicators. Not only T₂ can be distinguished from the distribution of free, crystal, bound, and immobile water, but it can also reflect the chemical exchange between free and hydration water. A₂ depicts the amount of water among different components (Pan et al., 2021).

Three components referred to as T_{2b} , T_{21} , and T₂₂ were found in all samples. A minor component, T_{2b}, between 0 - 10 ms, represents bound water, which is tightly associated with the hydrophilicity of macromolecules (Li et al., 2020). The relaxation time T_{2b} in the group added with fruit and vegetable powders was significantly lower than that of the control (C1, 0.93 ms; C2, 0.97 ms; C3, 1.00 ms) (p <0.05), reduced from approximately 1.00 to 0.77 ms, thus indicating that the tight water was stably combined with myofibre in the groups added with fruit and vegetable powders. The T_{2b} values decreased slightly as the addition of fruit and vegetable powders increased. This result indicated that the decrease in T_{2b} could be due to fruit and vegetable powders inducing a slight change in the protein structure, thereby strengthening the interaction between water and protein macromolecules (Zhang et al., 2020).

A significant component of T_{21} between 10 - 100 ms is called immobilised water, which is the dominant water component, and can be entrapped in the myofibrillar network or between thin and thick filaments. T_{22} , which ranged between 100 - 1000 ms,

Table 3. Relaxation times (including T_{2b}, T₂₁, and T₂₂) and the corresponding peak area amplitudes (including A_{2b}, A₂₁, and A₂₂) of fried beef meatballs under different levels of addition.

'	Ingredi	Ingredient addition		T_2			\mathbf{A}_2	
Treatment	NaNO2 (mg/kg)	Natural ingredient (g/kg)	T_{2b}	T_{21}	Γ_{22}	$\mathbf{A}_{2\mathrm{b}}$	A_{21}	A22
Control (-) C1			0.93 ± 0.09^{B}	$15.07\pm0.65^{\mathrm{BC}}$	$122.00 \pm 6.74^{\rm FG}$	3.91 ± 0.36^{BC}	$136.90 \pm 0.54^{\rm FG}$	$3220\pm114.99^{\text{CD}}$
Control (+) C2	120		$0.97\pm0.06^{\mathrm{AB}}$	$13.45 \pm 0.37^{\text{CD}}$	112.50 ± 2.73^{GH}	$2.46\pm0.38^{\rm E}$	179.10 ± 19.75^{DE}	2882 ± 23.75^{FG}
Control (+) C3	09		$1.00\pm0.06^{\mathrm{A}}$	$14.54 \pm 0.37^{\rm CD}$	$103.89 \pm 1.35^{\mathrm{GH}}$	$3.60\pm0.42^{\mathrm{CD}}$	$86.40 \pm 0.95^{\rm FG}$	3400 ± 55.96^{BC}
	09	4	$0.87\pm0.03^{\mathrm{BC}}$	15.77 ± 1.84^{AB}	$133.43 \pm 14.83^{\rm EF}$	4.36 ± 0.16^{BC}	166.85 ± 4.29^{DE}	3883 ± 97.85^{A}
Celery powder +	09	9	$0.90\pm0.06^{\mathrm{BC}}$	$12.72\pm0.72^{\mathrm{CD}}$	166.32 ± 6.07^{CD}	3.59 ± 0.34^{CD}	$75.54\pm3.33^{\rm G}$	3141 ± 47.85^{CD}
$NaNO_2$	09	~	$0.80\pm0.00^{\rm C}$	$16.26\pm0.62^{\mathrm{AB}}$	107.22 ± 4.72^{GH}	4.37 ± 0.17^{BC}	162.96 ± 11.92^{DE}	2691 ± 23.68^{FG}
Group	09	10	$0.80\pm0.00^{\rm C}$	$12.04\pm0.61^{\mathrm{CD}}$	150.11 ± 16.85^{DE}	4.16 ± 0.29^{CD}	190.96 ± 12.87^{DE}	3459 ± 223.2^{B}
	09	12	0.77 ± 0.03^{C}	11.06 ± 0.70^{D}	91.97 ± 18.20^{GH}	$4.01\pm0.09^{\mathrm{CD}}$	$151.72 \pm 0.50^{\rm EF}$	$3083 \pm 83.68^{\text{CD}}$
	09	4	$0.90\pm0.06^{\mathrm{BC}}$	$16.02\pm1.50^{\mathrm{AB}}$	$106.50\pm0.71^{\mathrm{GH}}$	3.84 ± 0.24^{BC}	220.83 ± 6.50^{BC}	$2901\pm187.8^{\rm EF}$
Chinese cabbage	09	9	$0.87 \pm 0.07^{\mathrm{BC}}$	$15.65\pm0.95^{\mathrm{BC}}$	100.50 ± 9.19^{GH}	3.71 ± 0.07^{BC}	216.30 ± 3.50^{CD}	2355 ± 32.72^{G}
$powder + NaNO_2$	09	8	$0.80\pm0.00^{\rm C}$	$13.30\pm1.50^{\mathrm{CD}}$	$85.50\pm4.95^{\mathrm{H}}$	3.79 ± 0.21^{BC}	176.65 ± 8.00^{DE}	$2462\pm67.16^{\rm G}$
Group	09	10	$0.80\pm0.00^{\rm C}$	$16.65\pm1.65^{\mathrm{A}}$	$104.00 \pm 11.31^{\rm GH}$	3.37 ± 0.21^{DE}	279.07 ± 4.22^{A}	2557 ± 77.84^{G}
	09	12	$0.87\pm0.07^{\mathrm{BC}}$	$12.67\pm1.52^{\mathrm{CD}}$	91.57 ± 7.31^{GH}	3.28 ± 0.24^{DE}	192.18 ± 11.86^{DE}	2470 ± 5.35^{G}
	09	9	$0.93\pm0.07^{\mathrm{AB}}$	$14.01\pm0.59^{\mathrm{CD}}$	172.68 ± 16.78^{CD}	3.75 ± 0.27^{BC}	201.87 ± 17.52^{DE}	$2514\pm91.93^{\rm G}$
Cranberry powder	09	8	$0.90\pm0.06^{\mathrm{BC}}$	$14.43\pm0.64^{\mathrm{CD}}$	207.96 ± 24.78^{A}	$3.63\pm0.18^{\mathrm{CD}}$	$243.11 \pm 20.00^{\mathrm{AB}}$	2958 ± 84.66^{DE}
+ NaNO2	09	10	$0.83\pm0.06^{\mathrm{BC}}$	$14.43\pm0.36^{\mathrm{CD}}$	149.32 ± 28.28^{DE}	$4.30\pm0.45^{\mathrm{BC}}$	174.56 ± 12.67^{DE}	$3025\pm59.83^{\rm DE}$
Group	09	12	$0.80\pm0.00^{\rm C}$	$13.53\pm0.09^{\mathrm{CD}}$	148.35 ± 17.93^{DE}	4.69 ± 0.46^{AB}	173.42 ± 9.05^{DE}	2999 ± 92.20^{DE}
	09	14	$0.80\pm0.00^{\rm C}$	12.87 ± 0.23^{CD}	191.38 ± 12.81^{AB}	$4.75\pm0.43^{\rm A}$	223.74 ± 6.46^{BC}	2571 ± 19.22^{FG}
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Values are mean \pm standard error. Means followed by different uppercase superscripts in the same column for treatment are significantly different (p < 0.05). Control C1 (-): no sodium nitrite. Control (+) C2: added 120 mg/kg sodium nitrite. Control (+) C3: added 60 mg/kg sodium nitrite.

represents free water, is defined as water that exists within the space between fibre bundles (Li *et al.*, 2020). There was no significant difference (p > 0.05) in the T_{21} of the fried beef meatballs as the addition of the cranberry powder increased.

There was no significant trend in A_{2b} of the fried beef meatball samples, thus indicating that it had little effect on the bound water content. Pearce *et al.* (2011) reported similar results where the bound water content in meat did not change with microstructural changes.

 A_{21} values of the cranberry powder were significantly higher than those of control C3, whereas the A_{22} values were lower. The changes in A_{21} and A_{22} indicated that free water in the intercellular space migrated into immobilised water entrapped in the

myofibrillar network. Furthermore, cranberry powder accelerates water movement from looser structures to protein-dense networks (Deng *et al.*, 2021). These findings were consistent with Han *et al.* (2014) who reported that the samples had a higher water-holding capacity because free water was converted to immobilised water. This was the same as the trend change of moisture content in cranberry powder.

Sensory evaluation

The effects of fruit and vegetable powders in combination with NaNO₂ on the sensory attributes of fried beef meatballs, including colour, odour intensity, taste intensity, and overall acceptability, are displayed in Table 4.

Table 4. Effects of fruit and vegetable powder addition levels on sensory qualities of fried beef meatballs.

	Ingredient addition		- Sancary characteristics				
Treatment	NaNO ₂ (mg/kg)	Natural	Sensory characteristics				
1 reatment		ingredient	Colour	Odour	Tasty	Overall	
		(g/kg)		intensity	intensity	acceptability	
Control (-) C1			3.33 ± 0.33^{E}	4.67 ± 0.67	3.67 ± 0.33	3.68 ± 0.33^{B}	
Control (+) C2	120		5.33 ± 0.33^{AB}	4.67 ± 0.57	4.33 ± 0.89	6.33 ± 0.33^{A}	
Control (+) C3	60		4.67 ± 0.33^{CD}	5.00 ± 0.67	3.67 ± 0.33	5.33 ± 0.33^{AB}	
	60	4	4.33 ± 0.33^{CD}	4.33 ± 0.33	4.00 ± 0.33	5.00 ± 0.58^{AB}	
	60	6	4.00 ± 0.00^{DE}	4.67 ± 0.33	4.33 ± 0.33	4.67 ± 0.33^{AB}	
Celery powder + NaNO ₂	60	8	4.33 ± 0.33^{CD}	4.33 ± 0.33	4.33 ± 0.33	5.33 ± 0.33^{AB}	
Group	60	10	4.33 ± 0.33^{CD}	4.67 ± 0.33	4.33 ± 0.33	5.00 ± 0.58^{AB}	
	60	12	4.33 ± 0.33^{CD}	4.33 ± 0.33	4.67 ± 0.33	5.00 ± 0.58^{AB}	
	60	4	4.33 ± 0.33^{CD}	4.67 ± 0.33	4.67 ± 0.33	$4.00\pm0.58^{\mathrm{B}}$	
Chinese cabbage powder +	- 60	6	4.33 ± 0.33^{CD}	4.33 ± 0.33	4.00 ± 0.58	5.00 ± 0.58^{AB}	
$NaNO_2$	60	8	4.33 ± 0.33^{CD}	4.67 ± 0.33	4.33 ± 0.67	3.67 ± 0.67^{B}	
Group	60	10	4.33 ± 0.33^{CD}	5.00 ± 0.58	4.33 ± 0.67	5.00 ± 0.58^{AB}	
	60	12	4.33 ± 0.33^{CD}	4.67 ± 0.33	4.67 ± 0.33	$4.00\pm0.58^{\mathrm{B}}$	
	60	6	5.00 ± 0.00^{BC}	5.00 ± 0.58	5.00 ± 0.58	4.67 ± 0.58^{AB}	
Cranberry powder +	60	8	5.00 ± 0.00^{BC}	5.00 ± 0.58	4.67 ± 0.89	$4.33\pm0.33^{\mathrm{B}}$	
$NaNO_2$	60	10	$5.67\pm0.33^{\mathrm{A}}$	5.00 ± 0.58	5.00 ± 0.58	5.00 ± 0.41^{AB}	
Group	60	12	5.00 ± 0.00^{BC}	4.68 ± 0.33	5.00 ± 0.58	5.00 ± 0.58^{AB}	
	60	14	5.00 ± 0.00^{BC}	4.67 ± 0.33	5.00 ± 0.58	5.33 ± 0.58^{AB}	

Values are mean \pm standard error. Means followed by different uppercase superscripts in the same column for treatment are significantly different (p < 0.05). Control C1 (-): no sodium nitrite. Control (+) C2: added 120 mg/kg sodium nitrite. Control (+) C3: added 60 mg/kg sodium nitrite.

The results showed that the colour of sample with $10\,$ g/kg cranberry powder $(5.67)\,$ was significantly higher than the other samples (p < 0.05), which could be attributed to the presence of nitrite, and the production of nitrous myoglobin. Similar findings on the sensory characteristics of frankfurter-type sausage were also found in previous studies (Xi et al., 2012). There was no significant difference in odour and taste intensity among all samples for various fruit and vegetable powders. The overall acceptance of the groups had no difference. However, the pleasing colour is one of the main factors that attract consumers (Huang et al., 2014).

Influence of different marinating times on physicochemical and sensory aspects of fried beef meatballs with the addition of cranberry powder

The present work found that 10 g/kg cranberry powder and 60 mg/kg NaNO₂ were the best options for partial nitrite substitution because this group had the highest a^* values, highest colour sensory characteristics, and lowest residual nitrite content. In the following studies, the effect of different marinating times on the physicochemical and sensory aspects of fried beef meatballs with the addition of cranberry powder was further characterised.

Colour

The colour developed during the marinating period (from 2 to 16 h) was due to loss of water and pigment formation. As depicted in Figure 1a, L^* values of samples incubated for 2 h (46.77) were lower than those of the other treatments (p < 0.05). L^* values were similar for samples incubated for 4 - 16 h. These results were consistent with those of Sindelar $et\ al.\ (2007)$ who found that different marinating times did not affect L^* values.

The significant differences (p < 0.05) observed in different marinating times on a^* values of the fried beef meatballs are illustrated in Figure 1a. The samples had the highest a^* value (13.34) after 12 h due to the formation of nitrosyl myoglobin. The addition of cranberry powder and nitrite influenced the colour of fried beef meatballs over time, such that more NO-haemachromes progressively formed from NO-Mb as marinating time increased. These results contradicted those of Choi et al. (2020) who found that white kimchi powder, combined with different marinating times, did not affect a^* values. However, no differences in CIE b^* values were found between the fried beef meatballs, thus indicating that incubation time did not affect b* values. A similar effect was observed by Bae et al. (2020) who

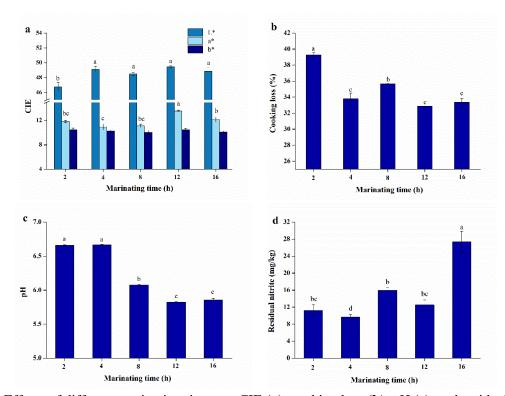


Figure 1. Effects of different marinating times on CIE (a), cooking loss (b), pH (c), and residual nitrite (d) of fried beef meatballs. Values are mean \pm standard error. Means followed by different lowercase superscripts are significantly different (p < 0.05).

demonstrated that the incubation time did not affect the b* values of cooked sausages treated with reddish powder.

Cooking loss and pH

The significant differences (p < 0.05) observed in different marinating times in the cooking loss of fried beef meatballs are shown in Figure 1b. The cooking loss was the lowest in the marinating time of 12 - 16 h. The results may be attributed to water evaporation during the marinating process. Khan *et al.* (2021) studies showed the same result that the minimum weight loss of meat was achieved with the longest marination time. However, our results disagreed with Bae *et al.* (2020) who reported that incubation time did not affect cooking yield.

In the present work, the pH values of fried beef meatballs decreased significantly from pH 6.66 at 2 h to pH 5.82 at 12 h (p < 0.05), as illustrated in Figure 1c. The pH reduction was probably caused by the cranberry being acidic and gradually infiltrating the samples during the marinating time. Similar studies described that samples with 2% cranberry extract had lower pH than samples without extract. Lower pH can affect protein coagulation, contribute to colour formation, and help stabilise the product (Tamkutė et al., 2019; Pennisi et al., 2020).

Residual nitrite

The residual nitrite content of the samples after different marinating times is displayed in Figure 1d. During the entire marinating period, marinating for 4 h resulted in the lowest levels of residual nitrite (9.7 mg/kg) (p < 0.05), whereas marinating for 16 h resulted in the highest value (27.33 mg/kg). However, the increase in residual nitrite might have been due to reduction reactions, nitrite oxidation, and nitrite binding to proteins, lipids, and other meat components. Cranberry powder has a reducing effect, reducing nitrite to NO gas. When time is prolonged, nitrous acid or nitric oxide (NO) in the matrix is transformed into nitrite, which has a greater nitrite concentration. It was previously known that 10 - 40% of nitrite is oxidised to nitrate during the curing process (Honikel, 2008). Overall, the residual nitrite at 12 h was relatively low.

Sensory analysis

Based on the sensory evaluation results (Table 5), it was clear that colour had the highest value (5.00) (p < 0.05) at 12 h, and taste intensity had a higher value at 4 - 16 h. The sensory perception of colour followed a pattern similar to the variations in a^* . The taste intensity score of fried beef meatballs increased as the marinating time increased. The tasty intensity increased from 3.23 to 4.68. However, there was no significant difference in odour throughout the This result agreed marinating period. Zarringhalami et al. (2009) who found no significant difference in the combined effect of annatto percentage and storage time for odour in the formulation containing 70% meat. Regarding overall acceptability, we found that there was no difference in marinating time.

Table 5. Effects of different marinating times on sensory qualities of fried beef meatballs with 10 g/kg cranberry powder and 60 mg/kg NaNO₂ addition.

	Sensory characteristic						
Marinating time	Colour	Odour intensity	Tasty intensity	Overall acceptability			
2	3.13 ± 0.33^{C}	3.21 ± 0.33	$3.23\pm0.33^{\mathrm{B}}$	$3.92\pm0.33^{\mathrm{A}}$			
4	3.75 ± 0.00^{BC}	3.80 ± 0.33	4.12 ± 0.33^{AB}	$3.92\pm0.33^{\mathrm{A}}$			
8	4.06 ± 0.33^{BC}	4.09 ± 0.33	$4.70\pm0.33^{\rm A}$	4.64 ± 0.33^{A}			
12	$5.00\pm0.33^{\mathrm{A}}$	4.68 ± 0.33	$5.00\pm0.33^{\mathrm{A}}$	$5.00\pm0.33^{\mathrm{A}}$			
16	4.38 ± 0.33^{AB}	4.38 ± 0.33	5.00 ± 0.33^{A}	4.64 ± 0.33^{A}			

Values are mean \pm standard error. Means followed by different uppercase superscripts in the same column for treatment are significantly different (p < 0.05). Control C1 (-): no sodium nitrite. Control (+) C2: added 120 mg/kg sodium nitrite. Control (+) C3: added 60 mg/kg sodium nitrite.

Conclusion

The present work found that a marination time of 12 h with 10 g/kg cranberry powder and 60 mg/kg NaNO₂ was the best option during fried beef meatball preparation. Based on this procedure, we not only reduced the addition level of nitrite, but we also observed that the cooking loss of fried beef meatballs was reduced, and higher a* values was acquired. Further research on the effects of other fruit and vegetable powder on fried beef meatballs is required to better understand their effects.

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