

COLOUR STABILITY AT THE END OF THE SHELF LIFE IN DRY-CURED PORK LOIN PRODUCED WITH AN ALTERNATIVE NATURAL NITRITE SOURCE

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ABSTRACT

This study aimed to evaluate the color stability of dry-cured pork loin made with a natural nitrite source (Swiss chard powder) combined with the starter culture BactoFerm Rosa. The assessment was done at the end of the product's shelf life, both, immediately after slicing and after exposure to daylight at $20 \pm 2^\circ\text{C}$ under daylight conditions for varying durations of 30, 60, 90, and 120 minutes. The research was conducted within industrial settings. Five groups of dry cured pork loins are examined, as follows: TSD (table salt and dextrose); NSD (nitrite salt and dextrose); NSDS (nitrite salt, dextrose and starter culture); TSDSF (table salt, dextrose, Swiss chard powder - from the first producer and starter culture) and TSDSS (table salt, dextrose, chard powder - from the second producer and starter culture). The measured L-value ranges from 26.10 (NSDS) to 33.42 (TSD). In the NSDS, the share of red color is the largest (11.05). The share of yellow color ranges from 1.79 (TSD) to 3.74 (TSDSF). The h-value ranges from 15.620 (NSDS) to 34.670 (TSD). The lowest C-value, i.e. the weakest color intensity, is observed in TSD (3.34), while NSDS has the highest color intensity (11.50). The incorporation of Swiss chard powder, acting as a natural source of nitrites, along with BactoFerm Rosa starter culture, contributed to maintaining stable and acceptable color in dry-cured pork loin as it approached the end of its shelf life.

Keywords: nitrate, starter culture, Swiss chard powder, synthetic nitrites.

INTRODUCTION

Throughout history, the creation of long-lasting cured meat products has been a practice dating back to ancient eras. This craft was rooted in accumulated wisdom and customs, transmitted from one generation of producers to the next. Over recent decades, notable strides in scientific understanding have played a crucial role in unraveling numerous biochemical intricacies inherent to the production procedure. This evolution has led to the refinement of the technological steps involved in crafting these products. The objective has been to introduce an exceptionally uniform product to the market that boasts distinct attributes such as a unique aroma, taste, and texture.

Numerous studies, within professional literature, highlight that the color of meat products stands as a pivotal factor when individuals make their selection. (Bendall & Swatland, 1988; Mancini & Hunt, 2005; Dragoev et al., 2014; Abdulhameed et al., 2016).

Pathare et al. (2013) state that the color of meat products serves as a significant indicator for assessing their freshness. Hammad et al. (2019) point out that microbiological and color changes in meat products are essential factors that affect the shelf life of the product itself. Meat processing manufacturers need to meet consumer expectations when it comes to the color

of the product (Crisosto et al., 2003). When these expectations are not fulfilled, consumers respond unfavorably to the product. Consumers anticipate a reddish-brown hue in meat and meat products, and if they encounter a dark, pale, or brown color, they invariably link it to diminished freshness, spoilage, and subpar quality.

Mancini & Hunt (2005) point out that the main carrier of color in meat is the pigment myoglobin, which is located in the sarcoplasm of muscle fibers. Feiner (2006) states that the color of fresh meat mainly depends on the representation of muscle pigment, primarily myoglobin (90 - 95 %), then hemoglobin (2 - 5 %) and small insignificant amounts of cytochromes, flavins, cobalamins, etc. Myoglobin, i.e. the monomeric globular protein, consists of colorless globin and heme, which is responsible for the color. The content of myoglobin in the muscles, and therefore the color of the meat, depends on a number of other factors such as: the type of animal, age, breed, the method of rearing and feeding, the activity, the type and chemical composition of the muscles, factors before and after slaughter (Danev, 1999; Mancini & Hunt, 2005).

In addition to myoglobin, the use of nitrites and nitrates is necessary to develop a typical, stable, red-pink color in meat products. (Andrée et al., 2010). They are added to meat products as a source of nitrogen monoxide and thereby ensure the creation of nitrosylmyoglobin. If nitrates are used, they must first be reduced to nitrites, accomplished through the activity of the bacterial enzyme nitrate reductase (Arnau et al., 2007; Gøtterup et al., 2008).

Nitrites and nitrates, beyond their role in color development within meat products, also serve as inhibitors of microorganism proliferation, notably targeting *Clostridium botulinum*. Additionally, they thwart the oxidation of fats and thus contribute to the development of flavor in meat products (Alahakoon et al., 2015; Govari & Pexara, 2015). For these reasons Ferysiuk & Wojciak (2020) state that nitrites and nitrates are irreplaceable additives in the processing industry.

Nitrites and nitrates can further react with secondary amines in meat products to form harmful N-nitrosamine compounds. N-nitrosamines are divided into volatile and nonvolatile N-nitrosamines (Xie et al., 2023). A number of epidemiological studies suggest that nitrosamines are etiological agents for the development of various types of cancer in humans. (Alexander et al., 2010; Chan et al., 2011; Johnson et al., 2013; Pham, et al., 2014; Zhu et al., 2015). Zhang et al. (2023) point out that the multiple function of nitrites in the production of meat products cannot be entirely substituted.

Indicated by the accumulated nitrates within vegetables, quantified as nitrate content (mg/kg FW), Santamaria (2005) divide vegetables into the following groups: vegetables that contain more than 2500 mg/kg FW, such as: celery, parsley, spinach, lettuce, arugula, etc.; vegetables that contain from 1000 to 2500 mg/kg FW, such as: Chinese cabbage, endive, leek, parsley, etc.; vegetables that contain from 500 to 1000 mg/kg FW, such as: beets, Savoy cabbage, cabbage, etc., vegetables that contain from 200 to 500 mg/kg FW, such as: carrots, cucumbers, zucchini, and broccoli, etc.; vegetables that contain less than 200 mg/kg FW, such as: potatoes, tomatoes, onions, mushrooms, asparagus, etc.

Nitrates, present in vegetables, are reduced to nitrites with the help of nitrate-reducing bacteria, represented in the starter cultures. (Choi et al., 2017; Shin et al., 2017; Hwang et al., 2018; Sucu & Turp, 2018; Ozaki et al., 2021).

As an alternative natural source of nitrites in meat products, vegetables containing high concentrations of nitrates can be used in combination with starter cultures, which contain nitrate-reducing bacteria Sebranek & Bacus (2007).

The primary objective of this study is to assess the color stability of dry-cured pork loin. This evaluation involves utilizing an alternative natural nitrate source along with the BactoFerm Rosa starter culture. The assessment is conducted at the end of the product's shelf

life, both in terms of freshly cut cross-sections and in situations where these sections are maintained at a consistent temperature of 20°C, with a maximum allowable temperature fluctuation of $\pm 2^\circ\text{C}$, under daylight conditions for varying durations of 30, 60, 90, and 120 minutes.

MATERIALS AND METHODS

Materials

The research was conducted in industrial conditions. In three iterations, the following five groups of dry-cured pork loin were produced as follows:

TSD-negative control group (95.5% frozen pork loin; 4% table salt and 0.5% dextrose).

NSD-positive control group (95.5% frozen pork loin; 4% nitrite salt and 0.5% dextrose).

NSDS group (95.48% frozen pork loin; 4% nitrite salt; 0.5% dextrose and 0.025% starter culture).

TSDSF group (95.23% frozen pork loin; 4% table salt; 0.5% dextrose 0.25% Swiss chard powder (first producer) and 0.025% starter culture).

TSDSS group (95.23% frozen pork loin; 4% table salt; 0.5% dextrose 0.25% Swiss chard powder (second producer) and 0.025% starter culture).

The frozen pork loin (*m. longissimus dorsi*) was thawed by dry defrosting. The fatty tissue and fascia were meticulously removed from the pieces, which were subsequently given a desired shape. Upon shaping, the process of tenderization commenced. A uniform application of dry curing was carefully spread over the surface of the pork loin pieces. These salted pieces were then placed on carts and left to mature for 21 days in a dark room. The ambient conditions during this period were maintained at a consistently cool temperature ranging from 0 to 4°C, accompanied by a relative air humidity ranging from 85% to 90%. Following this, the pork loin pieces were subjected to a draining process within an air-conditioning chamber for 24 hours, maintaining a temperature of 24°C and a relative humidity of 85%. This step was followed by a single day of cold smoking, where smoking occurred for approximately 40 minutes at four-hour intervals.

After the smoking phase, a span of 18 days was dedicated to ripening (fermentation). This stage initiated with a temperature of 22°C and a relative humidity of 82%. Subsequently, the temperature was gradually reduced to 12°C while the relative humidity decreased to 72%. The dry-cured pork loin pieces were vacuum-sealed in polyethylene bags and stored at a temperature of 4°C for a duration of six months, totaling 180 days.

Methods

Instrumental color analysis was conducted using the Dr. Lange spectro color chromometer. This device enabled the measurement of sample color within the fundamental X, Y, Z color system, employing coordinates Y, x, and y. Alternatively, measurements were carried out within the derived color system, with the L*, a*, and b* values being the most notable components of this system (CIE, 1976), which has the following values:

L - value describes the lightness of the sample: + L- lighter и - L- darker.

Values a and b describe the color hue: + a - more red (less green), - a - more green (less red), +b - more yellow (less blue) and - b - more blue (less yellow).

The Dr. Lange spectro color chromometer has an opening diameter of 8 mm and the color is measured on that surface. The measurement was made with illumination D-65 with a standard viewing angle of 8° . Before starting the measurement, the device is calibrated with a white standard (X= 4,09; Y= 4,34; Z= 4,69) and black standard (X= 83,68; Y= 88,41; Z= 90,42).

In the study, color analysis was performed subsequent to the expiration period of 180 days during which the products were stored at a temperature range of 0 to 4°C. The evaluation encompassed the measurement of color on a freshly cut cross-section at 0, 30, 60, 90, and 120

minutes. A total of fifteen concurrent measurements were carried out. The analysis was executed within an ambient temperature of 20°C, adhering to a maximum temperature deviation of $\pm 2^\circ\text{C}$ under natural daylight conditions.

Using the acquired L^* , a^* , b^* values, specific mathematical formulas were employed to compute two crucial color attributes: the hue angle (h) and color saturation (C). The hue angle (h) of a color characterizes the angle value on the a,b plane, moving in an anti-clockwise direction from the color point's position. This parameter denotes the direction in which the color's hue shifts. The hue angle (h) is determined using the following formula:

$$h = \tan^{-1} (b/a)$$

Color saturation (C) serves as an indicator of the intensity of color purity. It starts at 0 at the center of the coordinate system and escalates as the color moves farther away from the center towards the edges. The calculation of color saturation (C) is accomplished using the following formula:

$$C = \sqrt{a^2 + b^2}$$

RESULTS AND DISCUSSION

Table 1 shows the L-value measured at the end of the shelf life of the dry-cured pork loin. The lowest L-value, i.e. the darkest color of the fresh cross section, is observed in group NSDS, while the highest L-value, i.e. the lightest color of the fresh cross section, is observed in the negative control group TSD. It can be noticed that in dry-cured pork loin with added starter culture (NSDS, TSDSF and TSDSS groups), the fresh cross-section has a darker colour. The differences that appear between groups of dry-cured pork loin in terms of L-value are statistically significant ($p \leq 0.05$).

Table 1. Dynamics of L^* -values in dry-cured pork

Time of sampling	Groups of dry-cured pork loin				
	TSD	NSD	NSDS	TSDSF	TSDSS
	$\bar{x} \pm \text{SD}$				
0 min	$33.42 \pm 1.33^{\text{aB}}$	$33.45 \pm 1.47^{\text{aB}}$	$26.10 \pm 1.24^{\text{dBA}}$	$29.44 \pm 1.63^{\text{cBA}}$	$30.21 \pm 1.44^{\text{bA}}$
30 min	$32.97 \pm 1.31^{\text{aCB}}$	$33.00 \pm 1.45^{\text{aCB}}$	$25.84 \pm 1.23^{\text{cB}}$	$29.10 \pm 1.61^{\text{bBA}}$	$29.67 \pm 1.42^{\text{bBA}}$
60 min	$32.54 \pm 1.29^{\text{aC}}$	$32.54 \pm 1.43^{\text{aC}}$	$25.55 \pm 1.21^{\text{cB}}$	$28.57 \pm 1.58^{\text{bB}}$	$29.07 \pm 1.39^{\text{bB}}$
90 min	$33.22 \pm 1.32^{\text{aCB}}$	$33.31 \pm 1.46^{\text{aCB}}$	$26.00 \pm 1.23^{\text{cBA}}$	$29.21 \pm 1.62^{\text{bBA}}$	$29.77 \pm 1.42^{\text{bBA}}$
120 min	$36.75 \pm 1.46^{\text{aA}}$	$34.43 \pm 1.51^{\text{bA}}$	$26.53 \pm 1.26^{\text{dA}}$	$29.74 \pm 1.65^{\text{cA}}$	$30.37 \pm 1.45^{\text{cA}}$

$\bar{x}$ - mean value. SD standard deviation. statistically insignificant effect; means with a different letter (^{a-d}) within a row are statistically significantly different ($p \leq 0.05$; significance of group differences). means with a different letter (^{A-C}) within a column and parameter are statistically significantly different ($p \leq 0.05$; significance of sampling differences).

While maintaining the cross-section at a temperature of 20°C, with a maximum allowable temperature deviation of $\pm 2^\circ\text{C}$ under daylight conditions for durations of 30, 60, and 90 minutes, a consistent pattern is evident. Across all groups of dry-cured pork loin, there is a general reduction in the L-value. However, once the duration reaches 120 minutes, an inverse trend emerges, characterized by an increase in the L-value. Notably, these values span from 36.75 (TSD) to 26.53 (Group NSDS), (Table 1).

Differences in the L-value within the same groups over the average storage period at a room temperature of 20°C, with a maximum allowable temperature deviation of $\pm 2^\circ\text{C}$ under daylight conditions, are considered statistically significant ($p \leq 0.05$) (Table 1).

Anticipatedly, the lowest recorded a-value for a fresh cross-section, denoting the least amount of red color, is expected in Group TSD (2.68) due to its salting with table salt. Contrarily, the highest measured a-value is observed in Group NSDS (11.05), signifying that the inclusion of the starter culture significantly ($p \leq 0.05$) impacts the development of red color, as compared to the positive control Group NSD (Table 2).

Table 2. Dynamics of a*-values in dry-cured pork

Time of sampling	Groups of dry-cured pork loin				
	TSD	NSD	NSDS	TSDSF	TSDSS
	$\bar{x} \pm SD$				
0 min	2.68 ± 1.02^{eA}	9.27 ± 1.09^{bA}	11.05 ± 0.85^{aA}	7.47 ± 0.73^{cA}	6.89 ± 0.76^{dA}
30 min	2.65 ± 1.00^{eA}	9.15 ± 1.08^{bA}	10.93 ± 0.84^{aA}	7.34 ± 0.72^{cBA}	6.76 ± 0.75^{dBA}
60 min	2.60 ± 0.98^{eA}	9.02 ± 1.06^{bA}	10.80 ± 0.84^{aA}	7.20 ± 0.70^{cBA}	6.61 ± 0.73^{dBA}
90 min	2.51 ± 0.95^{eA}	8.73 ± 1.03^{bA}	10.61 ± 0.82^{aBA}	7.02 ± 0.69^{cCB}	6.41 ± 0.71^{dB}
120 min	2.31 ± 0.87^{eA}	8.13 ± 0.96^{bB}	10.25 ± 0.79^{aB}	6.69 ± 0.65^{cC}	6.03 ± 0.67^{dC}

$\bar{x}$ - mean value. SD standard deviation. statistically insignificant effect; means with a different letter (^{a-d}) within a row are statistically significantly different ($p \leq 0.05$; significance of group differences). means with a different letter (^{A-C}) within a column and parameter are statistically significantly different ($p \leq 0.05$; significance of sampling differences).

In the instances where Swiss chard powder was introduced along with the starter culture in the dry-cured pork loin groups, a notably lower a-value is statistically significant ($p \leq 0.05$) when contrasted with the positive control Group NSD and Group NSDS. Nonetheless, the development of red color remains acceptable in the nitrite-free TSDSF and TSDSS groups. Notably, the utilization of pre-converted nitrites derived from plant sources exhibits a more pronounced effect on the advancement of red color. This effect surpasses that of direct nitrite use. This observation is substantiated by information in professional literature, highlighting that employing pre-converted nitrites of plant origin contributes to a more substantial presence of red color, as indicated by the a-value, in meat products. (Hwang et al., 2018; Kim et al., 2019a; Kim et al., 2019b). Nevertheless, the prior conversion method, specifically the use of fermented cure containing nitrites, is prohibited for application in Europe. This stands in contrast to the regulations in the United States of America and Asia, where it is permissible for utilization in the production of meat products (Silovska Nikolova et al., 2022).

Table 2 provides evidence that while the cross-section is stored at a temperature of 20°C, with a maximum allowable temperature fluctuation of $\pm 2^\circ\text{C}$ under daylight conditions, a consistent decline in the a-value is evident across all groups of dry-cured pork loin. Kolev (2022) points out that air and light affect the reduction of the share of red color in meat products. The differences observed in the a-value among the groups for various storage durations of the cross-section at a room temperature of 20°C, with a maximum allowable temperature deviation of $\pm 2^\circ\text{C}$ under daylight conditions, are statistically significant ($p \leq 0.05$).

As evident from table 3, in the case of the fresh cross-section, the b-value varies from 1.79 (Group TSD) to 3.74 (Group TSDSF). The distinctions between the different groups of dry-cured pork loin are statistically significant ($p \leq 0.05$).

During the period when the cross-section is maintained at a room temperature of 20°C, with a maximum allowable temperature fluctuation of $\pm 2^\circ\text{C}$ under daylight conditions, there is a consistent decrease in the b-value across all groups of dry-cured pork loin. Furthermore, the differences in the measured b-value within the same groups for various durations of cross-

section storage at room temperature (20°C) with a maximum temperature deviation of $\pm 2^\circ\text{C}$ under daylight conditions are statistically significant ($p \leq 0.05$).

Table 3. Dynamics of b^* -values in dry-cured pork

Time of sampling	Groups of dry-cured pork loin				
	TSD	NSD	NSDS	TSDSF	TSDSS
	$\bar{x} \pm \text{SD}$				
0 min	$1.79 \pm 0.73^{\text{cA}}$	$3.01 \pm 1.04^{\text{bA}}$	$3.10 \pm 0.75^{\text{bA}}$	$3.74 \pm 1.23^{\text{aA}}$	$3.19 \pm 0.95^{\text{bA}}$
30 min	$1.77 \pm 0.72^{\text{cA}}$	$2.97 \pm 1.02^{\text{bA}}$	$3.06 \pm 0.75^{\text{bA}}$	$3.70 \pm 1.21^{\text{aA}}$	$3.14 \pm 0.93^{\text{bA}}$
60 min	$1.74 \pm 0.71^{\text{cA}}$	$2.93 \pm 1.01^{\text{bA}}$	$3.03 \pm 0.74^{\text{bA}}$	$3.63 \pm 1.19^{\text{aA}}$	$3.07 \pm 0.91^{\text{bA}}$
90 min	$1.72 \pm 0.70^{\text{cA}}$	$2.87 \pm 0.99^{\text{bA}}$	$2.98 \pm 0.72^{\text{bA}}$	$3.53 \pm 1.16^{\text{aA}}$	$3.00 \pm 0.89^{\text{bA}}$
120 min	$1.67 \pm 0.68^{\text{cA}}$	$2.79 \pm 0.96^{\text{bA}}$	$2.93 \pm 0.71^{\text{bA}}$	$3.46 \pm 1.14^{\text{aA}}$	$2.94 \pm 0.87^{\text{bA}}$

$\bar{x}$ - mean value. SD standard deviation. statistically insignificant effect; means with a different letter (^{a-d}) within a row are statistically significantly different ($p \leq 0.05$; significance of group differences). means with a different letter (^{A-C}) within a column and parameter are statistically significantly different ($p \leq 0.05$; significance of sampling differences).

The hue, represented by the h-value of the fresh cross-section, spans from 15,620 (Group NSDS) to 34,67 (Group TSD). Remarkably, Group NSDS displays the lowest h-value, indicating a proximity to the red (+a) color within the a,b plane. Conversely, Group TSD, where table salt is introduced, exhibits the highest h-value on the a,b plane, implying a tendency towards the yellow color (+b). Notably, the differences observed among the distinct groups are statistically significant ($p \leq 0.05$) (Table 4).

Throughout the storage of the cross-section at a temperature of 20°C, with a maximum allowable temperature deviation of $\pm 2^\circ\text{C}$ under daylight conditions, a consistent augmentation in the h-value is observable across all groups of dry-cured pork loin. Furthermore, the differences in the measured h-value within the same groups for diverse storage durations of the fresh cross-section at a temperature of 20°C with a maximum temperature deviation of $\pm 2^\circ\text{C}$ under daylight conditions are statistically significant ($p \leq 0.05$) (Table 4).

Table 4. Dynamics of h-values in dry-cured pork loin

Time of sampling	Groups of dry-cured pork loin				
	TSD	NSD	NSDS	TSDSF	TSDSS
	$\bar{x} \pm \text{SD}$				
0 min	$34.67 \pm 14.54^{\text{aA}}$	$17.98 \pm 6.06^{\text{cA}}$	$15.62 \pm 3.56^{\text{cA}}$	$26.21 \pm 7.50^{\text{bA}}$	$24.70 \pm 6.37^{\text{bA}}$
30 min	$34.70 \pm 14.54^{\text{aA}}$	$18.00 \pm 6.07^{\text{cA}}$	$15.64 \pm 3.56^{\text{cA}}$	$26.34 \pm 7.52^{\text{bA}}$	$24.74 \pm 6.38^{\text{bA}}$
60 min	$34.80 \pm 14.56^{\text{aA}}$	$18.00 \pm 6.07^{\text{cA}}$	$15.64 \pm 3.56^{\text{cA}}$	$26.39 \pm 7.53^{\text{bA}}$	$24.79 \pm 6.39^{\text{bA}}$
90 min	$35.34 \pm 14.65^{\text{aA}}$	$18.22 \pm 6.13^{\text{cA}}$	$15.64 \pm 3.56^{\text{cA}}$	$26.27 \pm 7.51^{\text{bA}}$	$24.91 \pm 6.41^{\text{bA}}$
120 min	$36.56 \pm 14.83^{\text{aA}}$	$18.92 \pm 6.33^{\text{cA}}$	$15.94 \pm 3.62^{\text{cA}}$	$26.94 \pm 7.64^{\text{bA}}$	$25.80 \pm 6.58^{\text{bA}}$

$\bar{x}$ - mean value. SD standard deviation. statistically insignificant effect; means with a different letter (^{a-d}) within a row are statistically significantly different ($p \leq 0.05$; significance of group differences). means with a different letter (^{A-C}) within a column and parameter are statistically significantly different ($p \leq 0.05$; significance of sampling differences).

The lowest C-value, indicating the least color intensity, is noted in Group TSD (3.34), in contrast to Group NSDS, which exhibits the highest color intensity for the fresh cross-section

at 11.50. The inclusion of the starter culture demonstrates a positive influence on color intensity. Notably, the observed disparity among the groups holds statistical significance ($p \leq 0.05$), (Table 5).

It is discernible that across all groups of dry-cured pork loin, storing the cross-section at a temperature of 20°C with a maximum allowable temperature fluctuation of $\pm 2^\circ\text{C}$ under daylight conditions leads to a decrease in the C-value and, subsequently, a reduction in the intensity of the color for the fresh cross-section. Moreover, the differences in the measured C-value within the same groups for varying storage durations of the fresh cross-section at a temperature of 20°C with a maximum temperature deviation of $\pm 2^\circ\text{C}$ under daylight conditions are statistically significant. ($p \leq 0.05$) (Table 5).

Table 5. Dynamics of C-values in dry-cured pork

Time of sampling	Groups of dry-cured pork loin				
	TSD	NSD	NSDS	TSDSF	TSDSS
	$\bar{x} \pm \text{SD}$				
0 min	$3.34 \pm 0.90^{\text{eA}}$	$9.80 \pm 1.11^{\text{bA}}$	$11.50 \pm 0.89^{\text{aA}}$	$8.42 \pm 0.91^{\text{cA}}$	$7.65 \pm 0.86^{\text{dA}}$
30 min	$3.29 \pm 0.89^{\text{eA}}$	$9.66 \pm 1.10^{\text{bA}}$	$11.37 \pm 0.88^{\text{aA}}$	$8.29 \pm 0.90^{\text{cBA}}$	$7.50 \pm 0.84^{\text{dBA}}$
60 min	$3.24 \pm 0.88^{\text{eA}}$	$9.53 \pm 1.08^{\text{bA}}$	$11.24 \pm 0.87^{\text{aA}}$	$8.13 \pm 0.89^{\text{cBA}}$	$7.33 \pm 0.82^{\text{dBA}}$
90 min	$3.15 \pm 0.85^{\text{eA}}$	$9.24 \pm 1.05^{\text{bA}}$	$11.04 \pm 0.86^{\text{aBA}}$	$7.92 \pm 0.86^{\text{cCB}}$	$7.12 \pm 0.80^{\text{dCB}}$
120 min	$2.95 \pm 0.78^{\text{eA}}$	$8.64 \pm 0.98^{\text{bB}}$	$10.68 \pm 0.83^{\text{aB}}$	$7.60 \pm 0.84^{\text{cC}}$	$6.75 \pm 0.76^{\text{dC}}$

$\bar{x}$ - mean value. SD standard deviation. statistically insignificant effect; means with a different letter (^{a-d}) within a row are statistically significantly different ($p \leq 0.05$; significance of group differences). means with a different letter (^{A-C}) within a column and parameter are statistically significantly different ($p \leq 0.05$; significance of sampling differences).

CONCLUSIONS

Drawing upon the findings derived from this research, several conclusions can be inferred:

The inclusion of the BactoFerm Rosa starter culture in Group NSDS of dry-cured pork loin leads to a notably stable color profile and a greater presence of red color, as indicated by the a-value, in comparison to the positive control Group NSD. The utilization of the BactoFerm Rosa starter culture exhibits a positive impact on color retention both up to the end of the fresh cross-section's shelf life and during the storage of the cross-section at room temperature under daylight conditions for durations of 30, 60, 90, and 120 minutes.

The nitrite-free groups of dry-cured pork loin, specifically Groups TSDSF and TSDSS, in combination with the BactoFerm Rosa starter culture, exhibit a satisfactory and consistent color stability extending up the fresh cross-section's shelf life. This color retention is, also, evident during the storage of the cross-section at room temperature under daylight conditions for durations of 30, 60, 90, and 120 minutes.

The use of Swiss chard powder, as the alternative natural choice of nitrites, in combination with the starter culture BactoFerm Rosa, will contribute to placing meat products on the market that will not contain synthetic nitrite.

Further research is needed on dry-cured pork loin produced with Swiss chard powder in combination with starter culture BactoFerm Rosa, which will focus on product safety. The integration of starter cultures within the meat industry holds the potential to enhance the availability of standardized meat products in the market.

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REFERENCES

- Abdulhameed, A. A., Yang, T. A., & Abdulkarim, A. A. (2016). Kinetic of texture and colour changes in chicken sausage during superheated steam cooking. *Polish Journal of Food and Nutrition Sciences*, 66(3), 199-209. <https://doi.org/10.1515/pjfn-2015-0044>.
- Alahakoon, A. U., Jayasena, D. D., Ramachandra, S., & Jo, C. (2015). Alternatives to nitrite in processed meat: Up to date. *Trends in Food Science and Technology*, 45(1), 37-49. <https://doi.org/10.1016/j.tifs.2015.05.008>.
- Alexander, D. D., Miller, A. J., Cushing, C. A., & Lowe, K. A. (2010). Processed meat and colorectal cancer: a quantitative review of prospective epidemiologic studies. *European Journal of Cancer Prevention*, 19(5), 328-341. <https://doi.org/10.1097/cej.0b013e32833b48fa>.
- Andrée, S., Jira, W., Schwind, K., Wagner, H., & Schwägele, F. (2010). Chemical safety of meat and meat products. *Meat Science*, 86(1), 38-48. <https://doi.org/10.1016/j.meatsci.2010.04.020>.
- Arnau, J., Serra, X., Comaposada, J., Gou, P., & Garriga, M. (2007). Technologies to shorten the drying period of dry-cured meat products. *Meat Science*, 77(1), 81-89. <https://doi.org/10.1016/j.meatsci.2007.03.015>.
- Bendall, J. R., & Swatland, H. J. (1988). A review of the relationships of pH with physical aspects of pork quality. *Meat Science*, 24(2), 85-126. [https://doi.org/10.1016/0309-1740\(88\)90052-6](https://doi.org/10.1016/0309-1740(88)90052-6).
- Chan, D. S. M., Lau, R., Aune, D., Vieira, R., Greenwood, D. C., Kampman, E., & Norat, T. (2011). Red and Processed Meat and Colorectal Cancer Incidence: Meta-Analysis of Prospective Studies. *PLOS ONE*, 6(6), e20456. <https://doi.org/10.1371/journal.pone.0020456>.
- Choi, Y., Kim, T., Jeon, K., Park, J., Kim, H., Hwang, K., & Kim, Y. (2017). Effects of Pre-Converted Nitrite from Red Beet and Ascorbic Acid on Quality Characteristics in Meat Emulsions. *Korean Journal for Food Science of Animal Resources*, 37(2), 288-296. <https://doi.org/10.5851/kosfa.2017.37.2.28>.
- CIE (1976). International Commission on Illumination, Colorimetry: Official Recommendation of the International Commission on Illumination Publication CIE No. (E-1.31). Bureau Central de la CIE, Paris, France.
- Crisosto, C. H., Crisosto, G. M., & Metheney, P. (2003). Consumer acceptance of ‘Brooks’ and ‘Bing’ cherries is mainly dependent on fruit SSC and visual skin color. *Postharvest Biology and Technology*, 28(1), 159-167. [https://doi.org/10.1016/S0925-5214\(02\)00173-4](https://doi.org/10.1016/S0925-5214(02)00173-4).
- Danev, M. (1999). Higijena i tehnologija na meso, riba, jajca i nivni proizvodi. [Hygiene and technology of meat, fish, eggs and their products] “Mikena”, Bitola.
- Dragoev, S.G., Staykov, A.S., Vassilev, K.P., Balev, D.K., Vlahova-Vangelova, D.B. (2014). Improvement of the Quality and the Shelf Life of the High Oxygen Modified Atmosphere Packaged Veal by Superficial Spraying with Dihydroquercetin Solution. *International Journal of Food Science* 2014:629062.
- Feiner G. (2006). *Meat products handbook - Practical science and technology*. Woodhead Publishing Limited, English.
- Ferysiuk, K., Wojciak, K.M., 2020. Reduction of nitrite in meat products through the application of various plant-based ingredients. *Antioxidants* 9 (8), 1-28. <https://doi.org/10.3390/antiox9080711>.

- Gøtterup, J., Olsen, K., Knøchel, S., Tjener, K., Stahnke, L. H., & Møller, J. F. (2008). Colour formation in fermented sausages by meat-associated staphylococci with different nitrite- and nitrate-reductase activities. *Meat Science*, 78(4), 492-501. <https://doi.org/10.1016/j.meatsci.2007.07.023>.
- Govari, M., & Pexara, A. (2015). Nitrates and Nitrites in meat products. *Journal of the Hellenic Veterinary Medical Society*, 66(3), 127. <https://doi.org/10.12681/jhvms.15856>.
- Hammad, H., Ma, M., Damaka, A., Elkhedir, A., & Jin, G. (2019). Effect of Freeze and Re-freeze on Chemical Composition of Beef and Poultry Meat at Storage Period 4.5 Months (SP4.5). *Journal of Food Processing & Technology*, 10, 791. <https://doi.org/10.4172/2157-7110.1000791>.
- Hwang, K., Kim, T., Kim, H., Seo, D., Kim, Y., Jeon, K., & Choi, Y. (2018). Effect of natural pre-converted nitrite sources on color development in raw and cooked pork sausage. *Asian-australasian Journal of Animal Sciences*, 31(8), 1358-1365. <https://doi.org/10.5713/ajas.17.0767>.
- Johnson, C.M., Wei, C., Ensor, J.E., Smolenski, D.J., Amos, C.I., Levin, B., Berry, D.A. (2013) Meta-analyses of colorectal cancer risk factors. *Cancer Causes Control* 24, 1207-1222.
- Kim, T., Hwang, K., Lee, M., Paik, H., Kim, Y., & Choi, Y. (2019a). Quality characteristics of pork loin cured with green nitrite source and some organic acids. *Meat Science*, 152, 141-145. <https://doi.org/10.1016/j.meatsci.2019.02.015>.
- Kim, T., Yong, H. I., Jang, H. W., Lee, H., Kim, Y., Jeon, K., & Choi, Y. (2019b). Quality of Sliced Cured Pork Loin with Spinach: Effect of Incubation Period with Starter Culture. *Journal of Food Quality*, 2019, 1-8. <https://doi.org/10.1155/2019/6373671>.
- Kolev, N. (2022). Natural antioxidants - an alternative for reduction of nitrites in cooked meat products. *Food Science and Applied Biotechnology*, 5(1), 64. <https://doi.org/10.30721/fsab2022.v5.i1.167>.
- Mancini, R., & Hunt, M. C. (2005). Current research in meat color. *Meat Science*, 71(1), 100-121. <https://doi.org/10.1016/j.meatsci.2005.03.003>.
- Ozaki, M. M., Munekata, P. E., Jacinto-Valderrama, R. A., Efraim, P., Pateiro, M., Lorenzo, J. M., & Pollonio, M. a. R. (2021). Beetroot and radish powders as natural nitrite source for fermented dry sausages. *Meat Science*, 171, 108275. <https://doi.org/10.1016/j.meatsci.2020.108275>.
- Pathare, P. B., Opara, U. L., & Al-Said, F. (2013). Colour measurement and analysis in fresh and processed foods: a review. *Food and Bioprocess Technology*, 6(1), 36-60. <https://doi.org/10.1007/s11947-012-0867-9>.
- Pham, N. M., Mizoue, T., Tanaka, K., Tsuji, I., Tamakoshi, A., Matsuo, K., Wakai, K., Nagata, C., Inoue, M., Tsugane, S., Sasazuki, S., Iwasaki, M., Otani, T., Sawada, N., Shimazu, T., Yamaji, T., Tsubono, Y., Nishino, Y., & Ito, H. (2014). Meat consumption and colorectal cancer risk: An evaluation based on a systematic review of epidemiologic evidence among the Japanese population. *Japanese Journal of Clinical Oncology*, 44(7), 641-650. <https://doi.org/10.1093/jjco/hyu061>.
- Santamaria, P. (2005). Nitrate in vegetables: toxicity, content, intake and EC regulation. *Journal of the Science of Food and Agriculture*, 86(1), 10-17. <https://doi.org/10.1002/jsfa.2351>.
- Sebranek, J. G., & Bacus, J. N. (2007). Cured meat products without direct addition of nitrate or nitrite: what are the issues? *Meat Science*, 77(1), 136-147. <https://doi.org/10.1016/j.meatsci.2007.03.025>.
- Shin, D., Hwang, K., Lee, C., Kim, T., Park, Y., & Han, S. G. (2017). Effect of Swiss Chard (*Beta vulgaris* var. *cicla*) as Nitrite Replacement on Color Stability and Shelf-Life of Cooked Pork Patties during Refrigerated Storage. *Korean Journal for Food Science of Animal Resources*, 37(3), 418-428. <https://doi.org/10.5851/kosfa.2017.37.3.418>.

Silovska Nikolova, A., Pejkovski, Z., Velkoska-Markovska, L., Belichovska, D., Nakov, D., Belichovska K. (2022). The effect of Swiss chard powder and starter cultures on colour development in smoked pork loin. *Journal of Agricultural, Food and Environmental Sciences*, 76, (2) 1-11.

Sucu, C., & Turp, G. (2018). The investigation of the use of beetroot powder in Turkish fermented beef sausage (sucuk) as nitrite alternative. *Meat Science*, 140, 158-166.

<https://doi.org/10.1016/j.meatsci.2018.03.012>.

Xie, Y., Geng, Y., Yao, J., Ji, J., Chen, F., Xiao, J., Hu, X., & Ma, L. (2023). N-nitrosamines in processed meats: Exposure, formation and mitigation strategies. *Journal of Agriculture and Food Research*, 13, 100645. <https://doi.org/10.1016/j.jafr.2023.100645>.

Zhang, Y., Zhang, Y., Jia, J., Peng, H., Qian, Q., Pan, Z., & Liu, D. (2023). Nitrite and nitrate in meat processing: Functions and alternatives. *Current Research in Food Science*, 6, 100470. <https://doi.org/10.1016/j.crfs.2023.100470>.

Zhu, B., Sun, Y., Qi, L., Zhong, R., & Miao, X. (2015). Dietary legume consumption reduces risk of colorectal cancer: evidence from a meta-analysis of cohort studies. *Scientific Reports*, 5, 8797. <https://doi.org/10.1038/srep08797>.