# Effect of acerola (*Malpighia emarginata* DC.) fruit extract on the quality of soft salami

## MARKÉTA SUCHOPÁROVÁ – ŠTĚPÁN JANOUD – LADISLAVA RÝDLOVÁ – FILIP BEŇO – VÁCLAV POHŮNEK – RUDOLF ŠEVČÍK

#### Summary

The demand for meat products without questionable food additives leads researchers to search for new ways to ensure the quality of the products. One of the possible approaches to achieve this goal is to use extracts from plants that have similar technological effects as commonly used additives. In this study, we examined the influence of the addition of the extract from acerola fruit on the key technological and organoleptic properties of soft salami. Samples of finely ground salami with various additions of acerola extract were produced in the laboratory of the Department of Food Preservation (University of Chemistry and Technology Prague, Prague, Czech Republic). The effect of acerola extract on the quality of soft salami was established through the measurement of the pH value, colour (using reflective spectrophotometry) and lipid oxidation (using thiobarbituric acid-reactive substances). The samples were also subjected to microbiological and sensory analysis. The addition of acerola slightly decreased the pH value and did not affect the microbial counts or sensorial quality of the salami. The incorporation of the extract had a positive effect on lipid stability and significantly improved the colour of the salami during storage. These results suggest that acerola fruit extract can be a suitable natural food additive for cooked meat products.

#### **Keywords**

natural food additive; meat product; soft salami; lipid oxidation; colour; sensorial property; technological property

In the production of meat products, various food additives and functional food ingredients are commonly added to raw meat. These food ingredients and additives ensure the characteristic properties of the product, such as texture, colour, taste or odour [1, 2]. Meat products of high quality contain the lowest amount of food additives as necessary. Food additives are not used to replace the meat or mask defects in the meat [2, 3]. The correct use of food additives is clearly defined by the legislation of the European Union [3], as well as their effects, dosage, composition and potential negative health consequences. However, the longterm attitude of the general public towards the use of food additives is negative [2, 4].

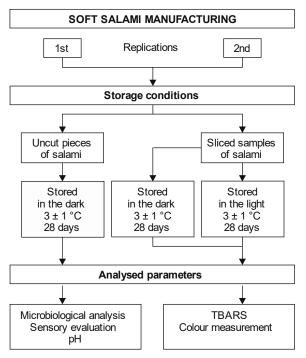
Thus, many studies have examined various ways to replace questionable artificial food additives without negatively affecting the overall quality of meat products. One of the possible approaches is to replace problematic food additives with natural compounds. However, there is no official general definition of natural compounds or natural food additives in the EU legislation. In the available literature, researchers use natural compounds from three possible sources [4, 5]. In most studies, natural compounds are obtained from plants [5-7]. In some studies, antimicrobial and other functional substances produced during microbial fermentation are rated among natural compounds [4, 5, 8]. And finally, some studies classify certain products of animal origin, such as the enzyme lysozyme, the proteins of albumen or the lactoperoxidase system of milk, as natural compounds [4, 5]. For the purposes of this study, natural substances of the plant origin will be discussed.

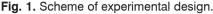
Natural compounds obtained from plants, similarly to commonly used additives, can have

Correspondence author: Markéta Suchopárová, e-mail: marketa.adamcova@vscht.cz

**Markéta Suchopárová, Štěpán Janoud, Ladislava Rýdlová, Filip Beňo, Václav Pohůnek, Rudolf Ševčík**, Department of Food Preservation, Faculty of Food and Biochemical Technology, University of Chemistry and Technology Prague, Technická 5, 166 28 Prague 6, Czech Republic.

a positive effect on the technological and sensory properties of meat products and, at the same time, are highly acceptable to consumers [4, 6]. The available studies focus mainly on natural substances with antimicrobial and antioxidant effects, as well as on the substances that can improve the colour of meat products [4, 6, 9]. Well-researched sources of natural compounds are, for example, various herbs (rosemary, thyme, oregano, sage) [10–12], spices (pepper, cinnamon, nutmeg) [6, 10, 11], fruits and vegetables (grapevine, grapefruit, cranberry, onion, garlic, capsicum, tomatoes) [6, 9, 12] and other plants such as aloe vera [13] and tea plant [14]. Natural compounds from the aforementioned sources are commonly added to meat products in the form of liquid or powder extracts [9]. Even though the natural compounds have a great potential for use in the production of meat products, several technological problems can be associated with their application. Natural compounds often have a strong taste and/or odour and, therefore, they may not be applicable to a wide range of meat products. Some compounds may not be stable during the processing and storage of meat products, while some may adversely affect the colour stability [4, 9, 10]. For this reason, new sources of natural compounds with a minimal negative impact on the overall quality of meat products are being sought out.





TBARS – thiobarbituric acid-reactive substances.

One of the less explored sources of natural compounds applicable in the production of meat products is acerola (Malpighia emarginata DC.) [15-18]. Acerola fruit is a rich natural source of ascorbic acid (10-45 g·kg<sup>-1</sup> of fruit) [15]. Furthermore, it contains other bioactive compounds such as carotenoids (for example  $\beta$ -carotene, β-cryptoxanthin, lutein), phenolic acids and flavonoids [15, 19]. Currently, acerola fruit is used mainly for direct consumption, in the beverage industry or in the production of food supplements [19, 20]. Even though acerola fruit is a wellknown source of bioactive compounds, there are only few studies on the effect of the addition of acerola on the quality of cooked meat products. In the studies by REALINI et al. [16] and FRUET et al. [17], the effects of acerola extract on the stability of beef patties were investigated. According to those studies, acerola extract had a positive effect on the oxidative and colour stability of meat products. A study of DE PAIVA et al. [18] examined the influence of acerola fruit powder on the stability of caiman meat nuggets during refrigerated storage. In that study, acerola had a positive effect on colour and did not affect the oxidative stability or sensorial quality of the nuggets. According to studies by SOUZA et al. [21] and ECA et al. [22], acerola fruit pulp or extracts can be incorporated into biodegradable packaging materials. Active substances contained on the surface of the package are capable of being gradually released to the surface of the food. This can possibly be used in the packaging of meat products for deceleration of lipid and colour oxidation.

Due to the limited available knowledge on the usage of acerola in meat production, there is an increasing need to explore the use of this fruit in a broader range of meat products. Therefore, we examined the possible positive effect of acerola extract on the quality of soft salami. Furthermore, we investigated the possibility of replacing both industrially produced ascorbic acid and sodium ascorbate with acerola extract. Acerola extract could have the same or a similar effect on the oxidative and colour stability of soft salami as the traditionally used antioxidants. In addition, acerola extract has the benefit of being more attractive to consumers.

## MATERIALS AND METHODS

#### **Experimental design**

To examine the effect of acerola on the quality of soft salami, we proceeded according to the following scheme (Fig. 1). The whole experiment was performed in two replications to verify the repeatability.

# Materials

Fresh meat (Tab. 1) was obtained twice (first and second replication) at two-month intervals from a commercial processing plant in the Czech Republic. The meat was stored at  $3 \pm 1$  °C for 1-2 days before manufacturing. Acerola extract (content of ascorbic acid 172 g·kg-1; total phenolics content expressed as gallic acid equivalent 167 g·kg<sup>-1</sup>) was obtained from Alchimica (Prague, Czech Republic). Ascorbic acid and sodium ascorbate were obtained from Sigma Aldrich (St. louis, Missouri, USA). Curing salt (content of sodium chloride 994 g·kg-1; content of sodium nitrite (E250) 6 g·kg<sup>-1</sup>) was obtained from Trumf International (Brno, Czech Republic). Liquid polyphosphate and spices were obtained from Hubka-Petrášek a vnuci (Prague, Czech Republic).

# Soft salami manufacturing

Samples of soft salami were prepared according to a traditional recipe (Tab. 1) in the laboratory of the Department of Food Preservation (University of Chemistry and Technology Prague, Prague, Czech Republic). Fresh meat was ground in Meat Grinder HLM G12SS (Hsiao Lin Machine, Taichung, Taiwan) by passing through a 4 mm plate. Immediately after, the ground meat, ice, liquid polyphosphate, curing salt and spices were mixed and cut using a bowl cutter CM 41 (Mainca, Barcelona, Spain). The resulting meat batter was divided into four batches. The first two batches were mixed with a standard amount of either ascorbic acid (290 mg·kg<sup>-1</sup>, control sample 1 - AI) or sodium ascorbate (290 mg·kg-1, control sample 2 – AII). The third and fourth batches were mixed with acerola extract powder at 3.5 g·kg<sup>-1</sup> and 5 g·kg<sup>-1</sup>, respectively.

The meat batters with ascorbic acid, sodium ascorbate or acerola extract were filled in polyamide bags (Viscase, Lombard, Illinois, USA). Each piece of salami was approximately 30 cm long and weighed approximately 400 g. Then, the samples were cooked in a convection oven 20 GN (Fagor Industrial, Oñati, Spain) for 10 min, obtaining a temperature of 70 °C in the centre of the product according to the Czech regulation 69/2016 [23]. The samples were let to cool down to room temperature  $(20 \pm 2 °C)$  and divided into several groups (Fig. 1). A half of the samples were left uncut and these whole pieces of salami were placed in a refrigerator at  $3 \pm 1 °C$  in the dark. The rest of the samples were aseptically unpacked and

Tab. 1. Composition of soft salami.

Raw materials	[%]
Beef shank	20.0
Beef round	10.0
Pork lean	33.0
Pork arm shoulder	20.0
Water	14.6
Curing salt	1.9
Liquid polyphosphate	0.4
Spices (white pepper, nutmeg)	0.1

sliced into 2 cm thick slices and vacuum-packaged (90% vacuum) in transparent polyethylenepolyamide bags (Wipak, Helsinki, Finland). Afterwards, half of these prepared samples were stored at  $3 \pm 1$  °C in the dark, while the other half were stored at  $3 \pm 1$  °C in the light (fluorescent light, approximately 700 lx). These conditions were intended to simulate different ways of storage of meat products in shops and their effects on colour stability and lipid oxidation during storage. The samples were stored for 28 days.

# Microbiological analysis

Samples of soft salami were analysed according to Czech Standards ČSN EN ISO 6887-1 [24] and ČSN EN ISO 6887-2 [25].

Ten grams of the soft salami from each sample were weighed aseptically into a sterile blender bag (VWR, Philadelphia, Pennsylvania, USA), then mixed with 90 ml of sterile physiological saline solution and homogenized in a laboratory blender Mixwel plus MIXW 1002 (Alliance Bio Expertise, Guipry, France) for 2 min. Then, 1 ml of the appropriate dilution was pipetted onto a sterile Petri dish and mixed with Plate Count Agar (PCA; Merck, Darmstadt, Germany). PCA plates were incubated at 30 °C for 72 h. Samples were analysed on day 1 after packaging and then after 14 and 28 days of storage.

# pH measurement

pH values were measured using pH meter Seven Go SG2-B with combined pH puncture electrode InLab Solids (Mettler Toledo, Columbus, Ohio, USA). Each sample was measured three times on the first day of storage.

# Lipid oxidation

Lipid oxidation in samples was determined by measuring 2-thiobarbituric acid-reactive substances (TBARS, method of TARLADGIS et al. [26]) on days 1, 7, 14, 21 and 28 of storage for both the samples stored in the dark and in the light. The results were expressed as milligrams of malondialdehyde (MDA) per kilogram of soft salami.

## **Colour measurement**

The colour of the soft salami was measured using a Minolta CM 5 reflection spectrophotometer (Konica Minolta, Tokyo, Japan). Colour measurements were carried out in the CIE-Lab space with 10° standard observer, illuminant D65 and an aperture diameter of 30 mm. Previously sliced samples were packed in transparent polyethylene-polyamide bags (Wipak, Helsinki, Finland). This enabled the instrument use Specular Component Included (SCI) mode to measure the true colour of the sample without the influence of surface conditions of the polymer foil. The measured values of lightness  $(L^*)$ , redness  $(a^*)$ and yellowness  $(b^*)$  were evaluated by the programs Spectra Magic NX (Konica Minolta) and Microsoft Excel (Office 2016, Microsoft, Redmond, Washington, USA). Results were reported as average of 18 consecutive measurements from random locations of each sliced sample (three pieces of salami from each sample, six measurements of colour for each piece). Colour measurements were carried out on days 1, 7, 14, 21 and 28 of storage.

#### Sensory analysis

In all experiments, samples of soft salami were presented to a semi-trained sensory panel of 15 researchers from Department of Food Preservation (University of Chemistry and Technology Prague). Samples were coded with four-digit random numbers and were presented to the assessors in random order. Equally big pieces of salami were served cold along with unsalted crackers and water. Panellists were asked to evaluate sample colour, intensity and acceptability of offflavours and atypical taste, intensity of salty taste and overall sensory quality. These qualities were evaluated with the use of 10 cm non-structured linear scoring scale ranging from 0 (not intense; unacceptable) to 10 (extremely intense; acceptable). The optimum for the acceptability of salty taste was represented by a value of 5. The samples of soft salami were evaluated on day 7 of storage.

#### Statistical analysis

The data obtained from the measurements were evaluated regarding statistical significance by one-way ANOVA using Statistica 13.1 (Dell, Round Rock, Texas, USA). Significance of the differences among results were analysed using Tukey's post hoc test (at P < 0.05).

## **RESULTS AND DISCUSSION**

#### **Microbiological stability**

The addition of acerola had no significant effect (P > 0.05) on the microbiological stability of the salami during storage (Tab. 2). Similar results on the minimal effect of the addition of acerola extract on the microbial stability of beef patties were reported by REALINI et al. [16].

# pH values

The addition of acerola had a significant effect (P < 0.05) on the decrease in pH of the salami (Tab. 3). This was probably caused by the high contents of malic, citric and tartaric acids in the acerola extract [19]. Generally, the decrease in pH can have a positive effect on the slowing down of microbial growth and a negative effect on the water-holding capacity in meat products in the pH range of 5–7 [5, 27]. However, the decrease in

Replication	Day	Total microbial counts [log CFU·g <sup>-1</sup> ]			
	of storage	AI	All	В	С
1st	1	1.90 ± 0.01 ª	1.86 ± 0.01 ª	$1.75 \pm 0.03  {}^{b}$	$1.84 \pm 0.02^{ab}$
	14	$2.93\pm0.03$	$2.98 \pm 0.04$	$2.86 \pm 0.05$	2.86 ±0.08
	28	$4.81 \pm 0.04$	$4.86 \pm 0.02$	$4.88 \pm 0.01$	$4.87 \pm 0.01$
2nd	1	$2.39 \pm 0.04$	$2.50 \pm 0.03$	$2.39 \pm 0.04$	$2.37 \pm 0.01$
	14	$3.87\pm0.02$	$3.96 \pm 0.02$	$3.88\pm0.05$	$3.93 \pm 0.04$
	28	$6.61 \pm 0.02  a$	$6.38 \pm 0.04$ <sup>b</sup>	$6.17 \pm 0.12^{b}$	$6.59 \pm 0.02 ^{a}$

Tab. 2. Effects of the addition of acerola extract on the total microbial counts of soft salami during storage.

The results are presented as mean  $\pm$  standard deviation. The means with different superscript letters (a, b) in the row are significantly different (P < 0.05).

Al – control sample with the addition of ascorbic acid, All – control sample with the addition of sodium ascorbate, B – sample with the addition of acerola extract 3.5 g kg<sup>-1</sup>, C – sample with the addition of acerola extract 5 g kg<sup>-1</sup>.

Suchopárová, M. et al.

Replication		р	Н	
	AI	All	В	С
1st	6.23 ± 0.01 ª	$6.23 \pm 0.01 ^{a}$	6.17 ± 0.01 <sup>b</sup>	$6.17 \pm 0.01$ <sup>b</sup>
2nd	6.28 ± 0.01 ª	$6.28 \pm 0.01 ^{a}$	6.21 ± 0.01 <sup>b</sup>	$6.21 \pm 0.02  ^{b}$

Tab. 3. Effects of the addition of acerola extract on pH of soft salami.

The results are presented as mean  $\pm$  standard deviation. The means with different superscript letters (a, b) in the row are significantly different (P < 0.05).

Al – control sample with the addition of ascorbic acid, All – control sample with the addition of sodium ascorbate, B – sample with the addition of acerola extract 3.5  $g \cdot kg^{-1}$ , C – sample with the addition of acerola extract 5  $g \cdot kg^{-1}$ .

Tab. 4. Effects of the addition of acerola extract on the thiobarbituric acid-reactive substances values
of soft salami during storage in the dark and in the light.

Replication	Day	TBARS [mg·kg <sup>-1</sup> ]				
	of storage	AI	All	В	С	
Samples store	ed in the dark	ζ				
1st	1	$0.075 \pm 0.006$ <sup>B</sup>	0.070 ± 0.011 <sup>B</sup>	$0.073 \pm 0.003$ <sup>B</sup>	$0.066 \pm 0.009^{\text{B}}$	
	7	$0.123 \pm 0.002 ^{\text{aA}}$	$0.119 \pm 0.004 ^{aA}$	0.081 ± 0.001 <sup>bB</sup>	$0.082 \pm 0.007  ^{bAB}$	
	14	$0.087 \pm 0.012^{\text{B}}$	$0.073 \pm 0.005^{\text{B}}$	0.070 ± 0.010 <sup>B</sup>	$0.089 \pm 0.005$ <sup>A</sup>	
	21	$0.065 \pm 0.003$ <sup>B</sup>	$0.074 \pm 0.012^{\text{B}}$	$0.087 \pm 0.003  {}^{ m B}$	$0.080 \pm 0.004  ^{\text{AB}}$	
	28	$0.130 \pm 0.006$ <sup>A</sup>	$0.124 \pm 0.012^{A}$	$0.108 \pm 0.010$ <sup>A</sup>	$0.105 \pm 0.006$ <sup>A</sup>	
2nd	1	$0.153 \pm 0.009$ <sup>D</sup>	0.144 ± 0.010 <sup>C</sup>	$0.139 \pm 0.002^{\text{ D}}$	$0.146 \pm 0.010^{\text{B}}$	
	7	$0.181 \pm 0.009  \text{aC}$	$0.170 \pm 0.007  ^{abBC}$	$0.158 \pm 0.002  ^{bCD}$	0.131 ± 0.003 cB	
	14	$0.234 \pm 0.009  \text{aB}$	0.199 ± 0.003 <sup>aB</sup>	$0.169 \pm 0.015  {}^{ m bC}$	$0.139 \pm 0.023  ^{bB}$	
	21	$0.151 \pm 0.006  dD$	$0.196 \pm 0.006  ^{bB}$	$0.216 \pm 0.004  ^{aB}$	0.172 ± 0.005 <sup>cB</sup>	
	28	$0.277 \pm 0.007  \text{aA}$	$0.291 \pm 0.022 ^{aA}$	$0.247 \pm 0.012  {}^{bA}$	$0.230 \pm 0.004  {}^{bA}$	
Samples stored in the light						
	1	$0.068 \pm 0.005$ <sup>D</sup>	0.077 ± 0.016 <sup>D</sup>	$0.070 \pm 0.007$ <sup>D</sup>	$0.079 \pm 0.005 ^{\text{C}}$	
1st	7	$0.314 \pm 0.013  \text{aC}$	$0.297 \pm 0.019  ^{aC}$	$0.200 \pm 0.029  {}^{ m bC}$	$0.183 \pm 0.027  {}^{\mathrm{bC}}$	
	14	$0.769 \pm 0.029  ^{aB}$	$0.621 \pm 0.061  ^{bB}$	$0.583 \pm 0.028  ^{bB}$	$0.434 \pm 0.032^{\text{cB}}$	
	21	0.671 ± 0.073 <sup>B</sup>	0.631 ± 0.044 <sup>B</sup>	0.519 ± 0.041 <sup>B</sup>	$0.528 \pm 0.056^{B}$	
	28	1.195 ± 0.030 <sup>aA</sup>	1.157 ± 0.033 <sup>aA</sup>	1.003 ± 0.015 <sup>bA</sup>	$0.911 \pm 0.038  {}^{\mathrm{bA}}$	
2nd	1	0.152 ± 0.010 <sup>D</sup>	0.138 ± 0.005 <sup>D</sup>	0.140 ± 0.012 <sup>E</sup>	$0.142 \pm 0.006 ^{\text{E}}$	
	7	0.720 ± 0.061 <sup>aC</sup>	$0.613 \pm 0.006  ^{aC}$	$0.470 \pm 0.049  {}^{ m bD}$	$0.363 \pm 0.032  ^{bD}$	
	14	$0.908 \pm 0.028  ^{aB}$	1.009 ± 0.061 <sup>aB</sup>	$0.873 \pm 0.088  {\rm aC}$	$0.648 \pm 0.013  {}^{ m bC}$	
	21	$1.979 \pm 0.032  ^{\mathrm{aA}}$	$1.814 \pm 0.062 ^{aA}$	1.061 ± 0.062 <sup>bB</sup>	1.143 ± 0.058 <sup>bB</sup>	
	28	$2.016 \pm 0.039$ <sup>aA</sup>	$1.928 \pm 0.084 ^{aA}$	1.638 ± 0.043 <sup>bA</sup>	1.397 ± 0.068 <sup>cA</sup>	

The results are presented as mean  $\pm$  SD. The means with different superscript capital letters in the column (A–E) or lowercase letters in the row (a–d) are significantly different (P < 0.05).

TBARS – thiobarbituric acid-reactive substances (expressed as milligrams of malondialdehyde per kilogram of soft salami). Al – control sample with the addition of ascorbic acid, All – control sample with the addition of sodium ascorbate, B – sample with the addition of acerola extract 3.5 g kg<sup>-1</sup>, C – sample with the addition of acerola extract 5 g kg<sup>-1</sup>.

pH in the range of 6.23–6.17 in the first replication and of 6.28–6.21 in the second replication was probably too low for these changes to appear.

## Thiobarbituric acid-reactive substances

TBARS values fluctuated during storage in the dark (Tab. 4). The fluctuations could be caused by the further reactions of aldehydes in the oxidized lipids or by the possible reactions of the secondary

oxidation products of lipids with proteins and other food components. Such reaction products were not detected by the TBARS measurement [28]. Despite the fluctuating nature of the measured values, all samples stored in the dark in both replications had significantly higher TBARS values (P < 0.05) at the end of storage.

TBARS values of the samples stored in the light grew significantly faster than of those stored

in the dark (Tab. 4). The faster oxidation of lipids in the light was probably supported by the oxidation of photosensitive heme pigments of heat-treated meat products nitrosylmyoglobin and its detaturated pink form nitrosylmyochrome [29-32]. The oxidation of these pigments in the light, in combination with the presence of oxygen even at low oxygen levels in the headspace of the package, leads to further oxidative reactions with metmyoglobin and hemin as main products [29, 31-33]. The formation of these compounds negatively affects the lipid stability and leads to oxidative rancidity, the development of off-flavours and product discoloration [29]. Oxidized flavour could be, according to CAMPO et al. [34], perceived by consumers in the range of MDA contents of  $0.5-1.0 \text{ mg}\cdot\text{kg}^{-1}$  in pork and  $0.6-2.0 \text{ mg}\cdot\text{kg}^{-1}$  in beef. Thus, it could be perceived on day 14 in the first replication and on Day 7 in the second replication. The rapid oxidation of the samples in the light also resulted in significant discolouration, which is discussed in the section Colour measurements.

Ascorbic acid and sodium ascorbate belong to the most important antioxidants used in heattreated meat products. During the manufacturing, they participate in the formation of the typical pink colour of the product by reducing metmyoglobin to myoglobin and nitrites to nitric oxide. Their addition can also decelerate the oxidation and discolouration of the meat products during storage [35, 36]. The replacement of ascorbic acid and sodium ascorbate with acerola extract positively affected the oxidation stability of the soft salami. When stored in the dark, the samples with the addition of acerola showed lower TBARS values (P < 0.05) than the control samples on Day 7 in the first replication and on Days 7, 14 and 28 in the second replication. On the other days of dark storage, there were no significant differences in TBARS values between the samples (P > 0.05). During the storage in light, the samples with the addition of acerola were statistically less oxidized (P < 0.05) on Days 7 and 28 in the first replication and on Days 7, 21 and 28 in the second replication. The reason for the improvement of the oxidative stability of salami with the addition of acerola, compared to the control samples with ascorbic acid and sodium ascorbate, could be the synergistic effect of ascorbic acid, carotenoids and phenolic compounds contained in acerola. These are, for example, lutein,  $\beta$ -carotene, quercetin and anthocyanins, which may have a positive effect on the oxidative stability of foods [19, 37]. Our results agree with the study of REALINI et al. [16] and FRUET et al. [17], where the oxidative stability of beef patties with acerola was better than that of the control samples. In contrast, the study of DE PAIVA et al. [18] did not show a significant effect of acerola on improving the oxidative stability of caiman nuggets. To the best of our knowledge, there are not many studies that deal with the effect of the replacement of ascorbic acid or sodium ascorbate by acerola extract on the oxidative stability of cooked meat products.

## **Colour measurements**

Replacement of ascorbic acid and sodium ascorbate by acerola extract had a significant effect on the instrumental colour of salami stored in the dark and in the light (P < 0.05). Samples with the acerola extract had a statistically lower lightness ( $L^*$ ), higher redness ( $a^*$ ) and higher yellowness ( $b^*$ ) at the beginning of storage (Fig. 2, Fig. 3). This agrees with the study of REALINI et al. [16] and FRUET et al. [17]. The lower lightness of the acerola-containing salami samples was related to higher redness and yellowness that was probably caused by the carotenoids and anthocyanins contained in the acerola extract [19, 38].

When stored in the dark, all samples of the soft salami became discoloured during the storage period (P < 0.05). However, samples with the addition of acerola extract were superior to the control samples because they maintained lower  $L^*$ , higher  $a^*$  and higher  $b^*$  values throughout the storage period (Fig. 2). Discolouration of the samples was probably caused by microbial growth, oxidation of lipids and/or oxidation of heme pigments [29, 39]. Storage in the light resulted in more significant discolouration in comparison to storage in the dark (Fig. 3). This could be related to the sensitivity of nitroxymyochrome and nitroxymyoglobin to light in the presence of oxygen and to the associated oxidative changes [29, 30]. In the second replication, these changes were more pronounced probably due to the more advanced lipid oxidation (Tab. 4) and/or due to the higher microbial counts (Tab. 2). All samples showed a significant decrease in redness and a significant increase in lightness and yellowness (P < 0.05). However, the addition of acerola had a significant positive effect on maintaining the colour of soft salami. Samples with acerola had significantly lower  $L^*$ and higher  $a^*$  values than the control samples throughout the storage period. Improved colour stability in the samples with acerola was probably caused by a combination of factors. The addition of acerola slowed down both the oxidation of lipids (Tab. 4) and the oxidation of heme pigments due to the synergistic effect of ascorbic acid, carotenoids and other phytochemicals contained in the extract [19, 37]. Furthermore, carotenoid pigments

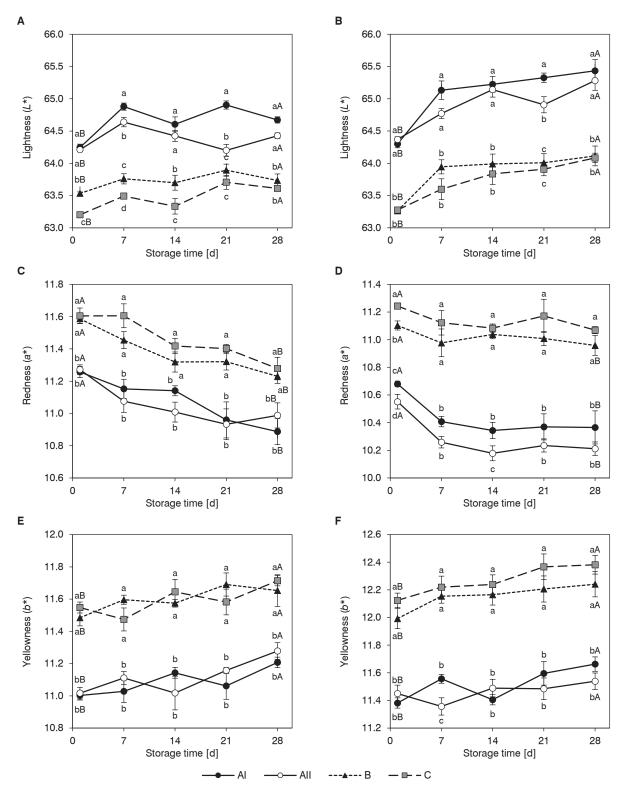


Fig. 2. Effects of the addition of acerola extract on the colour of soft salami during storage in the dark.

A – lightness during the first replication; B – lightness during the second replication; C – redness during the first replication; D – redness during the second replication; E – yellowness during the first replication; F – yellowness during the second replication. Markers represent the means and the error bars represent the standard deviations. The means with different superscript lowercase letters (a–d) are significantly different between the samples (P < 0.05) and different capital letters (A–B) indicate significant difference between the first and last day of storage (P < 0.05).

AI – control sample with the addition of ascorbic acid, AII – control sample with the addition of sodium ascorbate, B – sample with the addition of acerola extract 3.5 g kg<sup>-1</sup>.

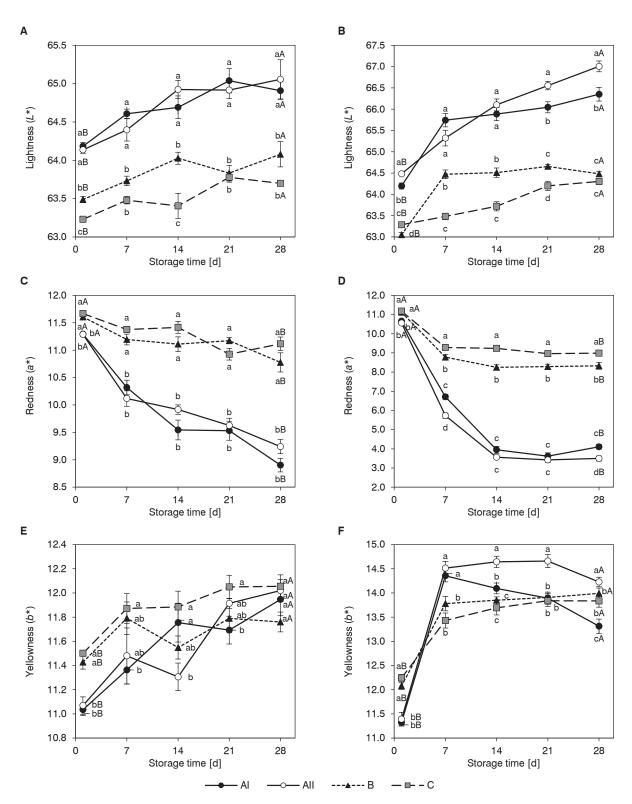


Fig. 3. Effects of the addition of acerola extract on the colour of soft salami during storage in the light.

A – lightness during the first replication; B – lightness during the second replication; C – redness during the first replication; D – redness during the second replication; E – yellowness during the first replication; F – yellowness during the second replication. Markers represent the means and the error bars represent the standard deviations. The means with different superscript lowercase letters (a–d) are significantly different between the samples (P < 0.05) and different capital letters (A–B) indicate significant difference between the first and last day of storage (P < 0.05).

AI – control sample with the addition of ascorbic acid, AII – control sample with the addition of sodium ascorbate, B – sample with the addition of acerola extract 3.5 g kg<sup>-1</sup>.

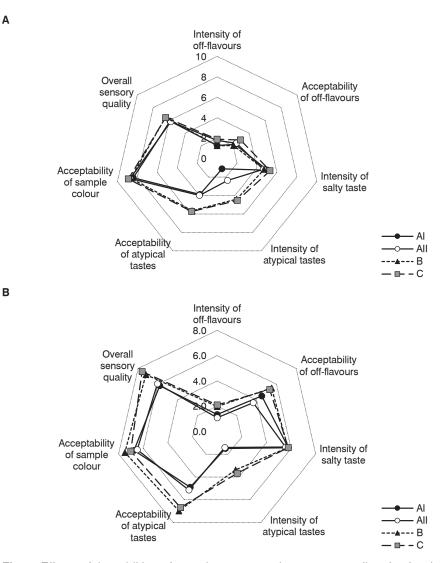


Fig. 4. Effects of the addition of acerola extract on the sensory quality of soft salami.

A - first replication, B - second replication.

Al – control sample with the addition of ascorbic acid, All – control sample with the addition of sodium ascorbate, B – sample with the addition of acerola extract 3.5 g kg<sup>-1</sup>, C – sample with the addition of acerola extract 5 g kg<sup>-1</sup>.

are relatively stable during the manufacturing of meat products. Moreover, the loss of carotenoids during the light storage is not so significant if the meat product is vacuum-packed [28]. Similar results on the positive effects of acerola on the colour of meat products were reported in the studies of BLOUKAS et al. [38], REALINI et al. [16], FRUET et al. [17] and DE PAIVA et al. [18].

#### Sensory analysis

The addition of acerola extract had a significant effect (P < 0.05) on increasing the intensity of foreign taste, which, however, was highly acceptable for the panellists (Fig. 4). Samples with acerola extract had higher overall quality in the second replication (P < 0.05). This could be related to statistically lower lightness, higher redness and higher yellowness in the samples with acerola on Day 7 of storage (Fig. 2), when the samples were sensorially evaluated. Other assessed parameters were comparable in all respects (P > 0.05). This agrees with the study of REALLINI et al. [16] and DE PAIVA et al. [18], who investigated the effect of the addition of acerola extract on the quality of meat products. Our results suggest that the addition of acerola extract had no negative effects on the sensory quality of soft salami.

# CONCLUSION

Our results showed that the addition of acerola extract to the soft salami vastly improves the colour and helps to considerably slow down the discolouration during storage in the dark and in the light. Moreover, the addition of acerola extract had a comparable or even better antioxidant effect than the standard used ascorbic acid or sodium ascorbate in soft salami. The addition of acerola in the range of 3.5-5 g·kg<sup>-1</sup> had no negative impact on the microbiological stability or sensorial quality. Thus, acerola could enable the replacement of questionable food additives in cooked meat products. The positive effect of acerola on the oxidative and colour stability of soft salami would be even more significant in the combination of functional packaging with an oxygen absorber.

Although acerola extract had a positive effect on the quality of soft salami, there are some limitations and challenges which relate to the use of acerola extract in the meat industry. One of them is the significantly higher price of the extract in comparison with the traditionally used antioxidants and colourants. However, the price is most likely to drop in the near future due to the growing market of acerola extracts. Another challenge for the use of acerola extract in meat products is the legislation. Acerola extract is not permitted as a food additive in the European Union. Some producers recommend labelling the usage of acerola on the package of the meat product as "acerola flavouring", which supposedly follows the Regulation (EC) No. 1334/2008 [40]. However, this way of labelling does not describe the real purpose of the use of acerola extract in meat products. To be classified as an approved food additive, antioxidant or colourant, acerola should undergo the authorization procedure according to the Regulation (EC) No. 1331/2008 [41].

Another drawback of the use of acerola extract in the meat industry is the need to find the right amount of the extract for every type of meat product. There is also a need for the analysis of the content of bioactive compounds in the individual batches of extract. Some limitations could also be related to the manufacturing of the meat products. The acerola extract used in our study was a fine powder, which made it more difficult to obtain a uniform blend throughout the meat batter than in the case of using ascorbic acid or sodium ascorbate.

Despite these limitations, we believe that acerola could be a suitable natural food additive for a wide range of meat products because of its positive effect on the oxidative and colour stability of the meat products, its lack of strong taste or odour and its attractiveness for consumers.

#### Acknowledgements

This work was supported from the grant of Specific University Research – grant No. A1\_FPBT\_2021\_004. Authors are grateful to the technical staff of the Department of Food Preservation for the verification of the content of bioactive compounds in the acerola extract.

## REFERENCES

- 1. Zeece, M.: Chapter Seven Food Additives. In: Zeece, M.: Introduction to the chemistry of food. Cambridge : Academic Press, 2020, pp. 251–311. ISBN: 978-0-12-809434-1. DOI: 10.1016/B978-0-12-809434-1.00007-4.
- Pipek, P.: Přídatné látky v masných výrobcích věčné téma. (Food additives in meat products – The neverending story.) Maso, *30*, 2019, pp. 4–10. ISSN: 1210-4086. In Czech.
- 3. Regulation (EC) No 1333/2008 of the European Parliament and of the Council of 16 December 2008 on food additives. Official Journal of the European Union, *51*, 2008, L354, pp. 16–33. ISSN: 1725-2555. <a href="http://data.europa.eu/eli/reg/2008/1333/oj">http://data.europa.eu/eli/reg/2008/1333/oj</a>
- Carocho, M. Morales, P. Ferreira, I. C. F. R.: Natural food additives: Quo vadis? Trends in Food Science and Technology, 45, 2015, pp. 284–295. DOI: 10.1016/j.tifs.2015.06.007.
- Adams, M. R. Moss, M. O. (Eds.): Food microbiology. 3rd edition. Cambridge : Royal Society of Chemistry, 2007. ISBN: 978-0-85404-284-5. DOI: 10.1039/9781847557940.
- Falowo, A. B. Fayemi, P. O. Muchenje, V.: Natural antioxidants against lipid-protein oxidative deterioration in meat and meat products: A review. Food Research International, *64*, 2014, pp. 171–181. DOI: 10.1016/j.foodres.2014.06.022.
- Hygreeva, D. Pandey, M. C. Radhakrishna, K.: Potential applications of plant-based derivatives as fat replacers, antioxidants, and antimicrobials in fresh and processed meat products. Meat Science, 98, 2014, pp. 47–57. DOI: 10.1016/j.meatsci.2014.04.006.
- Adamcová, M. van Andel, V. Strohalm, J. Houška, M. – Ševčík, R.: Effect of high-pressure processing and natural antimicrobials on the shelflife of cooked ham. Czech Journal of Food Sciences, 37, 2019, pp. 57–61. DOI: 10.17221/204/2018-CJFS.
- Lorenzo, J. M. Pateiro, M. Domínguez, R. Barba, F. J. – Putnik, P. – Kovačević, D. B. – Shpigelman, A. – Granato, D. – Franco, D.: Berries extracts as natural antioxidants in meat products: A review. Food Research International, *106*, 2018, pp. 1095–1104. DOI: 10.1016/j.foodres.2017.12.005.
- Alirezalu, K. Pateiro, M. Yaghoubi, M. Alirezalu, A. – Peighambardoust, S. H. – Lorenzo, J. M.: Phytochemical constituents,

advanced extraction technologies and techno-functional properties of selected Mediterranean plants for use in meat products. A comprehensive review. Trends in Food Science and Technology, *100*, 2020, pp. 292–306. DOI: 10.1016/j.tifs.2020.04.010.

- Van Haute, S. Raes, K. Van der Meeren, P. Sampers, I.: The effect of cinnamon, oregano and thyme essential oils in marinade on the microbial shelf-life of fish and meat products. Food Control, 68, 2016, pp. 30–39. DOI: 10.1016/j.foodcont.2016.03.025.
- Mariutti, L. R. B. Orlien, V. Bragagnolo, N. Skibsted, L. H.: Effect of sage and garlic on lipid oxidation in high-pressure processed chicken meat. European Food Research and Technology, 227, 2008, pp. 337–344. DOI: 10.1007/s00217-007-0726-5.
- Soltanizadeh, N. Ghiasi-Esfahani, H.: Qualitative improvement of low meat beef burger using *Aloe vera*. Meat Science, *99*, 2015, pp. 75–80. DOI: 10.1016/j.meatsci.2014.09.002.
- Bozkurt, H.: Utilization of natural antioxidants: Green tea extract and *Thymbra spicata* oil in Turkish dry-fermented sausage. Meat Science, *73*, 2006, pp. 442–450. DOI: 10.1016/j.meatsci.2006.01.005.
- Mezadri, T. Pérez-Gálvez, A. Hornero-Méndez, D.: Carotenoid pigments in acerola fruits (*Malpighia emarginata* DC.) and derived products. European Food Research and Technology, 220, 2005, pp. 63–69. DOI: 10.1007/s00217-004-1042-y.
- Realini, C. E. Guàrdia, M. D. Díaz, I. García-Regueiro, J. A. – Arnau, J.: Effects of acerola fruit extract on sensory and shelf-life of salted beef patties from grinds differing in fatty acid composition. Meat Science, 99, 2015, pp. 18–24. DOI: 10.1016/j.meatsci.2014.08.008.
- Fruet, A. P. B. Nörnberg, J. L. Calkins, C. R. De Mello, A.: Effects of different antioxidants on quality of beef patties from steers fed lowmoisture distillers grains. Meat Science, 154, 2019, pp. 119–125. DOI: 10.1016/j.meatsci.2019.04.014.
- 18. de Paiva, G. B. Trindade, M. A. Romero, J. T. da Silva-Barretto, A. C.: Antioxidant effect of acerola fruit powder, rosemary and licorice extract in caiman meat nuggets containing mechanically separated caiman meat. Meat Science, *173*, 2021, article 108406. DOI: 10.1016/j.meatsci.2020.108406.
- Belwal, T. Devkota, H. P. Hassan, H. A. Ahluwalia, S. – Ramadan, M. F. – Mocan, A. – Atanasov, A. G.: Phytopharmacology of acerola (*Malpighia* spp.) and its potential as functional food. Trends in Food Science and Technology, 74, 2018, pp. 99–106. DOI: 10.1016/j.tifs.2018.01.014.
- Moura, C. F. H. Oliveira, L. d. S. Souza, K. O. Franca, L. G. – Ribeiro, L. B. – Souza, P. A. – Miranda M. R. A.: Acerola – *Malpighia emarginata*. In: Rodrigues, S. – de Oliviera Silva, E. – de Brito, E. (Eds.): Exotic fruits. Reference guide. Amsterdam : Academic Press, 2018, pp. 7–14. ISBN: 978-0-12-803138-4. DOI: 10.1016/B978-0-12-803138-4.00003-4.
- 21. Souza, C. O. Silva, L. T. Silva, J. R. Lopez, J. A. Veiga-Santos, P. – Druzian J. I.: Mango and acerola

pulps as antioxidant additives in cassava starch bio-based film. Journal of Agricultural and Food Chemistry, *59*, 2011, pp. 2248–2254. DOI: 10.1021/ jf1040405.

- 22. Eça, K. S. Machado, M. T. C. Hubinger, M. D. Menegali, F. C.: Development of active films from pectin and fruit extracts: Light protection, antioxidant capacity, and compounds stability. Journal of Food Science, *80*, 2015, pp. C2389–C2396. DOI: 10.1111/1750-3841.13074.
- 23. Vyhláška č. 69/2016 Sb. O požadavcích na maso, masné výrobky, produkty rybolovu a akvakultury a výrobky z nich, vejce a výrobky z nich. (Regulation No 69/2016 of the Czech Parliament of 1 August 2016 on requirements for meat, meat products, fishery and aquaculture products and products thereof, eggs and products thereof.) Sbírka zákonů, 26, 2016, pp. 714–759. ISSN: 1211-1244. In Czech. <https:// www.zakonyprolidi.cz/cs/2016-69>
- 24. ČSN EN ISO 6887-1:2017. Microbiology of the food chain – Preparation of test samples, initial suspension, and decimal dilutions for microbiological examination. Part 1: General rules for the preparation of the initial suspension and decimal dilutions. Prague: Office for Standards, Metrology and Testing, 2017.
- 25. ČSN EN ISO 6887-2:2017. Microbiology of the food chain – Preparation of test samples, initial suspension, and decimal dilutions for microbiological examination. Part 2: Specific rules for the preparation of meat and meat products. Prague: Office for Standards, Metrology and Testing, 2017.
- 26. Tarladgis, B. G. Watts, B. M. Younathan, L. Dudan Jr., L.: A distillation method for the quantitative determination of malonaldehyde in rancid foods. Journal of the American Oil Chemists' Society, 37, 1960, pp. 44–48. DOI: 10.1007/BF02630824.
- Harding Thomsen, H. Zeuthen. P.: The influence of mechanically deboned meat and pH on the waterholding capacity and texture of emulsion type meat products. Meat Science, 22, 1988, pp. 189–201. DOI: 10.1016/0309-1740(88)90046-0.
- Velíšek, J. Hajšlová, J.: Chemie potravin. (Food Chemistry.) 3rd edition. Tábor : OSSIS, 2009. In Czech. ISBN: 978-80-86659-15-2.
- Munk, M. B. Huvaere, K. Bocxlaer, J. V. Skibsted, L. H.: Mechanism of light-induced oxidation of nitrosylmyoglobin. Food Chemistry, *121*, 2010, pp. 472–479. DOI: 10.1016/j.foodchem.2009.12.067.
- Skibsted, L. H.: Nitric oxide and quality and safety of muscle-based foods. Nitric Oxide, 24, 2011, pp. 176–183. DOI: 10.1016/j.niox.2011.03.307.
- Siripatrawan, U. Noipha, S.: Active film from chitosan incorporating green tea extract for shelf-life extension of pork sausages. Food Hydrocolloids, 27, 2012, pp. 102–108. DOI: 10.1016/j.foodhyd.2011.08.011.
- Böhner, N. Rieblinger, K.: Impact of different visible light spectra on oxygen absorption and surface discoloration of bologna sausage. Meat Science, *121*, 2016, pp. 207–209. DOI: 10.1016/j.meatsci.2016.06.019.
- 33. Larsen, H. Westad, F. Sørheim, O. Nilsen, L. H.:

Determination of critical oxygen level in packages for cooked sliced ham to prevent color fading during illuminated retail display. Journal of Food Science, 71, 2006, S407–S413. DOI: 10.1111/j.1750-3841.2006.00048.x.

- 34. Campo, M. M. Nute, G. R Hughes, S. I. Enser, M. – Wood, J. D. – Richardson, R. I.: Flavour perception of oxidation in beef. Meat Science, 72, 2006, pp. 303–311. DOI: 10.1016/j.meatsci.2005.07.015.
- Varvara, M. Bozzo, G. Celano, G. Disanto, Ch. Pagliarone, C. N. – Celano, G. V.: The use of ascorbic acid as a food additive: technical-legal issues. Italian Journal of Food Safety, *5*, 2016, article 4314. DOI: 10.4081/ijfs.2016.4313.
- Pipek, P.: Technologie masa. Vol. 2. (Meat Technology.) 3rd edition. Praha : VŠCHT, 1998. In Czech. ISBN: 80-7192-283-8.
- 37. Chang, S. K. Alasalvar, C. Shahidi, F.: Superfruits: Phytochemicals, antioxidant efficacies, and health effects – A comprehensive review. Critical Reviews in Food Science and Nutrition, 59, 2019, pp. 1580–1604. DOI: 10.1080/10408398.2017.1422111.
- Bloukas, J. G. Arvanitoyannis, I. S. Siopi, A. A.: Effect of natural colourants and nitrites on colour attributes of frankfurters. Meat Science, 52, 1999,

pp. 257-265. DOI: 10.1016/S0309-1740(98)00174-0.

- Borch, E. Kant-Muermans, M. L. Blixt, Y.: Bacterial spoilage of meat and cured meat products. International Journal of Food Microbiology, *33*, 1996, pp. 103–120. DOI: 10.1016/0168-1605(96)01135-X.
- 40. Regulation (EC) No 1334/2008 of the European Parliament and of the Council of 16 December 2008 on flavourings and certain food ingredients with flavouring properties for use in and on foods and amending Council Regulation (EEC) No 1601/91, Regulations (EC) No 2232/96 and (EC) No 110/2008 and Directive 2000/13/EC. Official Journal of the European Union, 51, 2008, L354, pp. 34–50. ISSN: 1725-2555. <http://data.europa.eu/eli/reg/2008/1334/ oj>
- 41. Regulation (EC) No 1331/2008 of the European Parliament and of the Council of 16 December 2008 establishing a common authorisation procedure for food additives, food enzymes and food flavourings. Official Journal of the European Union, 51, 2008, L354, pp. 1–6. ISSN: 1725-2555. <a href="http://data.europa.eu/eli/reg/2008/1331/oj">http://data.europa.eu/eli/reg/2008/1331/oj</a>>

Received 9 May 2022; 1st revised 3 October 2022; accepted 19 October 2022; published online 14 November 2022.