Scientific Papers. Series D. Animal Science. Vol. LXVII, No. 1, 2024
ISSN 2285-5750; ISSN CD-ROM 2285-5769; ISSN Online 2393-2260; ISSN-L 2285-5750

ENHANCING ANTIOXIDANT CAPACITY IN FUNCTIONAL MEAT PRODUCTS THROUGH INFUSION WITH SEA BUCKTHORN OIL TO COMBAT INHERENT ANTIOXIDANT DEFICIENCY

Bianca Georgiana ANCHIDIN, Florin-Daniel LIPȘA, Diana-Remina MANOLIU, Marius Mihai CIOBANU, Mihai Cătălin CIOBOTARU, Ioana GUCIANU, Paul Corneliu BOIȘTEANU

"Ion Ionescu de la Brad" Iasi University of Life Sciences, 3 Mihail Sadoveanu Alley, 700490, Iasi, Romania

Corresponding author email: paul.boișteanu@iuls.ro

Abstract

Given the growing concern in recent years for a healthier diet, attention must also be directed towards improving the quality profile of meat products and transforming them, as much as possible, into functional foods that combine the benefits of plant-based products with those of animal-origin products. With this in mind, we aimed to develop a functional meat product, given the recent scrutiny these products have faced, by using an oil with antioxidant effects to enhance the antioxidant profile of products with insignificant endogenous antioxidant levels. To achieve this, three batches of pork tenderloin were injected with 1%, 3%, and 5% sea buckthorn oil, and were analyzed in terms of antioxidant capacity, physicochemical and microbiological quality, and sensory perception. Most results showed highly significant differences ($p < 0.001$) between batches, with superior quality observed in the batch injected with 5% sea buckthorn oil and subjected to heat treatment. However, consumers preferred the batch injected with 3% sea buckthorn oil due to its more balanced taste. This research underscores the potential to develop meat-based functional foods with enhanced nutritional benefits.

Key words*: antioxidants, functional meat products, meat products, sea buckthorn oil.*

INTRODUCTION

Meat and meat products are important foods with essential nutritional components such as essential amino acids, fatty acids, vitamins, and minerals, which form a significant component for normal physiological and biochemical processes (Kausar et al., 2019).

There is an increasing demand for healthier meat and meat products that contain lower levels of fat, reduced cholesterol, reduced sodium chloride and nitrite content, an updated fatty acid profile, and the addition of health-enhancing ingredients among consumers worldwide (Kausar et al., 2019).

The term of functional foods was first introduced in Japan in the mid-1980s and refers to processed foods that contain ingredients which help specific functions of the body in addition to being nutritious (Kumar et al., 2012). The concept of functional foods stems from the traditional paradigm of providing methods for preventing nutritional deficiencies; this paradigm includes foods that offer health benefits through micronutrient fortification (Katan & De Roos, 2004). To date, Japan is the only country that has formulated a specific regulatory approval process for functional foods. Known as Foods for Specified Health Use (FOSHU), these foods are eligible to bear an approval seal from the Japanese Ministry of Health and Welfare (Kumar et al., 2012).

The European Commission's Concerted Action on Functional Food Science in Europe (FuFoSE), coordinated by the International Life Sciences Institute (ILSI) Europe, defined functional foods as: 'food products that, in addition to their basic nutritional impact, have beneficial effects on one or more functions of the human body, thereby improving general and physical conditions and/or reducing the risk of disease progression. The amount of consumption and the form of functional foods should be as typically expected, consumed in the daily diet. Therefore, they should not be in the form of pills or capsules, but only in the form of conventional foods' (Hoffmann et al., 2010). The International Life Sciences Institute

of North America (ILSI) states that functional foods are 'foods that, due to physiologically active food components, provide health benefits beyond basic nutrition.' This definition is vague, but encompasses the scope of functional foods (Vattem & Maitin, 2016).

One of the most interesting functional ingredients that can be added to meat products is sea buckthorn oil, which contains high concentrations of vitamin C, carotenoids, tocopherols, and other bioactive compounds with a strong antioxidant role (Anchidin et al., 2023).

Common sea buckthorn (*Hippophae rhamnoides*), also known as Siberian pineapple, is a spiny, dioecious shrub (or tree) from the family Elaeagnaceae that can grow up to 7 meters in height (Fu et al., 2014; Wang et al., 2014). Sea buckthorn has a unique nutritional composition, containing vitamins (A, C, D, E, F, K, P, and the B complex), 18 free amino acids, and a unique profile of unsaturated fatty acids, making it the only plant source of omega-7. Its bioactive phytochemical compounds possess various biological activities such as antioxidant, immunomodulatory, anticarcinogenic, hepatoprotective, cardioprotective, anti-atherogenic, and radioprotective properties (Shah et al., 2021).

Sea buckthorn fruit oil contains an average of 35% palmitoleic acid (16:1n-7) (Suryakumar & Gupta, 2011). Sea buckthorn oil is rich in antioxidant compounds such as vitamins A and E, sterols, and flavonoids (Ursache et al., 2017). It has been used in traditional medicine in Eastern Europe and Asia for the treatment of asthma, circulatory disorders, and other conditions (Suryakumar & Gupta, 2011; Zadernowski et al., 1997). Seed oil contains high concentrations of tocopherols (140 mg/100 ml), 1% phytosterols, and small amounts of tocotrienols (Yang & Kallio, 2001).

The antioxidant effects in fruits and vegetables may arise from phenolic compounds such as flavonoids and phenolic acids, or from nitrogenous compounds such as alkaloids, chlorophyll derivatives, amino acids, and amines. These flavonoids and other phenolic compounds of plant origin have been reported as scavengers and inhibitors of lipid peroxidation (Song et al., 2013). Sea buckthorn oil has been reported as a potent antioxidant in all in vitro model systems it has been added to (Chauhan et al., 2007).

This study aimed to assess the effect of sea buckthorn oil injection on relevant quality parameters of pork tenderloin (psoas major) and derived products with compact texture subjected to heat treatment. The central objective was to investigate the potential of this oil as a natural antioxidant, with the potential to replace synthetic antioxidants. Thus, the influence of sea buckthorn oil on essential physicochemical aspects of meat, as well as its antioxidant and antimicrobial capacity, was analyzed, with a focus on products exposed to heat treatment.

MATERIALS AND METHODS

In order to achieve the aim of the study, three functional products with pork tenderloin were developed, by injection with 1%, 3%, and 5% sea buckthorn oil. The pork tenderloin, as the raw material, was purchased from Metro Cash & Carry România S.R.L. in Iași. The organic sea buckthorn fruit oil (*Hippophae rhamnoides* L.) used for injecting the experimental batches was obtained from Zorian Export S.R.L. in Iași County, produced through cold pressing and without any chemical treatment.

The total 6 experimental batches are presented in Table 1, along with the ingredients used in their manufacture and the heat treatment applied to them. For the laboratory analyses necessary for characterizing the studied products, samples from these batches were accompanied by samples of sea buckthorn oil (*Hippophae rhamnoides* L.) and of the meat used as raw material in the batches. The laboratory analyses conducted included physicochemical antioxidant capacity, and microbiological analyses. The experimental batches and the qualitative analyses were carried out at the "Ion Ionescu de la Brad" Iași University of Life Sciences (IULS) and at the Institute of Research for Agriculture and Environment (ICAM).

The experimental protocol for the pork tenderloin samples (M1S, M2S, and M3S) involved salting the meat pieces with 2% salt (dry brining) and injecting them with sea buckthorn oil, in the concentrations presented in Table 1. The prepared pieces were vacuum-

sealed to facilitate better absorption of the oil into the meat and refrigerated under vacuum for 3 days until physicochemical analyses were conducted. The pastrami batches (P1S, P2S, and P3S) followed the same initial experimental protocol as the M1S, M2S, and M3S batches, but in addition to these, they underwent a heat treatment (Table 1) consisting of 4 stages, each lasting between 20 and 30 minutes.

Table 1. The composition of the experimental batches and the applied heat treatments

Experimental batches	Ingredients				Heat treatment (°C)			
	Pork tenderloin (%)	Sea buckthorn oil (%)	Salt (%)	Pepper crust (present or absent)	Drying I	Smoking	Boiling	Drying II
U	-	100	-	-	-			
C	100	-	-	-	-			
M1S	97	1	2	-				
M2S	95	3	2	-	-			
M3S	93	5	2	-	-			
P1S	97	1	2	X	65	65	72	80
P2S	95	3	2	X	65	65	72	80
P3S	93	5	2	X	65	65	72	80

The crude chemical determinations included quantitative analysis of moisture content, protein quantity, collagen, fat content, and salt concentration using a versatile method of near-infrared spectroscopic determination (NIR) (Gucianu et al., 2023) employing the Food Check meat analyzer (Bruins Instruments, Germany).

The assessment of the antioxidant activity of the experimental batches involved determining the content of polyphenols, flavonoids, and the scavenging activity of DPPH and ABTS radicals.

The extraction of compounds exerting antioxidant capacity is a crucial step in determining the antioxidant capacity of any food (Echegaray et al., 2022; López-Fernández et al., 2020). Meat is no exception, and extraction processes are particularly important for subsequent correct analysis. In this regard, solid-liquid extraction is the most commonly used in meat matrices. However, this process is performed in various ways, as different conditions can be used for this purpose (e.g., different solvents, different extraction times, and different extraction temperatures) (Wu et al., 2008). For this reason, we decided to use multiple methods for determining antioxidant compounds, as can be seen above. The solvents we used were represented by hexane-acetone (ABTS), ethanol (DPPH, polyphenols, and flavonoids).

For the total polyphenol content (TPC), the Folin-Ciocâlteu colorimetric method was used, as described by Blainski et al. (2013). The flavonoid content of the samples was determined using the method described by Zhishen et al. (1999) and Dewanto et al. (2002).

The determination of antioxidant capacity by the DPPH method was carried out following the procedure described by Pires et al. (2017), while that by the ABTS assay was conducted according to the method described by Dumitrașcu et al. (2022).

To detect the absorption of the samples, a UV-vis spectrophotometer Specord Plus 210 (Analytik Jena, Germany) was used. Sampling was done after vortex mixing to ensure reproducibility. The results were expressed as mg/g DW (for flavonoids), mg GAE/g DW (for polyphenols), µMol Trolox/g DW (for the DPPH assay), and µMol Trolox/g DW (for the ABTS assay). The solutions used in the antioxidant value determination analyses were purchased from Sigma Aldrich Steinheim (Darmstadt, Germany).

For the microbiological analyses, the following solutions/culture media bases were used: peptone water, Rapid *Staph* Agar base, Rapid *E. coli* 2 Agar base, Rapid *Salmonella* Agar base, and Plate Count Agar (PCA). All of these were purchased from Bio-Rad (Marnes-la-Coquette, France), except for the last culture medium, which was purchased from Scharlau (Barcelona, Spain).

The steps of evaluating the microbiological contamination of the meat and oil samples

studied were as follows: (a) homogenization and weighing of the samples, (b) dilution, (c) inoculation on appropriate media, (d) incubation at specific temperatures and times, and (e) counting specific colonies indicating the presence of microorganisms (Otero et al., 1998). All analytical procedures performed to determine the microbial load were carried out in a sterile environment, with five replicates for each sample. One gram of meat/oil was homogenized with 9 milliliters of peptone water. This represented a 10-1 dilution, followed by vortexing and serial dilution of each analyzed sample to a concentration of 10-3, by taking one milliliter from the previous dilution and diluting it in 9 mL of peptone water. One milliliter of the prepared samples was inoculated onto Petri dishes, onto which the specific culture medium previously prepared according to the instructions on the technical sheet was added. After the incubation period, colonies of microorganisms from the meat and oil samples were manually counted, and the results were expressed as logarithmic colony forming units per gram (log CFU/g).

The color of the samples was determined using the portable colorimeter Konica Minolta CR-410, in the CIE tridimensional color system, measuring the color parameters L*, a*, and b* with illuminant D65 at a 10-degree observation angle (Boișteanu et al., 2023; Manoliu et al., 2023). The instrument was calibrated on a white calibration plate for standard values before starting the measurements.

The colorimetric determinations for Chroma and Hue angle were determined using formulas (1) and (2), following the model of Turgut et al., 2017.

$$H(*) = tan^{-1} \times b^*/a^* \quad (1)$$

$$Chroma\ (C^*) = \sqrt{(a*)^2 + (b*)^2} \quad (2)$$

pH measurements were conducted using a HANNA HI 99163 Meat pH-meter. The electrode was inserted into the meat following prior calibration in buffer solutions with known pH values (an acidic solution with pH 4.01 and a neutral solution with pH 7.01). The probe of the meter was cleaned with distilled water after calibration and between readings to ensure that the results obtained were not influenced (Ciobanu et al., 2022).

A sensory evaluation of the pork tenderloin samples that were thermally treated (pastrami – P1S, P2S, and P3S) was conducted 24 hours after completing all technological stages by 12 experienced panellists (6 males and 6 females). The samples were labeled with random three-digit numbers. Generalized Procrustes Analysis (GPA) was used for processing the sensory data to evaluate the results of the sensory tests conducted by the tasting panel. All these analyses were performed using XLSTAT, an add-in software for Microsoft Office Excel (Trial Version 2024, Addinsoft, Paris, France).

All experiments were performed in five replicates. The results are expressed as mean ± standard deviation. Statistical comparisons were conducted using one-way analysis of variance (ANOVA) with IBM SPSS Statistics V21 software. Differences were considered significant when p-values were less than 0.05.

RESULTS AND DISCUSSIONS

Table 2 presents the main physico-chemical characteristics analyzed within the studied batches (moisture, proteins, collagen, fat, salt, and pH).

The batches of pork tenderloin injected with sea buckthorn oil and not thermally treated (M1S, M2S, and M3S) exhibited lower average moisture values of 75.74 ± 0.114% (M1S), 75.46 ± 0.114% (M2S), and 74.68 ± 0.109% (M3S) compared to the control batch (C), where this parameter value was 76.02 ± 0.130%. However, these values were higher than those of the batches of pork tenderloin thermally treated for the same oil injection amount (P1S, P2S, and P3S). The P1S batch obtained the highest moisture value among the thermally treated batches (75.54 ± 0.114%), with this value increasing directly proportional to the increase in the amount of oil contained in the pork tenderloin. The batch treated with 3% sea buckthorn oil (P2S) showed a decrease in moisture to 74.74 ± 0.251%, while the batch injected with 5% sea buckthorn oil and

thermally treated (P3S) recorded the lowest moisture among all studied batches, at 74.30 ± 0.071% (Table 2). The decrease in moisture following thermal treatment is a consequence of the increased temperature in the center of the product. The amount of moisture lost depends on the method of thermal treatment and the amount of connective tissue in the meat (Vinnikova et al., 2019). Pork tenderloin is known to be one of the most tender anatomical regions with a low content of connective tissue, as shown by the study of Nishimura et al. (2009). The progressive decrease in moisture in both thermally treated and untreated samples is also due to the increased addition of sea buckthorn oil. As the amount of lipids in meat or meat products increases, whether endogenous or exogenous, it leads to a decrease in moisture, as observed by Petrov et al. (2008) for meat with different amounts of fat.

The amount of fat (Table 2) exhibited progressive increases among batches, concurrent with the percentage of sea buckthorn oil added to them. The lowest fat content was observed in the control batch (C), at 1.82 ± 0.148%, which is expected considering that the psoas major is one of the anatomical portions with the lowest fat quantities. The batches not thermally treated had lower fat values, at 2.38 ± 0.179% (M1S), 3.06 ± 0.167% (M2S), and 3.80 ± 0.187% (M3S), compared to the batches of pork tenderloin that were thermally treated, where the mean values obtained were 2.54 ± 0.114% (P1S), 3.22 ± 0.192% (P2S), and 4.00 ± 0.158% (P3S). These changes in the fat percentage are attributed to the exogenous addition of vegetable fat (sea buckthorn oil) and the loss of moisture during the thermal process, but may also correlate with an inhibition of lipid oxidation induced by sea buckthorn oil. Both within batches subjected to the same thermal treatment and across all batches analyzed together statistically, very significant statistical differences are observed ($p < 0.001$) (Table 2). These results largely correlate with those obtained by us in previous research on this type of thermally treated product. In the current study, the mean value of the fat parameter increases directly proportional to the increase in the quantity of sea buckthorn oil added, which contrasts with our previous study where in the batch injected with 3% sea buckthorn oil, the value is lower than in the batches injected with 1% and 5% sea buckthorn oil (Anchidin et al., 2023). This discrepancy could be due to faulty injection of the sea buckthorn oil into the meat pieces, uneven distribution within the product mass, and sampling from a portion where an inadequate amount of sea buckthorn oil reached in line with the injected value. The issue we consider to have led to this "error" might be the type of injection used, namely manual injection.

Regarding the protein content in the studied batches, a slight gradual decrease is observed in the batches that were not thermally treated compared to the control batch (C) (Table 2). The mean protein content in this latter batch is 22.00 ± 0.100%, which is the highest value in our study for this qualitative parameter. From this value, it decreases successively to 21.80 ± 0.070% (batch M1S), 21.74 ± 0.134% (batch M2S), and 21.34 ± 0.114% (M3S) in the case of thermally untreated pork tenderloin. Regarding the batches of pork tenderloin that were thermally treated, since heat can modify/decompose proteins, it can significantly influence the nutritional properties of meat products (Yu et al., 2017). For this reason, in some of the thermally treated batches with the same quantity of injected sea buckthorn oil, a slightly lower mean value of this qualitative parameter is observed, as is the case with batches M1S and P1S, where the recorded values are 21.80 ± 0.070% and 21.42 ± 0.070%, respectively. The same situation was observed in batches M2S and P2S, where the obtained values were 21.74 ± 0.134% and 21.58 ± 0.084%, respectively. The only batches that did not follow this trend were the ones injected with 5% sea buckthorn oil (M3S and P3S), where the protein value of the thermally untreated sample was 21.34 ± 0.114%, lower than the 21.36 ± 0.134% obtained by the batch injected with 5% sea buckthorn oil and thermally treated. Thermal treatment had a lesser influence on the fluctuation of the mean protein value compared to the addition of sea buckthorn oil, but the differences between the studied batches were still distinctly significant (0.010**) (Table 2).

The collagen content of the pork tenderloin samples injected with sea buckthorn oil shows,

similar to moisture and protein content, a decreasing trend as the percentage of injected oil increases. The only analyzed sample that does not follow this trend is the pork tenderloin injected with 1% sea buckthorn oil and thermally treated (P1S), where the highest collagen value was recorded, 20.60 ± 0.100%, which is even higher than the control batch (C), where its value was 20.38 ± 0.084. This represents the only instance where a parameter of chemical quality, which typically decreases with increasing sea buckthorn oil quantity injected, shows an increase in its value surpassing the control batch (Table 2). The rest of the mean collagen values for pork tenderloin samples injected with sea buckthorn oil, like moisture, showed successive decreases from the control batch value (20.38 ± 0.084) as follows: 20.24 ± 0.089 in batch M1S, 20.06 ± 0.114 in batch M2S, 19.78 ± 0.148 in batch M3S. Similarly, the values of batches subjected to thermal treatment also showed a decrease in the mean collagen value compared to the control batch (C), starting from batch P2S, with a value of 19.83 ± 0.130%, and continuing with P3S - 19.78 ± 0.084%. The differences recorded following the application of the analysis of variance (ANOVA) for collagen value variation were distinctly significant (0.007**) for the thermally treated pork tenderloin batches (P1S, P2S, and P3S) and highly significant (0.000***) for all analyzed batches (Table 2).

Table 2. Means ± standard deviation of the physicochemical results obtained by the studied batches

Experimental groups	Physicochemical parameters (%)					
	Moisture	Protein	Collagen	Fat	Salt	pH
M1S	75.74 ± 0.114^{c}	21.80 ± 0.070^{b}	20.24 ± 0.089^{b}	2.38 ± 0.179^{a}	1.92 ± 0.109^{a}	5.91 ± 0.035^{a}
M2S	75.46 ± 0.114^{b}	21.74 ± 0.134^{a}	20.06 ± 0.114^{b}	3.06 ± 0.167^{b}	2.22 ± 0.084^{b}	6.09 ± 0.047^{b}
M3S	74.68 ± 0.109^{a}	21.34 ± 0.114^{a}	19.78 ± 0.148^{a}	3.80 ± 0.187^{c}	2.22 ± 0.084^{b}	6.09 ± 0.046^{b}
p-value	**0.000 (***)**	**0.000 (***)**	**0.000 (***)**	**0.000 (***)**	**0.000 (***)**	**0.000 (***)**
P1S	75.54 ± 0.114^{c}	21.42 ± 0.070^{a}	20.60 ± 0.100^{b}	2.54 ± 0.114^{a}	1.98 ± 0.164^{a}	6.184 ± 0.029^{a}
P2S	74.74 ± 0.251^{b}	21.58 ± 0.084^{b}	19.83 ± 0.130^{a}	3.22 ± 0.192^{b}	2.14 ± 0.134^{a}	6.22 ± 0.029^{a}
P3S	74.30 ± 0.071^{a}	21.36 ± 0.134^{a}	19.78 ± 0.084^{a}	4.00 ± 0.158^{c}	2.12 ± 0.084^{a}	6.24 ± 0.036^{a}
p-value	**0.000 (***)**	**0.010 (**)**	**0.007 (**)**	**0.000 (***)**	0.154 (ns)	0.064 (ns)
C	76.02 ± 0.130^{d}	22.00 ± 0.100^{d}	20.38 ± 0.084^{b}	1.82 ± 0.148	–	5.79 ± 0.079
Total p-value	**0.000 (***)**	**0.000 (***)**	**0.000 (***)**	**0.000 (***)**	**0.001 (***)**	**0.000 (***)**

Different letters on the same column indicate significant difference (estimated by ANOVA analysis and Tukey's test, $p \leq 0.05$).
M1S - pork tenderloin injected with 1% sea buckthorn oil, untreated thermally; M2S - pork tenderloin injected with 3% sea buckthorn oil, untreated thermally; M3S - tenderloin injected with 5% sea buckthorn oil, untreated thermally; P1S - tenderloin (pastrami) injected with 1% sea buckthorn oil, thermally treated; P2S - tenderloin (pastrami) injected with 3% sea buckthorn oil, thermally treated; P3S - tenderloin (pastrami) injected with 5% sea buckthorn oil, thermally treated.

The values for the salt content of the studied batches do not include the control sample (C) because it was not subjected to the salting process like the other studied batches. In the batches where sea buckthorn oil was injected, a prior salting process also took place. The values referring to the salt content of the studied samples do not show major fluctuations; however, the batches that were not subjected to thermal treatment recorded highly significant differences ($p < 0.001$) among the studied batches during statistical analysis. These results are in contrast to those obtained for the thermally treated samples, where the differences were not significant ($p > 0.05$). The differences observed between the batches subjected to thermal treatment and those that were not treated thermally were extremely significant, presenting a statistical significance level of up to 0.001*** (Table 2).

The highest average value for the salt content was obtained in batches M2S and M3S, both registering 2.22 ± 0.084%. The lowest value for the same parameter was observed in one of the

batches that did not undergo thermal treatment, namely M1S, with a value of 1.92 ± 0.109% (Table 2). Intermediate values were recorded in batches that underwent thermal treatment – P1S, P2S, and P3S, with values of 1.98 ± 0.164%, 2.14 ± 0.134%, and 2.12 ± 0.084%, respectively.

Statistical differences for the pH of the analyzed samples were, as with the salt content, non-significant (0.064) for the batches subjected to thermal treatment and highly significant (0.000***) for the batches only injected with sea buckthorn oil and not subjected to other technological operations. The statistical analysis of all studied batches, as shown in Table 2, revealed highly significant differences (0.000***) among them. pH values showed a continuous increase, concurrent with the increase in the percentage of injected oil, starting from the value of the control batch (5.79 ± 0.079%). Thus, batches M1S, M2S, and M3S recorded mean pH values of 5.91 ± 0.035%, 6.09 ± 0.047%, and 6.09 ± 0.046%, respectively. An increase in pH is also observed after the thermal treatment stage, with the following results obtained: 6.184 ± 0.029% (P1S), 6.22 ± 0.029% (P2S), and 6.24 ± 0.036% (P3S).

In continuation of this study, we evaluated the antioxidant capacity of the batches injected with sea buckthorn oil (M1S, M2S, M3S, P1S, P2S, P3S). The spectrophotometric analyses used to measure the antioxidant activity in the aforementioned batches consisted of determinations for: total flavonoid content (TFC), total polyphenol content (TPC), DPPH assay, and ABTS assay.

Sea buckthorn oil is rich in antioxidant compounds such as vitamins A and E, sterols, and flavonoids (Ursache et al., 2017). In the non-heat-treated batches, the lowest value of total flavonoid content was observed in batch M1S, at 0.10 ± 0.004 mg CE/g DW, gradually increasing in batches M2S and M3S (where average values of 0.14 ± 0.014 and 0.20 ± 0.014 were obtained), while in the heat-treated batches, these values increased significantly ($p < 0.05$) up to 0.43 ± 0.005 (P3S). This significant increase in phenolic content suggests that temperature has an effect on the stability of flavonoids and their biological activity (Chaaban et al., 2017). Additionally, we observe a consistent increase in antioxidant activity parallel to the increase in the percentage of sea buckthorn oil in the analyzed products.

The analysis of total polyphenolic compounds in the batches of injected and non-heat-treated pork tenderloin revealed significantly different values ($p < 0.05$) from one sample to another, with values of 0.23 ± 0.036 mg GAE/g DW (M1S), 0.33 ± 0.016 mg GAE/g DW (M2S), and 0.47 ± 0.012 mg GAE/g DW (M3S). This increase could reflect the percentage variations in the injection of batches with sea buckthorn oil. Furthermore, the batches of injected pork tenderloin subjected to heat treatment showed a significant increase ($p < 0.05$) in polyphenolic content compared to the non-heat-treated batches, with values ranging from 0.87 ± 0.069 to 1.08 ± 0.017 mg GAE/g DW (Table 3). This increase could be attributed to the synergistic effects between heat treatment and the compounds in the injected oil, which could promote the release or formation of new polyphenolic compounds. The antioxidant activity of food matrices containing phenolic compounds, after heat treatment and exposure to light, may remain constant, increase, or decrease. The evolution depends on the interactions between molecules and food matrices, as well as operating conditions (Ioannou et al., 2020).

The DPPH assay, like the other antioxidant capacity analyses in the present study, highlights positive effects on antioxidant activity directly proportional to the increase in the percentage of sea buckthorn oil added to the products, as well as to the heat treatment. Batches M1S and M2S show relatively similar levels of μMol Trolox/g DW, at 4.62 ± 0.150 μMol Trolox/g DW and 4.99 ± 0.030 μMol Trolox/g DW, respectively, with no significant differences between them ($p > 0.05$). From the injection of 5% sea buckthorn oil into the pork tenderloin, a more significant increase in antioxidant activity is observed, reaching 7.10 ± 0.112 μMol Trolox/g DW (Table 3). According to statistical analysis, the differences between these three described batches are highly significant ($p < 0.001$). Significant differences ($p < 0.05$) are observed between batches M1S, M2S, M3S, and between the batches of pork tenderloin injected with sea

buckthorn oil and subjected to heat treatment (P1S, P2S, and P3S). Batch P1S, injected with 1% oil and subjected to heat treatment, obtained a significantly different value ($p < 0.05$) of 9.60 ± 0.152 µMol Trolox/g DW compared to batch M3S, injected with 5% sea buckthorn oil but not subjected to heat treatment, which measured 7.10 ± 0.112 µMol Trolox/g DW. Batches P2S and P3S, injected with 3% and 5% sea buckthorn oil, respectively, showed values with nonsignificant differences ($p<0.05$) between them (10.28 ± 0.152 µMol Trolox/g DW and 10.42 ± 0.188 µMol Trolox/g DW, respectively). Statistical differences show distinct significant differences ($p<0.01$) between the batches subjected to heat treatment (P1S, P2S, and P3S) for the DPPH test (Table 3).

The last test used in our study to determine antioxidant activity was the ABTS assay, as shown in Table 3. The highest values of antioxidant activity were obtained in this assay. In the untreated batches, batch M3S (injected with 5% sea buckthorn oil) obtained the highest antioxidant value in this category, at 1617.17 ± 14.753 µMol Trolox/g DW, with the lowest being obtained by the batch injected with 1% sea buckthorn oil (M1S). In the case of batches subjected to the ABTS assay that underwent heat treatment, a different situation was observed compared to the results of the other antioxidant analyses conducted (Table 3). This translates to a lack of successive increase in antioxidant capacity values, as seen in the other tests. Instead, a decrease in antioxidant capacity was observed from 1688.84 ± 15.238 µMol Trolox/g DW (P1S – pastrami injected with 1% sea buckthorn oil) to 1522.24 ± 4.681 µMol Trolox/g DW (P3S - pastrami injected with 3% sea buckthorn oil). This latter value is the lowest antioxidant activity value obtained using the ABTS assay among the samples injected with sea buckthorn oil, regardless of the presence or absence of heat treatment. This result could be due to uneven distribution of sea buckthorn oil in the product mass or defective extraction, considering that batch P3S obtained the highest antioxidant value in this study, at 1802.28 ± 3.770 µMol Trolox/g DW (Table 3). In all phytochemical activity characterization tests, highly significant differences ($p<0.001$) were observed within the same test for all analyzed samples (Table 3).

Table 3. Characterization of the phytochemicals in the studied batches

Experimental groups	Phytochemical content			
	TFC, mg/g DW	TPC, mg GAE/g DW	DPPH µMol Trolox/g DW	ABTS µMol Trolox/g D.W.
M1S	0.10 ± 0.004^{b}	0.23 ± 0.036^{b}	4.62 ± 0.150^{b}	1566.81 ± 9.002^{c}
M2S	0.14 ± 0.014^{c}	0.33 ± 0.016^{c}	4.99 ± 0.030^{b}	1584.35 ± 6.179^{c}
M3S	0.20 ± 0.014^{d}	0.47 ± 0.012^{d}	7.10 ± 0.112^{c}	1617.17 ± 14.753^{d}
p-value	**0.000 (***)**	**0.000 (***)**	**0.000 (***)**	**0.003 (**)**
P1S	0.31 ± 0.016^{e}	0.87 ± 0.069^{e}	9.60 ± 0.152^{d}	1688.84 ± 15.238^{e}
P2S	0.38 ± 0.008^{f}	0.97 ± 0.025^{f}	10.28 ± 0.152^{e}	1522.24 ± 4.681^{b}
P3S	0.43 ± 0.005^{g}	1.08 ± 0.017^{g}	10.42 ± 0.188^{e}	1802.28 ± 3.770^{f}
p-value	**0.000 (***)**	**0.003 (**)**	**0.002 (**)**	**0.000 (***)**
C	0.05 ± 0.015^{a}	0.09 ± 0.013^{a}	3.06 ± 0.176^{a}	651.25 ± 16.74^{a}
p-value	**0.000 (***)**	**0.000 (***)**	**0.000 (***)**	**0.000 (***)**

Different letters on the same column indicate significant difference (estimated by ANOVA analysis and Tukey's test, $p \leq 0.05$).
M1S - pork tenderloin injected with 1% sea buckthorn oil, untreated thermally; M2S - pork tenderloin injected with 3% sea buckthorn oil, untreated thermally; M3S - tenderloin injected with 5% sea buckthorn oil, untreated thermally; P1S - tenderloin (pastrami) injected with 1% sea buckthorn oil, thermally treated; P2S - tenderloin (pastrami) injected with 3% sea buckthorn oil, thermally treated; P3S - tenderloin (pastrami) injected with 5% sea buckthorn oil, thermally treated.

The results regarding antioxidant activity show that the ABTS assay recorded the highest values of antioxidant activity compared to the DPPH assay, with significant differences (Table 3). These highly different results can be explained by the type of antioxidants each of these two tests can measure. The ABTS assay measures both hydrophilic and lipophilic antioxidants, while the DPPH assay applies only to hydrophobic systems (Munteanu & Apetrei, 2021).

Following the Pearson correlations between phytochemical and physicochemical parameters, strong and significant relationships

($p<0.01$) were identified among all studied parameters (Table 4).

The strongest positive correlations of TFC were observed with TPC (0.992**) and DPPH (0.977**). Very strong correlations were also observed between TPC and DPPH (0.987**), indicating a close association between flavonoid and phenolic content and antioxidant activity. Very strong positive correlations between the results of these antioxidant capacity determination analyses and pH were observed, indicating that an increase in pH value leads to an increase in antioxidant activity. Strong negative correlations of TFC, TPC, and DPPH are observed with the chemical parameters moisture, protein, and collagen (Table 4). The chemical parameter fat strongly correlates with the results of antioxidant capacity determination tests, as seen in Table 4. These results are compatible with those in Tables 2 and 3, where an increase in the amount of fat in the batch is observed as the percentage of sea buckthorn oil injected increases, but there is also an increase in antioxidant activity in batches with a higher addition of sea buckthorn oil. Moisture, protein, and collagen show significantly negative correlations with TFC, TPC, DPPH, and ABTS (Table 4). These results suggest that batches with lower moisture, protein, and collagen content had a higher concentration of antioxidant compounds.

Table 4. Pearson correlations between the results of the phytochemical compound analysis and those of the physicochemical analysis

Parameters	TFC	TPC	DPPH	ABTS	Moisture	Protein	Collagen	Fat	Salt	pH
TFC	1	**0.992****	**0.977****	**0.637****	**-0.765****	**-0.694****	**-0.630****	**0.673****	**0.562****	**0.875****
TPC		1	**0.987****	**0.635****	**-0.711****	**-0.697****	**-0.634****	**0.624****	**0.550****	**0.874****
DPPH			1	**0.672****	**-0.721****	**-0.758****	**-0.672****	**0.642****	**0.605****	**0.906****
ABTS				1	**-0.581****	**-0.717****	**-0.731****	**0.682****	**0.941****	**0.785****
Moisture					1	**0.709****	**0.663****	**-0.941****	**-0.577****	**-0.775****
Protein						1	**0.929****	**-0.731****	**-0.611****	**-0.788****
Collagen							1	**-0.741****	**-0.658****	**-0.770****
Fat								1	**0.685****	**0.777****
Salt									1	**0.782****
pH										1

** Correlation is significant at the 0.01 level.

For the determination of the color of all analyzed samples, the CIELab system was used, and its results are presented in Table 5, along with the chromatic parameters Hue Angle and Chroma, which were calculated based on the values obtained using the colorimeter. The CIELab model consists of the color brightness parameters L*, a*, and b*, where L* = 0 and L* = 100 are considered to be black and white, respectively. The parameter a* is used to represent negative values for green and positive values for red, while negative values for blue and positive values for yellow are displayed by b*.

The ranges of the L*, a*, b*, H*, and C* values were 38.36 ± 1.21 - 52.90 ± 1.486, 13.49 ± 0.358 - 21.82 ± 0.912, 4.87 ± 0.240 - 20.68 ± 0.738, 0.30 ± 0.023 - 0.84 ± 0.030, and 16.70 ± 0.951 - 28.69 ± 1.081, respectively (Table 5). All values of the CIE a* and CIE b* colorimetric parameters were positive, indicating that the studied samples fall within the red and yellow color range. By analyzing the mean values of the control samples, it can be observed that the luminosity value (L*) decreases with the addition of sea buckthorn oil, with lot M3S (injected with 5% sea buckthorn oil) showing the lowest value (38.36 ± 1.210) for this parameter among all the studied lots, while the control lot that was not injected with sea buckthorn oil (52.90 ± 1.486) obtained the highest value (Table 5). Following heat treatment, an increase in the luminosity of the samples is observed up to a value of 52.00 ± 0.626 (P3S) within the lot injected with 5% sea buckthorn oil.

The CIE a* value initially decreases in the lot injected with 1% sea buckthorn oil and not subjected to heat treatment (13.49 ± 0.358), compared to the control lot (C), which obtained a value of 15.97 ± 1.000. Subsequent values of the parameter a* for the lots that were not heat-treated reached a maximum value of 21.82 ± 0.912 for the lot injected with 5% sea buckthorn oil (M3S). Regarding the values for the heat-treated samples, there is also an increase in the a* parameter up to a maximum of 18.70 ± 0.668 in the lot injected with 5% sea

buckthorn oil and subjected to heat treatment (P3S), which is lower than that obtained in the lot injected with the same percentage of sea buckthorn oil but not subjected to heat treatment (M3S).

As with the CIE a* parameter, the CIE b* parameter also shows a continuous increase, up to the maximum value of 20.68 ± 0.738 in the pastrami lot injected with 5% sea buckthorn oil and subjected to heat treatment (P3S, Table 5). These continuous increases, directly proportional to the amount of sea buckthorn oil injected, indicate an intensification of the yellow color, which is characteristic of sea buckthorn oil. The intense color of this oil is due to its concentration of carotenoids (Koskovac et al., 2017). In the case of lots M1S and M2S (Table 5), a decrease in the b* parameter value is observed from 10.07 ± 0.516 to 9.75 ± 0.331, even though the latter lot was injected with 3% sea buckthorn oil, and the former with only 1%. However, in the lot injected with 5% sea buckthorn oil belonging to the same category (without heat treatment), a substantial increase in the average value of the b* parameter is observed (18.60 ± 1.316), a value lower than that for the heat-treated lots injected with 3% and 5% sea buckthorn oil, where values of 19.70 ± 0.552 and 20.68 ± 0.738 were obtained, respectively. From this, we can deduce a combined effect of heat treatment and the addition of sea buckthorn oil on the value of the CIE b* parameter. The results are similar to those obtained by Bobko et al. (2019) on minced pork meat to which sea buckthorn juice and oil, both organic, were added.

The Hue values for the untreated muscle vary slightly but fall within a relatively narrow range (0.53 ± 0.009 - 0.71 ± 0.043). These values are lower than those for the heat-treated muscle, which range from 0.83 ± 0.025 to 0.84 ± 0.030, indicating a different color hue induced by heat treatment. Although meat is a dead skeletal muscle tissue, it is not inert. The complex interactions between myoglobin (Mb) and biomolecules in the muscle's food matrix critically influence the internal color of heat-treated meat (Suman et al., 2016). The control sample has the lowest Hue value (0.30 ± 0.023), indicating a distinct hue compared to the other samples (Table 5).

Table 5. Chromatic parameters (Luminance L*, color coordinates a* and b*, Hue angle, and Chroma)

The tested samples	L*	a*	b*	Hue angle*	Chroma*
M1S	41.06 ± 0.775^{b}	13.49 ± 0.358^{a}	10.07 ± 0.516^{b}	0.64 ± 0.013^{c}	16.83 ± 0.582^{a}
M2S	39.88 ± 0.521^{ab}	16.88 ± 0.616^{bc}	9.75 ± 0.331^{b}	0.53 ± 0.009^{b}	19.50 ± 0.674^{b}
M3S	38.36 ± 1.210^{a}	21.82 ± 0.912^{e}	18.60 ± 1.316^{d}	0.71 ± 0.043^{d}	28.69 ± 1.081^{e}
p-value	**0.001 (***)**	**0.000 (***)**	**0.000 (***)**	**0.000 (***)**	**0.000 (***)**
P1S	46.68 ± 0.742^{c}	14.26 ± 0.374^{a}	15.52 ± 0.548^{c}	0.83 ± 0.014^{e}	21.08 ± 0.582^{c}
P2S	49.61 ± 0.996^{d}	17.84 ± 0.777^{cd}	19.70 ± 0.552^{de}	0.84 ± 0.030^{e}	26.59 ± 0.563^{d}
P3S	52.00 ± 0.626^{e}	18.70 ± 0.668^{d}	20.68 ± 0.738^{e}	0.83 ± 0.025^{e}	27.89 ± 0.712^{de}
p-value	**0.000 (***)**	**0.000 (***)**	**0.000 (***)**	0.870 (ns)	**0.000 (***)**
C	52.90 ± 1.486^{e}	15.97 ± 1.000^{b}	4.87 ± 0.240^{a}	0.30 ± 0.023^{a}	16.70 ± 0.951^{a}
Total p-value	**0.000 (***)**	**0.000 (***)**	**0.000 (***)**	**0.000 (***)**	**0.000 (***)**

Different letters on the same column indicate significant difference (estimated by ANOVA analysis and Tukey's test, $p \leq 0.05$).
M1S - pork tenderloin injected with 1% sea buckthorn oil, untreated thermally; M2S - pork tenderloin injected with 3% sea buckthorn oil, untreated thermally; M3S - tenderloin injected with 5% sea buckthorn oil, untreated thermally; P1S - tenderloin (pastrami) injected with 1% sea buckthorn oil, thermally treated; P2S - tenderloin (pastrami) injected with 3% sea buckthorn oil, thermally treated; P3S - tenderloin (pastrami) injected with 5% sea buckthorn oil, thermally treated.

The untreated muscle batches exhibit highly significant differences (p = 0.000***) in Chroma among the studied lots, with values of 16.83 ± 0.582 (lot M1S), 19.50 ± 0.674 (lot M2S), and 28.69 ± 1.081 (lot M3S) (Table 5). Similarly, highly significant differences (p = 0.000***) are observed among the studied lots that underwent heat treatment (Table 5), where the values were higher than those in untreated lots for batches P1S (1% sea buckthorn oil injection) and P2S (3% sea buckthorn oil injection), with values of 21.08 ± 0.582 and 26.59 ± 0.563, respectively. Batch P3S, with a mean Chroma value of 27.89 ± 0.712, injected with 5% sea buckthorn oil and subjected to heat treatment, is the only batch that did not exceed the value obtained by this parameter for the untreated batch with the same injection

percentage (M3S), which had a value of 28.69 ± 1.081 (Table 3).

The differences were highly significant ($p<0.001$) for all studied batches regarding all colorimetric parameters, except for the heat-treated batches (P1S, P2S, and P3S) for the Hue angle.

In the present study, the in vitro antimicrobial activity of sea buckthorn (Hippophae rhamnoides) oil against microbial activity was evaluated qualitatively and quantitatively by the presence or absence of colony-forming units on Petri plates. For this purpose, bacterial colonies were analyzed for the following species: *Staphylococcus aureus*, *Escherichia coli*, and *Salmonella*. Additionally, the total number of microorganisms in the samples was measured (Plate Count Agar). The statistical results show a significant diversity among the values for the same microbiological analysis, as evidenced by the highly significant differences (0.000***) identified between the analyzed batches (Table 6).

Table 6. The mean values ± SE of the batches of pork tenderloin injected with sea buckthorn oil regarding microbiological contamination

Batch	*Staphylococcus aureus* (log CFU/g)	*Escherichia coli* (log CFU/g)	Plate count agar (log CFU/g)	*Salmonella* (log CFU/g)
O	0.00 ± 0.000^{a}	0.00 ± 0.000^{a}	0.00 ± 0.000^{a}	*Absent*
C	4.17 ± 0.032^{g}	3.99 ± 0.031^{f}	4.12 ± 0.026^{f}	
M1S	3.55 ± 0.040^{de}	2.91 ± 0.055^{c}	3.58 ± 0.026^{b}	
M2S	3.63 ± 0.010^{ef}	3.10 ± 0.012^{e}	3.70 ± 0.015^{c}	
M3S	2.59 ± 0.032^{b}	2.59 ± 0.021^{b}	4.02 ± 0.015^{e}	
P1S	3.70 ± 0.040^{f}	3.47 ± 0.021^{f}	4.03 ± 0.025^{e}	
P2S	3.20 ± 0.030^{c}	2.97 ± 0.020^{cd}	3.94 ± 0.015^{d}	
P3S	3.48 ± 0.035^{d}	3.04 ± 0.032^{de}	4.02 ± 0.015^{e}	
p-value	**0.000 (***)**	**0.000 (***)**	**0.000 (***)**	-

Different letters on the same column indicate significant difference (estimated by ANOVA analysis and Tukey's test, $p \leq 0.05$).
M1S - pork tenderloin injected with 1% sea buckthorn oil, untreated thermally; M2S - pork tenderloin injected with 3% sea buckthorn oil, untreated thermally; M3S - tenderloin injected with 5% sea buckthorn oil, untreated thermally; P1S - tenderloin (pastrami) injected with 1% sea buckthorn oil, thermally treated; P2S - tenderloin (pastrami) injected with 3% sea buckthorn oil, thermally treated; P3S - tenderloin (pastrami) injected with 5% sea buckthorn oil, thermally treated.

By analyzing Table 6, we can observe that the sea buckthorn oil samples (O) evaluated for all four microbiological analyses showed no microbial contamination. These results can be explained by the fact that plants produce a variety of antimicrobial compounds to protect themselves from biotic attacks (Gupta et al., 2011). Furthermore, experiments conducted by other authors have demonstrated that sea buckthorn oil exhibits pronounced antimicrobial activity (Yue et al., 2017). However, even though sea buckthorn oil influenced the antimicrobial activity of the samples in this study, the test results did not show consistently lower or higher values but rather appeared chaotic, with both decreases and increases observed (Table 6). Even in this scenario, the batches injected with sea buckthorn oil still exhibited lower mean values of log CFU/g compared to those obtained for the control batch C, where 4.17 ± 0.032 log CFU/g was identified for *Staphylococcus aureus*, 3.99 ± 0.031 log CFU/g for *Escherichia coli*, and 4.12 ± 0.026 log CFU/g for the total germ count (Table 6).

The analysis of the presence of *Staphylococcus aureus* showed that the mean values ranged from 2.59 ± 0.032 to 3.70 ± 0.040 log CFU/g. The highest value (3.70 ± 0.040 log CFU/g) was observed in batch P1S, which represents one of the thermally treated batches, followed by the value of batch M2S, at 3.63 ± 0.010 log CFU/g, which is relatively close to the former. In the batches injected with 3% sea buckthorn oil, a slight decrease in microbial load was observed after thermal treatment, with a value of 3.20 ± 0.030 log CFU/g obtained, while the untreated batch showed significant differences ($p < 0.05$), with a value of 3.63 ± 0.010. Batches injected with 1% (M1S and P1S) and 5% (M3S and P3S) sea buckthorn oil showed higher values for *Staphylococcus aureus* after thermal treatment.

The microbiological tests for *Escherichia coli* showed the same trend as in the case of tests for *Staphylococcus aureus*, with reductions

observed before and after thermal treatment in the microbial load only in the batch injected with 3% sea buckthorn oil (M2S and P3S). The results for this bacterial species ranged from 2.59 ± 0.021 log CFU/g, identified in batch M3S, to 3.47 ± 0.021 log CFU/g (Table 6).

Regarding the results for Plate Count Agar, we observe that the highest values obtained in this analysis, aside from the control sample, were obtained by batches P3S (4.02 ± 0.015 log CFU/g), P1S (4.03 ± 0.025 log CFU/g), and M3S (4.02 ± 0.015 log CFU/g), as shown in Table 6. The results for two of these batches, P1S and P3S, positively correlate with the results of the tests for the identification of the bacterial species *Staphylococcus aureus* and *Escherichia coli*, as these batches exhibited among the highest values for these bacteria among the studied product batches injected with sea buckthorn oil.

Salmonella was absent in all the batches studied, a result that complies with the microbiological requirements set by Commission Regulation (EC) No 2073/2005, similar to the values of all microbiological tests conducted in this study.

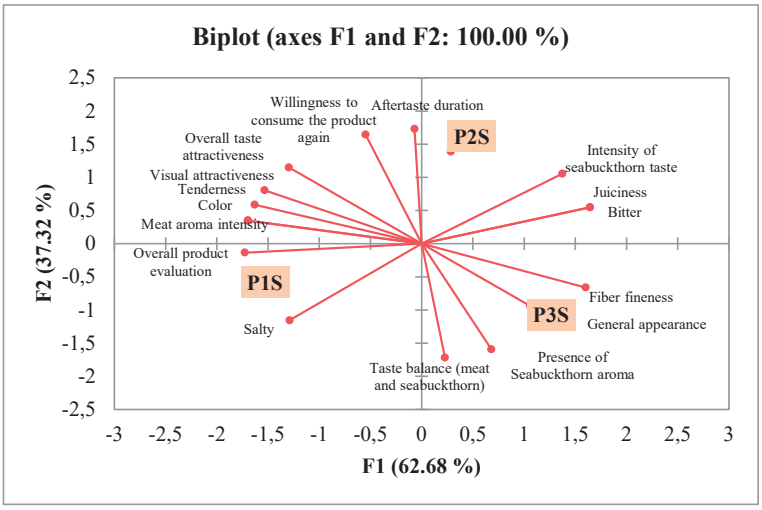


Figure 1. Representation of correlations between sensory traits using Principal Component Analysis (PCA)

For a comprehensive sensory characterization, we decided to perform a PCA graphic, considering the following characteristics: General appearance, Color, Visual attractiveness, Presence of Seabuckthorn aroma, Meat aroma intensity, Fiber fineness, Juiciness, Tenderness, Salty, Bitter, Intensity of seabuckthorn taste, Overall taste attractiveness, Taste balance (meat and seabuckthorn), Overall product evaluation, Willingness to consume the product again, Aftertaste duration (Figure 1). Figure 1 displays the products and attributes in a single plane, providing an overview of the relationships between products and attributes. PCA graph – principal component analysis involves evaluating the perception differences among the heat-treated batches in this study (batches P1S, P2S, and P3S). The biplot obtained through PCA analysis offers a clear view of the variations and relationships between the evaluated sensory characteristics and the batches of pork tenderloin injected with sea buckthorn oil that were heat-treated (pastrami).

By analyzing Figure 1, we can observe that the characteristics located in the upper-left quadrant are represented by Meat aroma intensity, Color, Tenderness, Visual attractiveness, Overall taste attractiveness, Willingness to consume the product again, and Aftertaste duration, which have negative contributions on axis F2 but positive contributions on axis F1. The positioning of these sensory characteristics suggests that product P2S (closest to these characteristics and positioned on the positive side of the Biplot) is perceived favorably by consumers in terms of aroma and appearance. However, it

also exhibits a higher intensity of seabuckthorn taste, greater juiciness, and a slightly bitter taste, as seen in batch P3S (the batch injected with 5% seabuckthorn oil). These latter characteristics, located in the upper-right quadrant, were perceived by consumers as positive.

Batch P1S, located in the bottom-left quadrant, is perceived as having certain negative characteristics. It is perceived as salty, even though its salt content, according to chemical analysis, was lower than in the other batches (Table 3), and it received a generally poor evaluation, meaning it obtained low scores for most of the evaluated characteristics. The perception of saltiness in the product could be due to its extremely low fat content, which fails to counterbalance this aspect.

Batch P3S, characterized primarily by a high degree of muscle fiber fineness, a pleasing overall appearance, the presence of seabuckthorn aroma, and, last but not least, a balanced taste between meat and seabuckthorn oil. However, it is less favored than batch P2S, which is preferred by consumers.

By analyzing the PCA results, we can observe that batch P2S (the batch injected with 3% seabuckthorn oil) emerged as the frontrunner in terms of consumer preferences. It was characterized by a strong seabuckthorn taste, juiciness, and an attractive appearance. Visual cues are often the first to be perceived and capable of influencing perception (Ciobanu et al., 2023). Batch P3S also received relatively high scores in the sensory analysis, similar to P2S. Panelists appreciated the balance of flavors and appearance in the muscle injected with 5% seabuckthorn oil (P2S). The only batch characterized negatively among all three was batch P1S (injected with 1% seabuckthorn oil), which received low scores in the sensory analysis and, consequently, an overall negative evaluation.

CONCLUSIONS

The analysis conducted on various batches of pork tenderloin reveals significant variations in physicochemical characteristics among them. Although some batches recorded higher values for certain parameters, there is no clearly superior batch in all aspects.

The analyses of the antioxidant capacity of pork tenderloin (psoas major) injected with sea buckthorn oil have shown positive effects of adding it to the analyzed samples. The most suitable method for determining antioxidant compounds, according to the results obtained, is ABTS. This method yielded values much higher than those obtained in the DPPH test, suggesting that the ABTS test is highly suitable for this type of food matrix.

By analyzing the parameters of the CIE*a*b* system, Hue angle, and Chroma, it can be observed that the addition of sea buckthorn oil positively influenced all colorimetric parameters, leading to their increase, especially in the case of samples subjected to heat treatment.

The microbiological analyses conducted indicate that the sea buckthorn oil injected into the pork tenderloin (psoas major) batches does not exert a strong antimicrobial effect, but it still has a slight positive influence, resulting in a minor reduction in microbial contamination.

Overall, our results indicate the valuable potential of *H. rhamnoides* in developing natural antimicrobial and antioxidant agents. Sensory-wise, the products were well received by consumers, especially the batches injected with 3% and 5% sea buckthorn oil. The batch injected with 1% sea buckthorn oil was negatively perceived by panelists due to low scores obtained for positive characteristics, resulting in an overall unfavorable evaluation. Panelists appreciated the presence of sea buckthorn flavor in the products and penalized the weak or imperceptible presence of it.

REFERENCES

Anchidin, B.G., Manoliu, D.R., Ciobotaru, M.C., Ciobanu, M.M., Gucianu, I., Sandu, G.A., & Boișteanu, P.C. (2023). Development of a Functional Meat Product with Sea Buckthorn Oil and Analysis of its Sensory and Physicochemical Quality. *Scientific Papers. Series D. Animal Science, 66*(1).

Blainski, A., Lopes, G.C., & De Mello, J.C.P. (2013). Application and analysis of the folin ciocalteu method for the determination of the total phenolic content from *Limonium brasiliense* L. *Molecules*, *18*(6), 6852-6865.

Bobko, M., Kročko, M., Haščík, P., Tkáčová, J., Bučko, O., Bobková, A., Čuboň, J., Češkovič, M., & Pavelkova, A. (2019). The effect of sea buckthorn (*Hippophae rhamnoides* L.) berries on parameters of quality raw cooked meat product. *Journal of*

microbiology, biotechnology and food sciences, 9(Special issue), 366-369.

Boișteanu, P.C., Manoliu, D.R., & Ciobanu, M. M. (2023). The Effect of Red Lentil Flour on the Quality Characteristics of Beef Burgers Obtained from Two Different Anatomical Regions. *Scientific Papers. Series D. Animal Science*, *66*(1).

Chaaban, H., Ioannou, I., Chebil, L., Slimane, M., Gérardin, C., Paris, C., ... & Ghoul, M. (2017). Effect of heat processing on thermal stability and antioxidant activity of six flavonoids. *Journal of food processing and preservation*, *41*(5), e13203.

Chauhan, A.S., Negi, P.S., & Ramteke, R.S. (2007). Antioxidant and antibacterial activities of aqueous extract of Seabuckthorn (*Hippophae rhamnoides*) seeds. *Fitoterapia*, *78*(7), 590-592.

Ciobanu, M.M., Manoliu, D.R., Ciobotaru, M. C., Postolache, A.N., Munteanu, M., & Boișteanu, P.C. (2022). Research on changes in pork quality parameters following different aging processes. *Scientific Papers Iasi University of Life Sciences, Series Agriculture*, https://repository.uaiasi.ro/xmlui/handle/20.500.12811/3241

Ciobanu, M.M., Manoliu, D.R., Ciobotaru, M.C., Anchidin, B.G., Matei, M., Munteanu, M., ... & Boișteanu, P.C. (2023). The Influence of Sensory Characteristics of Game Meat on Consumer Neuroperception: A Narrative Review. *Foods*, *12*(6), 1341.

Commission Regulation (EC) No 2073/2005 of 15 November 2005 on microbiological criteria for foodstuffs (Text with EEA relevance).

Dumitrașcu, L., Lanciu, A., & Aprodu, I. (2022). A preliminary study on using ultrasounds for the valorization of spent brewer's yeast. *The Annals of the University Dunarea de Jos of Galati. Fascicle VI-Food Technology, 46*(2), 141-153.

Echegaray, N., Munekata, P. E. S., Gullón, P., Dzuvor, C. K. O., Gullón, B., Kubi, F., & Lorenzo, J. M. (2022). Recent advances in food products fortification with anthocyanins. In Critical Reviews in Food Science and Nutrition (Vol. 62, Issue 6, pp. 1553–1567). *Taylor and Francis* Ltd. https://doi.org/10.1080/10408398.2020.1844141

Fu, L., Su, H., Li, R., & Cui, Y. (2014). Harvesting technologies for sea buckthorn fruit. *Engineering in Agriculture, Environment and Food*, *7*(2), 64–69. https://doi.org/10.1016/j.eaef.2013.10.002

Gucianu, I., Anchidin, B.G., Manoliu, D.R., Ciobotaru, M.C., Boișteanu, P.C., Postolache, A.N., & Ciobanu, M.M. (2023). Smoking Temperature Characteristics and Influence of Quality Indicators on Phytophagus Fillet (*Hypophthalmichthys molitrix*). *Scientific Papers. Series D. Animal Science*, *66*(2).

Gupta, S.M., Gupta, A.K., Ahmed, Z., & Kumar, A. (2011). Antibacterial and antifungal activity in leaf, seed extract and seed oil of seabuckthorn (*Hippophae salicifolia* D. Don) plant. *J. Plant Pathol. Microbiol.*, *2*(2), 1-4.

Hoffmann, M., Waszkiewicz-Robak, B., & Świderski, F. (2010). Functional Food of Animal Origin. Meat and Meat Products Functional food concept and strategies for functional products development. *Nauka Przyroda Technol Ogie*. http://www.npt.up-poznan.net

Ioannou, I., Chekir, L., & Ghoul, M. (2020). Effect of heat treatment and light exposure on the antioxidant activity of flavonoids. *Processes*, *8*(9), 1078.

Katan, M.B., & ROOS, N.M. (2004). Promises and problems of functional foods. Critical *Reviews in Food Science and Nutrition, 44*(5), 369-377.

Kausar, T., Hanan, E., Ayob, O., Praween, B., & Azad, Z. (2019). A review on functional ingredients in red meat products. *Bioinformation*, *15*(5), 358–363. https://doi.org/10.6026/97320630015358.

Koskovac, M., Cupara, S., Kipic, M., Barjaktarevic, A., Milovanovic, O., Kojicic, K., & Markovic, M. (2017). Sea buckthorn oil - A valuable source for cosmeceuticals. *Cosmetics*, *4*(4), 40.

Kumar, A., Kumar, S., Sharma, B. D., Mendiratta, S. K., Verma, O. P., & Patel, A. K. (2012). Functional meat and meat products: an overview. *Journal of Advanced Veterinary Research*, *2*(4), 314-324.

López-fernández, O., Domínguez, R., Pateiro, M., Munekata, P. E. S., Rocchetti, G., & Lorenzo, J. M. (2020). Determination of polyphenols using liquid chromatography–tandem mass spectrometry technique (LC–MS/MS): A review. *Antioxidants, 9(*6). https://doi.org/10.3390/antiox9060479.

Manoliu, D.R., Ciobanu, M.M., Ciobotaru, M.C., Postolache, A.N., Anchidin, B.G., & Boișteanu, P.C. (2023). Physico-Chemical And Sensory Evaluation Of Three Types Of Pork Mortadella Manufactured In The Iuls Meat Processing Microsection. *Scientific Papers. Series D. Animal Science, 66*(2).

Munteanu, I.G., & Apetrei, C. (2021). Analytical methods used in determining antioxidant activity: A review. *International journal of molecular sciences*, *22*(7), 3380.

Nishimura, T., Fang, S., Wakamatsu, J.I., & Takahashi, K. (2009). Relationships between physical and structural properties of intramuscular connective tissue and toughness of raw pork. *Animal Science Journal, 80*(1), 85-90.

Otero, A., Garci, M.L., & Moreno, B. (1998). Rapid microbiological methods in meat and meat products. *Meat science*, *49*, S179-S189.

Petrov, O.V., Hay, J., Mastikhin, I.V., & Balcom, B.J. (2008). Fat and moisture content determination with unilateral NMR. *Food research international, 41*(7), 758-764.

Pires, M.A., Munekata, P.E., Villanueva, N.D., Tonin, F. G., Baldin, J.C., Rocha, Y.J., ... & Trindade, M.A. (2017). The antioxidant capacity of rosemary and green tea extracts to replace the carcinogenic antioxidant (BHA) in chicken burgers. *Journal of Food Quality*, 2017.

Shah, R.K., Idate, A., & Poorva, V.U. (2021). Comprehensive review on sea buckthorn: Biological activity and its potential uses. *Pharma Innov, 10*, 942-953.

Song, H.S., Bae, J.K., & Park, I. (2013). Effect of heating on DPPH radical scavenging activity of meat substitute. *Preventive nutrition and food science, 18*(1), 80.

Suman, S.P., Nair, M.N., Joseph, P., & Hunt, M.C. (2016). Factors influencing internal color of cooked meats. *Meat Science*, *120*, 133-144.

Suryakumar, G., & Gupta, A. (2011). Medicinal and therapeutic potential of Sea buckthorn (*Hippophae rhamnoides* L.). *Journal of Ethnopharmacology, 138(*2), 268–278.

Turgut, S. S., Işıkçı, F., & Soyer, A. (2017). Antioxidant activity of pomegranate peel extract on lipid and protein oxidation in beef meatballs during frozen storage. *Meat Science, 129*, 111-119.

Ursache, F.M., Ghinea, I.O., Turturică, M., Aprodu, I., Râpeanu, G. & Stănciuc, N. (2017). Phytochemicals content and antioxidant properties of sea buckthorn (*Hippophae rhamnoides L.*) as affected by heat treatment – Quantitative spectroscopic and kinetic approaches. *Food Chemistry*, *233*, 442–449. https://doi.org/10.1016/j.foodchem.2017.04.107.

Ursache, F.M., Ghinea, I.O., Turturică, M., Aprodu, I., Râpeanu, G., & Stănciuc, N. (2017). Phytochemicals content and antioxidant properties of sea buckthorn (*Hippophae rhamnoides* L.) as affected by heat treatment–Quantitative spectroscopic and kinetic approaches. *Food Chemistry, 233*, 442-449.

Vinnikova, L., Synytsia, O., & Kyshenia, A. (2019). The Problems of Meat Products Thermal Treatment. *Food Science & Technology (2073-8684), 13*(2).

Wang, R., Zong, S.X., Yu, L.F., Lu, P.F., & Luo, Y.Q. (2014). Rhythms of volatile release from female and male sea buckthorn plants and electrophysiological response of sea buckthorn carpenter moths. *Journal of Plant Interactions*, *9*(1), 763–774.

Wu, C., Duckett, S. K., Neel, J. P. S., Fontenot, J. P., & Clapham, W. M. (2008). Influence of finishing systems on hydrophilic and lipophilic oxygen radical absorbance capacity (ORAC) in beef. *Meat Science*, *80*(3), 662–667.

Yang, B., & Kallio, H.P. (2001). Fatty acid composition of lipids in sea buckthorn (*Hippophae rhamnoides* L.) berries of different origins. *Journal of Agricultural and Food Chemistry*, *49*(4), 1939–1947.

Yu, T.Y., Morton, J.D., Clerens, S., & Dyer, J.M. (2017). Cooking-induced protein modifications in meat. Comprehensive Reviews in *Food Science and Food Safety, 16*(1), 141-159.

Yue, X.F., Shang, X., Zhang, Z.J., & Zhang, Y.N. (2017). Phytochemical composition and antibacterial activity of the essential oils from different parts of sea buckthorn (*Hippophae rhamnoides* L.). *Journal of food and drug analysis*, *25*(2), 327-332.

Zhishen, J., Mengcheng, T., & Jianming, W. (1999). The determination of flavonoid contents in mulberry and their scavenging effects on superoxide radicals. *Food chemistry, 64*(4), 555-559.