PAPER

APPLICATION OF ROSEMARY FOR THE PROLONGATION OF MICROBIAL AND OXIDATIVE STABILITY IN MECHANICALLY DEBONED POULTRY MEAT FROM CHICKENS

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ABSTRACT

In this study, we aimed to determine the effect of rosemary (*Rosmarinus officinalis* L.) preparations on microbial quality and oxidative stability in vacuum-packed mechanically deboned poultry meat (MDPM) from chickens stored at -18 °C for 4 months. We used MDPM originating from four production batches in which rosemary was added to in the form of dried spice (2.0%), extracts (2.0%) such as aqueous and ethanol (40 and 70% (v/v)), and essential oil (0.2%). MDPM control sample did not contain added rosemary. According to the results, the microbial quality of MDPM depended on the type of rosemary preparation used. Compared to the control sample, total bacterial count was considerably lower in samples with added essential oil and ethanol extract (70%, v/v). Essential oil was found to be the most effective in inhibiting psychrotrophic bacteria growth in vacuum-packed MDPM during storage. During the entire storage period, the use of rosemary preparations did not have a significant effect on the count of *Enterobacteriaceae*, but it significantly limited the growth of the coliform bacteria. Based on the index value of thiobarbituric acid reactive substances, rosemary preparations also showed, except for aqueous extract, a decrease in lipid oxidation in vacuum-packed MDPM from chickens stored at -18 °C for 4 months.

Keywords: antimicrobial effect, antioxidant effect, mechanically deboned poultry meat, rosemary, storage

1. INTRODUCTION

Mechanically deboned poultry meat (MDPM) obtained from chickens constitutes raw material commonly used in the meat industry, particularly in the production of homogenized products. The use of MDPM is justified for economic reasons as well as for the pursuit of rational usage of the elements of carcasses, which would be difficult to use otherwise (PIETRZAK *et al.*, 2011). The basic raw material to obtain MDPM from chickens is bones that remains from the deboning of the largest muscles (breast and thigh) and carcasses of chicken of lower quality (STANGIERSKI *et al.*, 2011).
The MDPM production, storage, and processing conditions have been established in Regulation (2004). Despite the continuous improvements in methods and machines used to obtain MDPM, Polish and European poultry industries most commonly use high-pressure methods for the production of MDPM, which is destructive for the bone structure (NAGY *et al.*, 2007; BOTKA-PETRAK *et al.*, 2011; BEŁKOT *et al.*, 2013). This leads to the lower stability of the raw material upon storage than hand-trimmed or machine-trimmed chicken meat. This poor stability is primarily due to the high level of fragmentation and aeration during production, which contributes to a higher susceptibility toward oxidation processes and an increase in the growth of microflora (GRABOWSKI and KIJOWSKI, 2004; MICHALSKI and POMYKAŁA, 2008).
In a situation of inability for immediate use of MDPM in processing, the raw material is stored in a frozen state (GRABOWSKI and KIJOWSKI, 2004). Addition of natural substances from plant origin, exhibiting antimicrobial and antioxidant effect, constitutes an additional factor in prolonging the stability of meat and meat products during storage. However, the latest literature search (SHAH *et al.*, 2014) reveals that the majority of the studies of the effectiveness of plant preparations on the stability of meat products during storage involves mammal meat and its products. Study results demonstrate that inter alia rosemary preparations may be used to minimize the oxidative changes in meat and meat products such as aged beef (COLLE *et al.*, 2016), raw pork batters (HERNÁNDEZ-HERNÁNDEZ *et al.*, 2009), fresh (GEORGANTELIS *et. al.*, 2007) and thermally processed pork sausage (SEBRANEK *et al.*, 2005), wiener (CORONADO *et al.*, 2002) and bologna sausages (VIUDA-MARTOS *et al.*, 2010), frankfurters (ESTÉVEZ and CAVA, 2006), and reduced nitrite liver pâtés (DOOLAEGE *et al.*, 2012). Due to the application of rosemary preparations, microbial quality of different meat products can be improved, inter alia in modified atmosphere-packaged fresh pork and vacuum-packed ham slices (ZHANG *et al.*, 2009), and in the African fresh sausage (MATHENJWA *et al.*, 2012). However, there is little information on the application possibilities of plant preparations for the prolongation of storage stability of MDPM (HASSAN and LAM SWET FAN, 2005; HAĆ-SZYMAŃCZUK *et al.*, 2014) and its products (MOHAMED and MANSOUR, 2012; JIRIDI *et al.*, 2015).
Rosemary (*Rosmarinus officinalis* L.), from the *Lamiaceae* family, is a plant with both strong antioxidant and antimicrobial activities. It is used in food in the form of fresh or dried leaves, essential oil, and aqueous and alcoholic extracts from leaves (ERKAN *et al.*, 2008; HAĆ-SZYMAŃCZUK *et al.*, 2010). Various studies have demonstrated that the complex biologically active substances of rosemary have an inhibitory effect on a wide spectrum of bacteria, including *Enterococcus faecalis*, *Staphylococcus aureus*, *Staphylococcus epidermidis*, *Bacillus subtilis*, and *Klebsiella pneumoniae* (DIMITRIJEVIC *et al.*, 2007; ZHANG *et al.*, 2009; HAĆ-SZYMAŃCZUK *et al.*, 2010). As an alternative to synthetic antioxidants, rosemary preparations have been used in food processing (BALENTINE *et al.*, 2006; VELASCO and WILLIAMS, 2011).
In this study, we aimed to determine the effect of rosemary (*R. officinalis* L.) on lipid oxidation and microbial quality of MDPM from chickens stored at -18 °C for 4 months.

The results of this study may contribute to the understanding of the innovative methods of MDPM preservation.

2. MATERIALS AND METHODS

We used MDPM that was obtained from a production plant in northeast Poland. MDPM was prepared using high-pressure separation method on breast muscle scraps of broilers. The chilled MDPM (6 kg) was collected from the wholesalers in Warsaw and transported to the Division of Food Biotechnology and Microbiology of the Faculty of Food Sciences under refrigerated conditions. Rosemary (*R. officinalis* L.) was added to the MDPM in dried form ("Kamis," McCormic, Stefanowo, Poland) and as an aqueous extract, ethanol extracts, and essential oil (own production) under laboratory conditions. In the following parts of the paper, they will be called rosemary preparations.

Each of the batches of MDPM from chickens was analyzed for fat, protein, and water content. The determination of these chemical components in MDPM was performed in accordance with the requirements of the Association of Official Analytical Chemists (AOAC, 2007). We used a FoodScan™Lab near-infrared spectrometer (Foss Analytical A/S, Hillerød, Denmark) working in the spectral range of 850-1050 nm and using a calibration based on the artificial neural network model.

The aqueous extract and ethanol extracts from dried rosemary ("Kamis," McCormic, Stefanowo, Poland) were obtained via continuous extraction in a Soxhlet apparatus (a universal extraction system B-811, Büchi Labortechnik AG, Flawil, Switzerland). The extraction process parameters were established in the preliminary study (results unpublished). For the preparation of each extract, 40 g of dried rosemary was distributed onto 8 extraction thimbles (5 g per thimble). Distilled water and ethyl alcohol with 40 and 70% (v/v) concentration were, respectively, used as solvents. The raw material in each thimble was extracted with 150 mL of the appropriate solvent for 15 cycles, maintaining the boiling point of a solvent. The portions obtained from each extract were combined, resulting in approximately 550 mL of raw extracts. The raw extracts were filtered using 180-μm thick filter paper (Whatman GE, LaboPlus Sp. z o.o., Warsaw, Poland). Subsequently, each extract was concentrated in a rotary evaporator (Rotovaporator R-205; Büchi Labortechnik AG) until approximately 40 g of the extract was left, corresponding to the weight of dried rosemary used to obtain the extract.

To obtain essential oil from rosemary, the method of BIAŁECKA-FLORIAŃCZYK and WŁOSTOWSKA (2007) was followed. Around 30 g of fresh rosemary leaves were crumbled and covered with 400 mL of water. This was subjected to distillation in a Deryng apparatus by Simax until essential oils were obtained. The chilled distillate was four times extracted using dichloromethane in a separatory funnel. Then, water was removed by adding anhydrous magnesium sulfate. The obtained extract was concentrated in a rotary evaporator (Rotovaporator R-205). The solvent was evaporated at a temperature of 30 °C and at a pressure of 540-560 hPa.

The chemical composition of the aqueous extract, ethanol extracts, and essential oil from rosemary was analyzed for the identification and determination of chemical compounds.

The determination of volatile compounds in essential oil was performed by gas chromatography (GC) equipped with flame ionization detector (FID) (Perkin Elmer, Autosystem XL) based on the literature (BURT, 2004; DJEDDI *et al.*, 2007). The following parameters were used for separation: HP-5 column (30 m × 0.32 mm × 0.25 μm), helium as a carrier gas (3 cm^3/min), split mode (1:100) for sample injection, injection temperature at 270 °C, and FID temperature at 300 °C. The following program of oven temperature was used: initial temperature 35 °C/5 min, 30 °C/min temperature increase up to 60 °C

followed by 6 °C/min to 200 °C and 30 °C/min until a temperature of 280 °C was achieved.
The identification and determination of the amount of selected chemical compounds in rosemary extracts was performed based on the literature (LONGARAY DELAMARE *et al.*, 2007, TAWAHA *et al.*, 2007). In this study, we performed high performance liquid chromatography (HPLC) using Agilent 1200 liquid chromatography coupled with diode array detector (DAD). Zorba Eclipse XDB C18 (4.6 × 150 mm) column was used with the following parameters: 5 μl injection volume, 0.8 cm^3/min flow rate, and UV detection at a wavelength of 210 and 325 nm. Separation was performed in gradient elution with two eluents: A-acetonitrile and B-0.05% trifluoroacetic acid. Data were analyzed using EZ Elite Chrome program.
In each experimental series, six samples of MDPM were prepared (each weighing 1 kg), differing in the type of rosemary preparation added: Control-sample without addition of rosemary, D-2.0% addition of dried rosemary, WE-2.0% addition of aqueous extract from rosemary, E40-2.0% addition of 40% (v/v) ethanol extract from rosemary, E70-2.0% addition of 70% (v/v) ethanol extract from rosemary, and EOS-0.2% addition of essential oil from rosemary.
The amount of rosemary preparations added to MDPM was established based on the recommendations of the producer or based on the literature (GEORGANTELIS *et al.*, 2007). After MDPM samples were thoroughly mixed with rosemary preparations, each sample was divided into four portions (250 g each) and vacuum-packed in plastic bags (PE/PA, thickness 75 μm) using a vacuum machine C200 (Multivac Sepp Haggenmüller GmbH & Co. K.G., Wolfertschwenden, Germany). These samples were stored at a temperature of -18° C for 4 months. After each month, microbial analyzes were performed and TBARS index values were determined for all MDPM samples including the control sample. Before the analyzes, each sample was defrosted (+4 °C, 4 h) without opening the packaging. Moreover, directly after the delivery of the raw material to the laboratory, the same determinations were carried out only on the MDPM control sample.
The microbial analyzes were conducted following Polish Standard (PN-EN ISO, 2005). They include the determination of the total bacteria count (TBC) (PN-EN ISO, 2013), number of psychrotrophic bacteria (PN-ISO, 2004), *Enterobacteriaceae* (PN-ISO, 2005), coliform bacteria (PN-ISO, 2007), and *Salmonella* spp. (PN-EN ISO, 2003). The number of bacteria was expressed as $\log_{10}$ colony forming units per gram (log CFU/g). TBARS was determined using the extraction method of PIKUL et al. (1989). TBARS index value was expressed in milligram of malondialdehyde per kilogram of sample (mg MAD/kg).
The experiment was repeated four times, preparing MDPM samples from different production batches. The statistical analysis of the results was performed using Statistica version 10.0 program (2011). The significance was tested using one-way analysis of variance (ANOVA) and Tukey's honest significant difference (HSD) test at a significance level of α=0.05.

3. RESULTS

In this study, the analysis of chemical composition of rosemary preparations revealed that they differed in the chemical profile. Each of them consisted of a complex mixture of different substances. The results of an earlier work (HAĆ-SZYMAŃCZUK *et al.*, 2015) demonstrated that the dominating compounds of the aqueous extract of rosemary were rosmarinic, ferulic, and chlorogenic acids. In the ethanol extracts of rosemary, the major compounds present were rosmarinic acid, carnosol, and ferulic acid (Table 1), whereas in

the essential oil, the major compounds present were camphor, borneol, and R(+)limonene (Table 2).

Table 1. Chemical composition of extracts from rosemary.

Chemical compound	Retention time (min)	Ethanol extract (40%, v/v)	Ethanol extract (70%, v/v)
		Concentration (mg/cm^3)	
Chlorogenic acid	2.83	0.068	0.108
Epicatechin	3.80	nd	0.002
Caffeic acid	4.33	0.012	0.026
Rutoside	5.83	0.060	0.104
p-coumaric acid	7.04	0.006	0.020
Ferrulic acid	8.08	0.112	0.166
Benzoic acid	11.87	0.012	0.018
Rosemarinic acid	12.70	4.006	5.756
Myricetin	12.80	nd	0.002
Resveratrol	15.51	nd	0.002
Quercetyn	18.69	0.006	0.012
Carnosol	25.73	0.468	0.178
Curcumin	26.03	nd	0.026

nd – not detected

Table 2. Chemical composition of essential oil from rosemary.

Chemical compound	Retention time (min)	Concentration (mg/cm^3)
α-pinene	7.88	0.20
β-pinene	8.79	0.10
Myrcene	9.21	0.22
1,4-cineole	9.72	0.10
p-cymene	9.92	0.53
R(+) limonene	10.13	22.47
γ-terpinene	10.71	0.75
Linalol	11.70	1.33
Camphor	12.26	51.87
Borneol	13.15	27.90
Carvone	14.93	0.44
Bergamol	15.23	0.18
Thymol	15.99	0.11
Carwacrol	16.20	6.70
Eugenol	17.42	0.92
β-caryophyllene	18.73	0.62

Based on the results of microbial analysis of MDPM during storage (Figs. 1-4), it was found that the tested rosemary preparations showed different antimicrobial activity. During the storage period, TBC was found to be highest in the control sample (Fig. 1). In the EOS, E70, and E40 samples, a reduction in TBC was observed from 2 months of

storage. After 4 months of storage, the EOS and E70 samples were characterized by significantly ($p \leq 0.05$) lower TBC than control sample. Of all the tested rosemary preparations, essential oil was found to be the most efficient in inhibiting the growth of psychrotrophic microorganisms (Fig. 2). EOS significantly lowered the number of microorganisms compared to control, D, and WE after 4 months of storage. In each of the examined MDPM samples, a significantly ($p \leq 0.05$) higher *Enterobacteriaceae* bacterial count was found after 2 months of storage (Fig. 3). The use of rosemary preparations did not significantly ($p > 0.05$) influence the count of *Enterobacteriaceae* in the MDPM samples during the entire storage period. In each of the examined MDPM samples, coliform bacteria was also detected (Fig. 4). However, in comparison to the control sample, addition of rosemary preparations to MDPM significantly ($p \leq 0.05$) restricted the growth of coliform bacteria during the entire storage period. *Salmonella* spp. was not determined in any of the examined MDPM samples.

The fat, protein, and water content in MDPM from chickens was on average 15.93, 15.72, and 66.31%, respectively.

Based on TBARS index values (Fig. 5.), the addition of rosemary preparations to MDPM had an influence on the course of oxidative changes in lipids. Among the tested preparations, the weakest antioxidant activity was exhibited by the aqueous rosemary extract. However, other rosemary preparations significantly ($p \leq 0.05$) slowed down the processes of lipid oxidation in the MDPM samples during the storage period. Our results also demonstrated that for EOS and E70 samples, the TBARS index value after 4 months of storage was significantly ($p \leq 0.05$) lower than the TBARS index value after 1 month of storage.

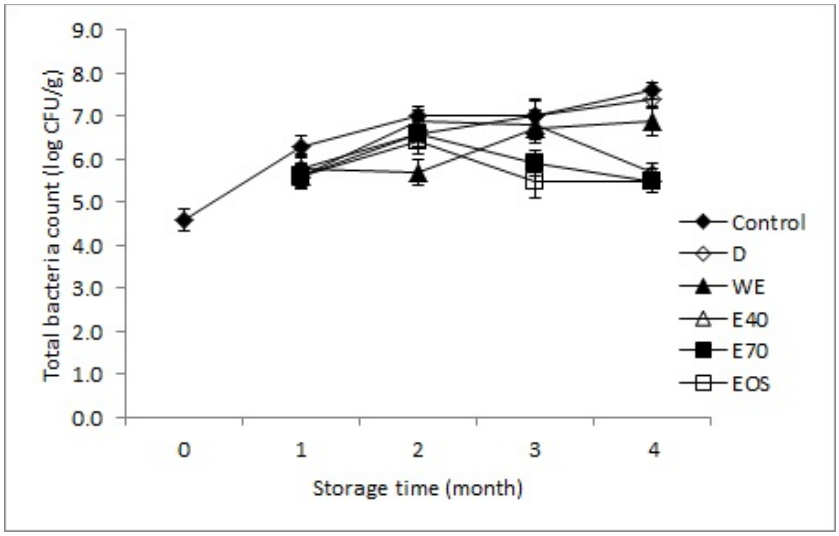

Figure 1. Effect of addition of rosemary preparations on the total bacteria count in mechanically deboned poultry meat (MDPM) from chickens stored at -18 °C. Notes: Control-control sample, without addition of rosemary; D-2.0% addition of dried rosemary; WE-2.0% addition of aqueous extract from rosemary; E40-2.0% addition of 40% (v/v) ethanol extract from rosemary; E70-2.0% addition of 70% (v/v) ethanol extract from rosemary; EOS-0.2% addition of essential oil from rosemary.

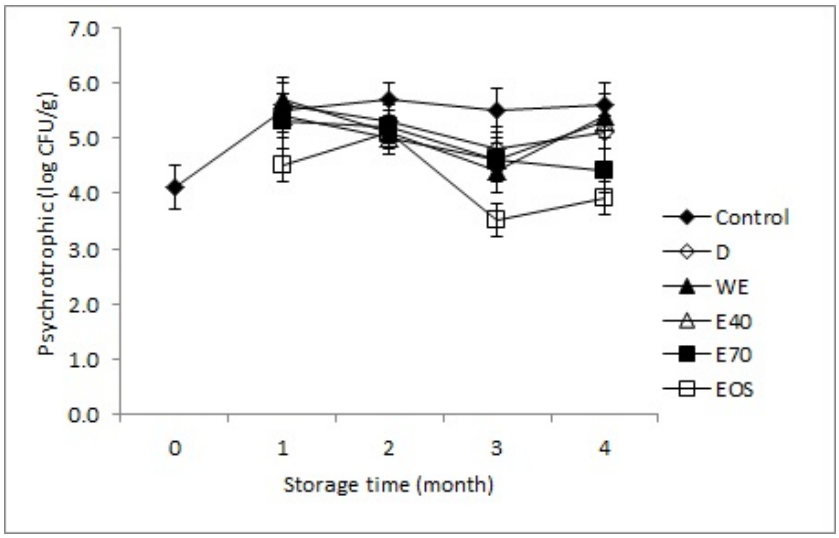

Figure 2. Effect of addition of rosemary preparations on the number of psychrotrophic bacteria in mechanically deboned poultry meat (MDPM) from chickens stored at -18 °C. Notes: Control-control sample, without addition of rosemary; D-2.0% addition of dried rosemary; WE-2.0% addition of aqueous extract from rosemary; E40-2.0% addition of 40% (v/v) ethanol extract from rosemary; E70-2.0% addition of 70% (v/v) ethanol extract from rosemary; EOS-0.2% addition of essential oil from rosemary.

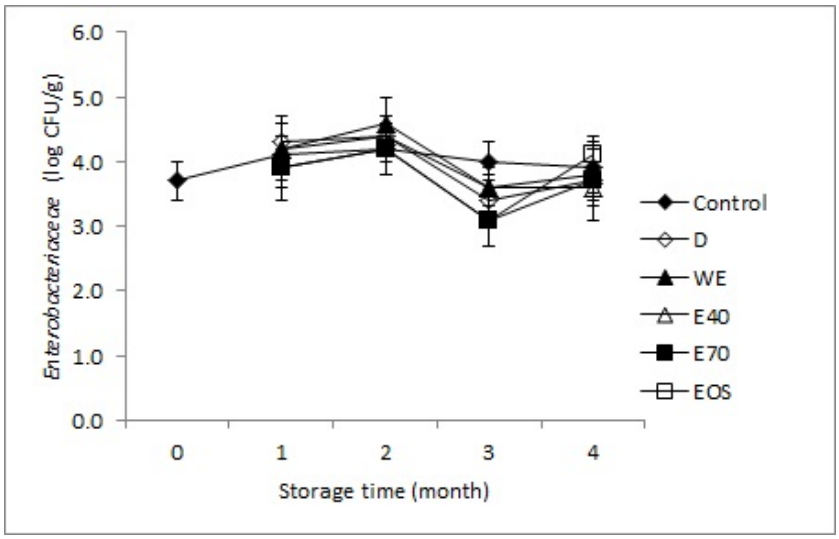

Figure 3. Effect of addition of rosemary preparations on the number of *Enterobacteriaceae* bacteria in mechanically deboned poultry meat (MDPM) from chickens stored at -18 °C. Notes: Control-control sample, without addition of rosemary; D-2.0% addition of dried rosemary; WE-2.0% addition of aqueous extract from rosemary; E40-2.0% addition of 40% (v/v) ethanol extract from rosemary; E70-2.0% addition of 70% (v/v) ethanol extract from rosemary; EOS-0.2% addition of essential oil from rosemary.

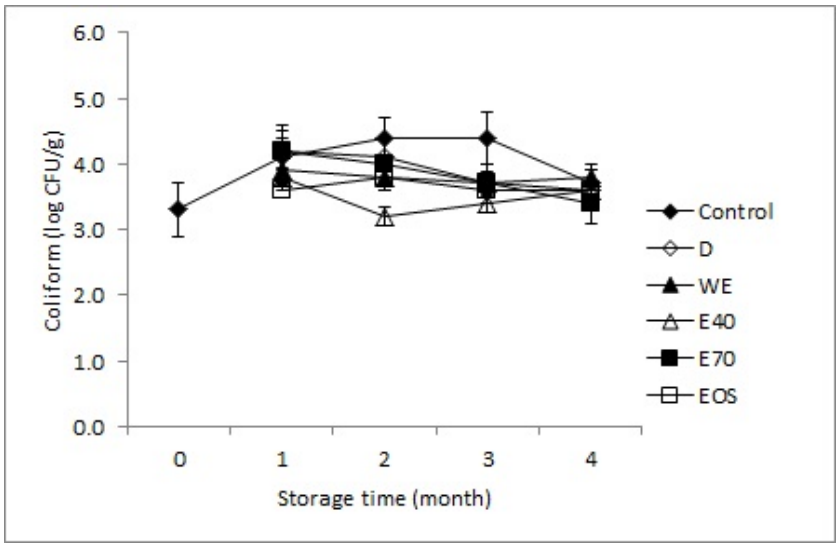

Figure 4. Effect of addition of rosemary preparations on the number of coliform bacteria in mechanically deboned poultry meat (MDPM) from chickens stored at -18 °C. Notes: Control-control sample, without addition of rosemary; D-2.0% addition of dried rosemary; WE-2.0% addition of aqueous extract from rosemary; E40-2.0% addition of 40% (v/v) ethanol extract from rosemary; E70-2.0% addition of 70% (v/v) ethanol extract from rosemary; EOS-0.2% addition of essential oil from rosemary.

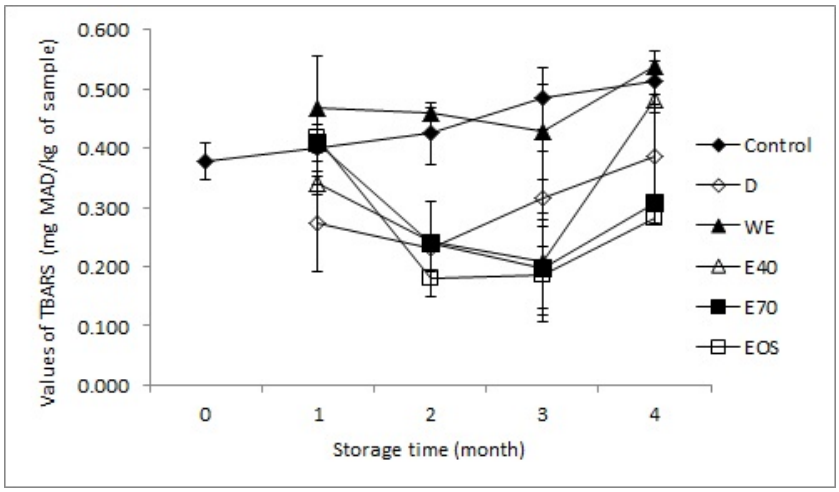

Figure 5. Effect of addition of rosemary preparations on thiobarbituric acid reactive substances (TBARS) index value in mechanically deboned poultry meat (MDPM) from chickens stored at -18 °C. Notes: Control-control sample, without addition of rosemary; D-2.0% addition of dried rosemary; WE-2.0% addition of aqueous extract from rosemary; E40-2.0% addition of 40% (v/v) ethanol extract from rosemary; E70-2.0% addition of 70% (v/v) ethanol extract from rosemary; EOS-0.2% addition of essential oil from rosemary.

4. DISCUSSION

The results of this study showed that MDPM from chickens contained a high amount of fat (15.93%). However, it was in compliance with the requirements of non-compulsory Polish Standard (PN, 1992), which states that MDPM from burring poultry should not contain fat more than 20%, protein less than 12%, and water more than 75%. The fat present in MDPM is susceptible to oxidation, which might be due to the presence of the unsaturated fatty acids and phospholipids along with the catalytic effects of heme iron

(GRABOWSKI and KIJOWSKI, 2004; PIETRZAK *et al*., 2011; BEŁKOT *et al*., 2013). Since lipid oxidation is the major cause of quality loss in MDPM, in our opinion the application of rosemary preparations as sources of natural antioxidants seems to be interesting option for preserving the shelf life of this raw material.

The raw material used in this study met the food safety criteria with respect to *Salmonella* and aerobic bacteria and *E. coli* count as specified in Commission Regulation (2005). More discussion in this area is difficult because the available literature lacks the information on antimicrobial effect of rosemary extracts in MDPM.

HAĆ-SZYMAŃCZUK *et al.* (2009) and OKOH *et al.* (2010) have found that the antimicrobial efficiency of rosemary extracts varies, which could be attributed to the type and method of its preparation (e.g., distillation and extraction). This could be due to the different chemical profiles of these preparations. In their study, OKOH *et al.* (2010) found that rosemary oil obtained by solvent free microwave extraction exhibited stronger inhibitory effect on the examined bacteria (*Staphylococcus aureu*s, *E. coli*, *Bacillus subtilis*, and *Klebsiella pneumoniae*) in comparison to the oil obtained through hydro-distillation. However, HAĆ-SZYMAŃCZUK *et al.* (2009) found that rosemary oil as well as aqueous extract did not inhibit the growth of *S. aureus* and *K. pneumoniae* on Mueller-Hinton Agar medium.

ROMANO *et al.* (2009) found that the addition of rosemary leaf extract limited the growth of *E. coli.* According to the authors the minimal inhibitory concentration (MIC) of this extract was 105 μg/mL. They found a stronger antimicrobial activity than benzoic acid and butylated hydroxytoluene (BHT), whose concentration was 250 μg/mL. However, based on the comparative study of antimicrobial properties of essential oils from *Lamiaceae* plants, ŽIŽOVIC *et al.* (2009) found that the minimum inhibitory concentration (MIC) for *Escherichia* and *Salmonella* bacteria was above 1250 μg/mL.

Similar studies on the use of rosemary preparations, solely or mixed with other components of plant origin, have demonstrated efficiency in inhibiting the growth of microflora in meat and meat products. According to ABDEL-HAMIED *et al.* (2009), a significant inhibition of psychrotrophic microorganisms in minced meat stored at 4 °C and -18 °C was obtained by using a mixed addition of rosemary and salvia extracts. According to them, the addition of 0.05% of extracts to the meat stored at 4 °C for 10 days reduced the number of psychrotrophic microbes from 31.64 log CFU/g in the control to 14.12 log CFU/g in the extract sample. For meat stored at -18 °C for 100 days, the bacterial count was 7.16 and 20.31 log CFU/g, respectively, for the extracts and control samples.

ZHANG *et al.* (2009) studied antimicrobial activity of 14 different extracts toward pathogenic bacteria causing pork meat spoilage such as *Listeria monocytogenes*, *E. coli*, *Pseudomonas fluorescens*, and *Lactobacillus sake*. According to their results, the modified-atmosphere-packed meat stored at 4 °C for 28 days demonstrated the effectiveness of combination of rosemary and liquorice extracts as a natural preservative, which significantly inhibited the growth of the studied microorganisms.

According to PHAM *et al.* (2013), a mixture of rosemary extract (addition level: 2000 ppm) and green tea extract (100–300 ppm) can be used to limit the growth of psychrotrophic bacteria in raw pork sausage stored at -20 °C for 6 months. According to MATHENJWA *et al.* (2012), the use of plant extracts and chitosan in the production of traditional South African pork and beef sausage can lower or eliminate the addition of sulfur dioxide (SO_2) as a preservative. They found that the combination of rosemary extract (addition level: 260 mg/kg), chitosan (10 mg/kg), and SO_2 (100 mg/kg) or rosemary extract with chitosan had an equally efficient antimicrobial effect in sausages as SO_2 (250 mg/kg).

The results of microbiological quality evaluation of MDPM obtained in this study confirm the bacteriostatic properties of rosemary formulations. Literature data indicate, however,

that some of the active substances present in these preparations may exhibit bactericidal effect. Thymol and carvacrol are chemical compounds whose mechanism of action on bacterial cells has been most comprehensively evaluated so far. The presence of these compounds has been confirmed in rosemary oil used in this study. Their mechanism of action on Gram-negative bacteria is based on the disintegration of the cell membrane, by releasing lipopolysaccharides (LPS) and increasing the permeability of the plasma membrane for adenosine triphosphate (ATP), the loss of which ultimately leads to cell death (HELANDER *et al*., 1998).

In case of Gram-positive bacteria, carvacrol interacts with the cell membrane, changing its permeability toward H+ and K+ cations. Change in the gradient of these cations causes disruption of the basic processes in cell and ultimately leads to cell death. In Gram-positive bacteria, increase in membrane permeability toward ATP is not observed as for Gram-negative bacteria (ULTEE *et al*., 2002).

The present literature does not provide information on the antioxidant activity of rosemary extracts in MDPM, and the majority of studies concern the storage stability of slaughtered mammal meat and its products (HERNÁNDEZ-HERNÁNDEZ *et al*., 2009; WÓJCIAK *et al*., 2011; PHAM *et al*., 2013; ARMENTEROS *et al*., 2016).

WÓJCIAK *et al*. (2011) compared the antioxidant activity of aqueous extracts from different plants added to pork meat and found that after 30 days of storage under refrigeration conditions, the highest efficiency was observed in rosemary extract. However, HERNÁNDEZ-HERNÁNDEZ *et al*. (2009) recommend the addition of alcoholic extract of rosemary based on the study performed on model pork batters to slow down the lipid oxidation processes. According to them, the strong antioxidant property of the extract might be due to high concentration of carnosic acid and carnosol and the presence of numerous other active components. The antioxidant activity of rosemary extracts was also confirmed by PHAM *et al*. (2013) on raw pork sausage and by MATHENJWA *et al*. (2012) on pork and beef sausage, which were stored in a frozen state for 180 and 100 days, respectively.

According to MIELNIK *et al*. (2003), the use of commercial rosemary preparations may also constitute an alternative method for the improvement of oxidative stability and prolongation of MDPM from turkey upon storage. To obtain a satisfactory quality of vacuum-packed raw material stored at -25 °C for 7 months, an individual selection of the type and amount of rosemary preparation is necessary, which complies with our results.

In contrast to the above-cited literature, COLLE *et al*. (2016) reported that the use of rosemary extract together with ascorbic acid did not significantly limit the processes of lipid oxidation in beef steaks in comparison to the control product. SZCZEPANIK (2007) conducted a comparative study of antioxidant activity of extracts from dill, coltsfoot, rosemary, horsetail, salvia, and thyme in the breast muscle of chickens and turkeys during a 6-month frozen storage (-25 °C). The author also found that none of the used extracts significantly slowed down the oxidation process of lipids contained in the chicken muscles.

Although we did not evaluate it in this work, the use of natural preservatives of plant origin could be helpful in controlling the oxidation of other ingredients that exhibit nutritional value in meat products. NIETO *et al*. (2013) explored the mechanisms behind the protection of protein against oxidation by natural plant antioxidants. The oxidative stability of the meat proteins in pork patties was evaluated as loss of thiols and as formation of myosin cross-links. Essential oil of rosemary was found to have an antioxidative effect on protein thiol loss. Furthermore, protein disulfide cross-link formation was inhibited in pork patties with added essential oil of rosemary. These and other properties of rosemary preparations are due to a large range of chemical compounds.

Both the results of the analysis of the chemical composition of rosemary preparations obtained by the authors of this study and those presented by other researchers (BURT, 2004; DJEDDI *et al*., 2007) demonstrate that these preparations are mixtures of many different compounds. According to DJEDDI *et al*. (2007) the chemical profile of preparations from rosemary depends not only on the methods of obtaining them but also on the habitat of plants. When reviewing the literature, the authors concluded that the climatic differences between south Europe and North Africa Mediterranean areas might have a significant impact on the content of ingredients such as 1,8-cineole, α-pinene, and camphor in rosemary essential oil. KASPARAVIČIENE *et al*. (2013) reported that ethanol extracts of rosemary contain primarily three groups of compounds: phenolic diterpenes, flavonoids, and phenolic acids. ABRAMOVIČ *et al*. (2012), among the dominant phenolic diterpenes of these extracts, mentioned-after COUVELIER *et al*. (1996) carnosol, carnosic acid, methyl carnosate, and phenolic acids from caffeinic and rosmarinic acid.

5. CONCLUSIONS

Our results indicate that the addition of rosemary preparations constitute an auxiliary factor in the preservation of MDPM from chickens stored in a frozen state for 4 months. The tested preparations differed in their chemical composition and antimicrobial and antioxidant activities. The addition of 0.2% essential oil and 2.0% of 70% (v/v) ethanol extract was the most efficient in restricting the growth of microflora and inhibiting lipid oxidation in MDPM from chickens.

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