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Korrespondenzadresse:
anelef@ap.teithe.gr

Summary

Zusammenfassung

¹Laboratory of Meat Technology, Department of Animal Production, School of Agricultural Technology, Alexander Technological Educational Institute, Thessaloniki, Greece;

²Laboratory of Hygiene and Technology of Food of Animal Origin, Veterinary Research Institute of Thessaloniki, National Agricultural Research Foundation, Greece

Changes in lipid oxidation and fatty acid composition in pork and poultry meat during refrigerated and frozen storage

Änderungen der Lipidoxidation und der Fettsäurezusammensetzung im Schweinefleisch und im Geflügelfleisch während der Kühlung und der Gefrierkühlung

A. Eleftheriadou¹, G. Samouris², M. Ioannidou², E. Kasapidou¹

This experiment studied changes in lipid oxidation and fatty acid composition in pork (*m. longissimus dorsi*) and chicken breast (muscle and skin tissue) samples during refrigerated (4 °C) and frozen storage (–18 °C). Pork samples were analysed after 0, 1, 3, 7, 10 and 14 days of refrigerated storage and chicken samples were analysed after 0, 1, 2, 5 and 7 days of refrigerated storage. Pork and chicken samples were analysed after 15, 30, 90 and 180 days of frozen storage.

Lipid oxidation increased ($P < 0.05$) during refrigerated and frozen storage in pork muscle samples. Refrigerated storage did not affect oxidation levels in chicken muscle samples whereas there was an increase ($P < 0.05$) in chicken skin samples. Oxidation levels increased significantly ($P < 0.05$) during frozen storage for both skin and muscle samples.

The levels of monounsaturated fatty acids of pork and chicken breast skin samples increased ($P < 0.05$) during refrigerated storage. In chicken breast muscle samples the levels of saturated fatty acids increased and the levels of polyunsaturated fatty acids decreased under refrigeration. Frozen storage resulted in decreases in the levels of saturated fatty acids in pork ($P < 0.05$) and chicken breast ($P < 0.05$) muscle samples. The levels of polyunsaturated fatty acids increased ($P < 0.05$) under frozen storage conditions in pork muscle samples. Slight differences were observed only in the levels of saturated and monounsaturated fatty acids in frozen stored chicken breast skin samples.

Keywords: storage, lipid oxidation, fatty acid composition, pork meat, poultry meat

Diese Studie untersucht Veränderungen der Lipidoxidation und der Fettsäurezusammensetzung in Schweinefleischproben (*M. longissimus dorsi*) und Brustproben (Muskelgewebe und Hautgewebe) von Masthühnern während einer Kühlung (4 °C) und einer Gefrierlagerung (–18 °C). Die gekühlten Schweinefleischproben wurden nach 0, 1, 3, 7, 10 und 14 Tagen Lagerung analysiert, die gekühlten Masthühnerproben wurden nach 0, 1, 2, 5 und 7 Tagen Lagerung untersucht. Nach 15, 30, 90 und 180 Tagen wurden die tiefgefrorenen Schweinefleisch- und Masthühnerproben analysiert.

In den Schweinefleischproben erhöhte sich die Lipidoxidation ($P < 0.05$) während der Kühlung und der Gefrierlagerung. Die Kühlung beeinflusste die Oxidation in den Masthühner-Muskelproben nicht, während es in den Masthühner-Hautproben eine Oxidations-Zunahme ($P < 0.05$) zu verzeichnen gab. Während der Gefrierlagerung erhöhte sich die Lipidoxidation signifikant ($P < 0.05$) in den Haut- und Muskelproben der Masthühner.

Die Konzentration der einfach ungesättigten Fettsäuren in den Schweinefleischproben und Masthühner-Hautproben erhöhte sich ($P < 0.05$) während der Kühlung. In den Masthühner-Muskelproben erhöhte sich der Gehalt an gesättigten Fettsäuren während der Kühlung, der Gehalt an mehrfach ungesättigten Fettsäuren nahm gleichzeitig ab. Durch die Gefrierkühlung nahm der Gehalt an gesättigten Fettsäuren in den Muskelproben des Schweinefleisches ($P < 0.05$) und der Masthühner ($P < 0.05$) ab. Die Konzentrationen der mehrfach ungesättigten Fettsäuren erhöhten sich ($P < 0.05$) unter Gefrierkühlungsbedingungen in den Schweinefleischproben. Geringfügige Unterschiede wurden nur in den Gehalten von gesättigten und einfach ungesättigten Fettsäuren der gefrorenen Masthühner-Hautproben beobachtet.

Schlüsselwörter: Lagerung, Lipidoxidation, Fettsäuremuster, Schweinefleisch, Geflügelfleisch

tor, Denmark). The fatty acid methyl esters were prepared with boron trifluoride in methanol solution. An appropriate quantity of the extracted fat was saponified by the addition of NaOH in methanol followed by heating at 100 °C for 15 minutes. Fatty acid methyl ethers were prepared by incubation at 100 °C for 5 minutes in boron trifluoride methanol reagent. The produced fatty acid methyl esters were extracted by the addition of hexane after vigorous agitation and washing with a saturated solution of potassium hydroxide (Christie, 2003). The fatty acid methyl esters were analyzed using a HP 5890 (Hewlett-Packard) gas chromatograph equipped with flame ionisation detector and a DB-23 (60 m x 0.25 mm x 0.25 µm) column (J & W Scientific, Inc., Folsom, California, USA). The GC (Hewlett-Packard, model 5890) conditions were: carrier gas He; packed mode injection; injector and flame ionisation detector temperatures 250 °C and 280 °C respectively; initial oven temperature 50 °C for 1 min, increased at 25 °C per min to 175 °C, increased at 4 °C per min to 230 °C, held at 230 °C for 5 min.

A 37 component mixture (Supelco, Bellefonte, Pennsylvania, USA) of fatty acids methyl esters (FAME) was used as a reference standard. The mixture was purchased as a 100-mg neat mixture, containing C4–C24 FAMES (2%–4% relative concentration). Fatty acids methyl esters were identified by comparing their retention times with the retention times of the FAME mixture. Fatty acids were quantified by peak area measurement and the results are expressed as percent (%) of the total fatty acids present in the sample.

Statistical analyses

One-way analysis of variance (ANOVA) was used to analyse differences between treatments. Post-hoc analysis was undertaken using Tukey's test at a 5% level of significance. Statistical software package SPSS version 13.0 (2004) for Windows (SPSS, Chicago, IL, USA) was used.

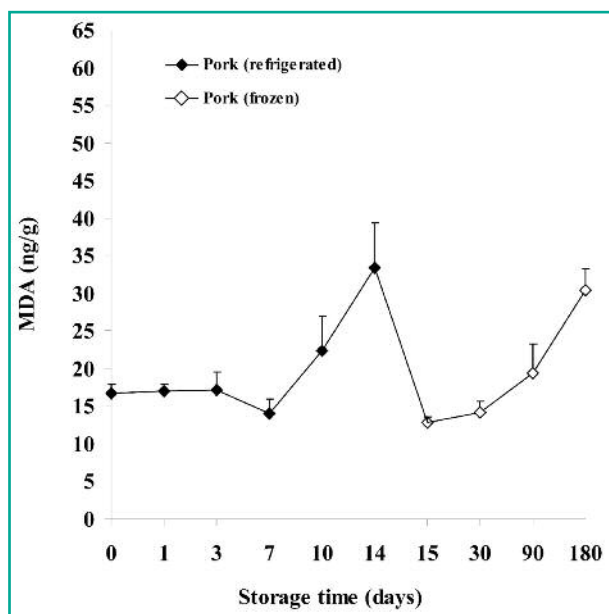


FIGURE 1: Changes in the levels of malondialdehyde in pork samples during storage at 4 °C and at –18 °C.

Results and discussion

Lipid oxidation

Lipid oxidation in pork samples during refrigerated and frozen storage is presented in Figure 1. Lipid oxidation increased ($P < 0.05$) as the storage period was extended in both conditions. Lipid oxidation remained virtually unchanged during the first 10 days of refrigerated storage whereas at day 14 there was a 2 fold increase in the concentration of MDA than the values initially observed (days 0–7). Regarding frozen storage, lipid oxidation was almost stable for the first 90 days. At 180 days of storage lipid oxidation increased 1.8–2.3 times than the values observed during the first 90 days of storage.

Lipid oxidation changes for chicken muscle and skin samples under refrigerated and frozen storage are illustrated in Figure 2. Lipid oxidation was overall higher in skin tissue due to its higher content of unsaturated and particularly polyunsaturated fatty acids (Tab. 1). Refrigerated storage did not affect oxidation in chicken muscle samples whereas there was a change ($P < 0.05$) in chicken skin samples. Chicken muscle samples were very stable to lipid oxidation unlike chicken skin samples that lipid oxidation exhibited a 3.6 times increase between refrigeration days 0 and 7.

Oxidation changes during frozen storage were statistically significant ($P < 0.05$) for both skin and muscle samples. Lipid oxidation in both muscle and skin increased with storage time. The changes in lipid oxidation levels were also more profound in skin samples than muscle samples. At 180 days of frozen storage lipid oxidation was 1.8 and 2.8 times higher in muscle and skin samples in relation to lipid oxidation at 15 days of frozen storage.

Regarding changes in lipid oxidation during refrigerated storage, the reported results are also supported by Park et al. (2008) that investigated lipid oxidation during refrigerated storage (8 °C) of air packed pork m. *longissimus dorsi* samples and reported similar changes in the MDA values. Min et al. (2008) reported a similar pattern for the

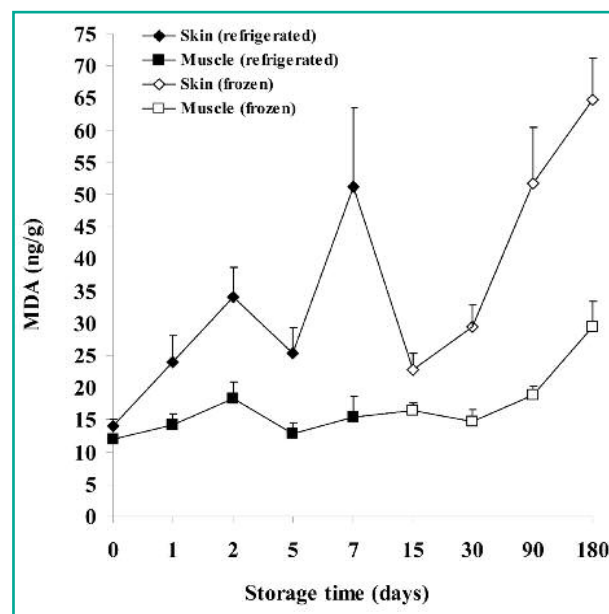


FIGURE 2: Changes in the levels of malondialdehyde in chicken breast (muscle and skin tissue) samples during storage at 4 °C and at –18 °C.

TABLE 1: Fatty acid composition (mean \pm standard deviation) of pork muscle, chicken skin and chicken breast muscle on storage day 0 (control samples).

Fatty acid	Pork (n = 6)	Chicken muscle (n = 6)	Chicken skin (n = 6)
14:0 Myristic	2.40 \pm 0.965	0.85 \pm 1.547	0.69 \pm 0.122
16:0 Palmitic	25.97 \pm 0.900	25.75 \pm 0.456	26.81 \pm 1.692
16:1 n-7 Palmitoleic	3.55 \pm 0.603	3.61 \pm 0.749	3.98 \pm 0.692
18:0 Stearic	12.21 \pm 3.238	7.13 \pm 0.845	8.12 \pm 1.280
18:1 n-9 Oleic	40.16 \pm 4.605	31.86 \pm 2.529	29.03 \pm 3.050
18:2 n-6 Linoleic	6.89 \pm 2.308	24.86 \pm 0.303	25.54 \pm 2.541
18:3 n-3 α -Linolenic	1.33 \pm 1.516	1.82 \pm 1.547	0.21 \pm 0.135
20:0 Arachidic	ND ¹	ND ¹	0.41 \pm 0.471
20:1 n-9 Eicosenoic	0.38 \pm 0.380	ND ¹	0.27 \pm 0.092
20:4 n-6 Arachidonic	ND ¹	ND ¹	0.16 \pm 0.085
22:0 Behenic	ND ¹	ND ¹	0.07 \pm 0.113
22:6 n-3 Docosahexanoic (DHA)	ND ¹	ND ¹	0.00 \pm 0.008
SFA	41.91 \pm 5.893	33.73 \pm 1.808	36.09 \pm 3.397
MUFA	44.09 \pm 5.228	35.47 \pm 0.728	33.28 \pm 2.649
PUFA	6.89 \pm 2.308	26.68 \pm 2.823	25.91 \pm 2.630

SFA: saturated fatty acids; MUFA monounsaturated fatty acids; PUFA polyunsaturated fatty acids; ND¹: not detectable.

development of lipid oxidation in ground patties of pork loin and chicken breast samples stored in air at 4 °C for 7 days.

The levels of polyunsaturated fatty acids were much higher in chicken muscle than pork muscle samples (Tab. 1). Despite differences in the levels of polyunsaturated fatty acids, pork and chicken muscle samples were equally susceptible to oxidation during refrigerated and frozen storage. Min et al. (2008) reported no proportional correlation between lipid oxidation and the content of polyunsaturated fatty acids in raw chicken thigh meat during storage. The same workers suggested that the oxidative stability of meat may not be proportionately related to its content in polyunsaturated fatty acids.

Lipid oxidation was far below reported threshold values for the detection of rancidity by expert taste panellists. The extent of lipid oxidation is usually reported with the term TBARS or TBA number which describes the thiobarbituric acid reactive substances. Malonaldehyde is the main substance reacting with the thiobarbituric acid. TBA number within the same animal species, though, are influenced by a number of factors such as animal nutrition i. e. dietary antioxidants and dietary fat content and type, whether the meat is raw, cooked or processed, the type of muscle, the type of taste panel i. e. trained or untrained and the method used for the determination of the TBA number. Different workers have reported different values for the detection of rancidity in sensory evaluation tests. Melton (1983) reported that oxidized flavours were detectable at TBA numbers in the range 0.3–1.0 mg malondialdehyde/kg tissue in beef or pork and 1.0 or 2.0 mg malondialdehyde/kg tissue in chicken. Tarladgis et al. (1960) reported that TBA values in the range of 0.6–2.0 mg malondialdehyde/kg tissue correspond to the threshold value for detection of rancidity in pork. According to O'Neill et al. (1998) the threshold value for warmed over flavour detection in cooked, dark chicken meat could be set at TBA values equal or higher than 0.8 mg of malondialdehyde/kg tissue. Warmed over

flavour and rancid flavour are closely correlated (Poste et al., 1986; Byrne et al., 2001). Moreover, detection of lipid oxidation is not related to acceptability of the meat by the panellists and the relevant TBA numbers should not be considered as a general reference number (Melton, 1983; Boles and Parish, 1990; Fernández et al., 1997).

Fatty acid composition

Fatty acid composition of pork longissimus dorsi muscle, chicken skin and chicken breast muscle for storage day 0 (control) is shown in Table 1. In all samples palmitic (16:0), oleic (18:1 n-9) and linoleic (18:2 n-6) acid were the major fatty acids in the saturated, monounsaturated and polyunsaturated lipid class respectively.

Refrigerated storage did not affect the levels of saturated, monounsaturated and polyunsaturated fatty acids in pork samples (Fig. 3). In general, after 14 days of storage the levels of monounsaturated fatty acids increased slightly. Morcuende et al. (2003) reported similar changes in the composition of neutral lipids in m. *longissimus dorsi* from free range reared Iberian pigs stored under

light in aerobic conditions at 4 °C for 10 days. Monin et al. (2003) reported statistically significant changes ($P < 0.05$) in the proportions of saturated fatty acids and no changes in the proportions of monounsaturated and polyunsaturated fatty acids in m. *longissimus lumborum* from Large White and Piétrain pigs that were packed under vacuum and stored at 3–4 °C for 9 days. In the latter study, the levels of polyunsaturated fatty acids were elevated in samples from both breeds compared to our study. These differences are probably linked to the genetic type of animals used in the study. Ramírez and Cava (2008) reported that refrigerated

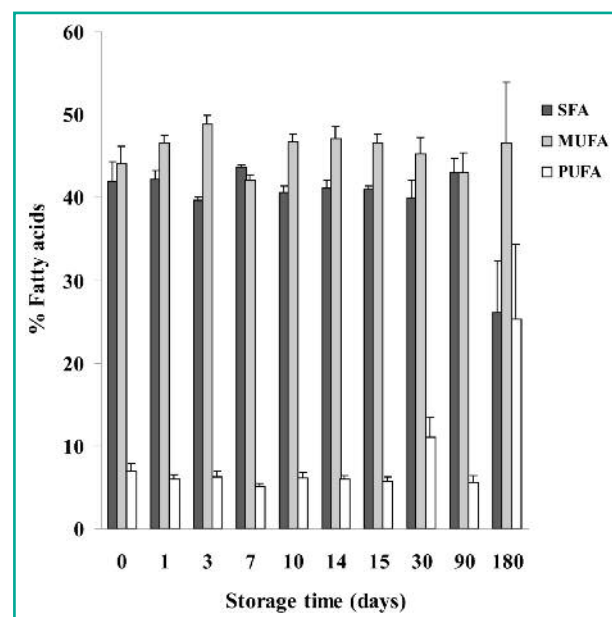


FIGURE 3: Changes in the percentages of fatty acid lipid classes during refrigerated and frozen storage of pork samples (SFA saturated fatty acids; MUFA monounsaturated fatty acids; PUFA polyunsaturated fatty acids).

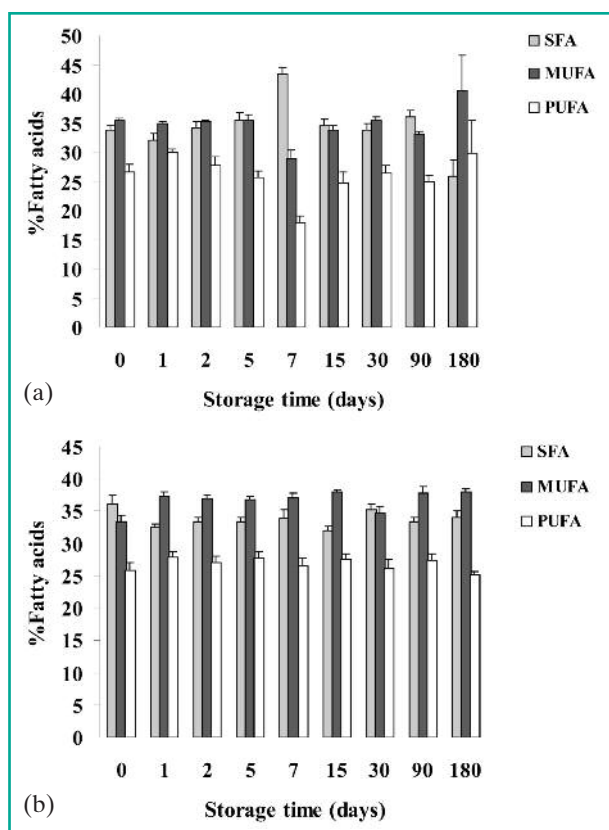


FIGURE 4: Changes in the percentages of fatty acid lipid classes during refrigerated and frozen storage of chicken breast (a) and skin (b) samples (SFA saturated fatty acids; MUFA monounsaturated fatty acids; PUFA polyunsaturated fatty acids).

storage did not change the fatty acid composition of *m. longissimus lumborum* from three different Iberian x Duroc genotype pigs stored under light in aerobic conditions at 4 °C for 10 days. Despite intrinsic differences in the fatty acid composition of intramuscular fat between different breeds, changes in the levels of saturated, monounsaturated and polyunsaturated fatty acids followed the same pattern in the studies reported above.

Frozen storage resulted in a decrease ($P < 0.05$) in the levels of saturated fatty acids and an increase ($P < 0.05$) in the levels of polyunsaturated fatty acids. The levels of monounsaturated fatty acids were not affected (Fig. 3). Hernández et al. (1999) reported no changes in the levels of saturated, monounsaturated and polyunsaturated fatty acids in the neutral lipid fraction of vacuum packed pork *m. longissimus dorsi* during frozen storage for 6 months. Approximately 80 % of total lipids consist of neutral lipids. Polyunsaturated fatty acids are mainly present in the phospholipid fraction. In the previous study there was a decrease in the proportion of polyunsaturated fatty acids after 6 months of storage whereas in our study an increase in the proportion of polyunsaturated fatty acids was observed.

The fatty acid composition of chicken breast muscle changed during refrigerated storage whereas there was no change in the fatty acid composition of chicken breast skin samples (Fig. 4). Chicken muscle fatty acid composition did not change during the first 5 days of refrigerated storage. Changes were observed on storage day 7 in the levels of all lipid classes.

Frozen storage resulted in a decrease ($P < 0.05$) in the levels of saturated fatty acids (Fig. 4) in breast muscle. Noticeable changes occurred after 90 days of storage. Conchillo et al. (2004) reported a similar pattern in the changes of fatty acid composition in chicken breast samples stored under vacuum for either 6 days at 4 °C or 90 days at -18 °C. Regarding chicken skin samples, frozen storage did not affect the levels of polyunsaturated fatty acids. There were significant fluctuations ($P < 0.05$) in levels of both saturated and monounsaturated fatty acids during storage.

Reported reasons for the changes in the fatty acid composition during refrigerated and frozen storage are the activities of lipolytic enzymes whose stability is affected by storage temperature (Rhee et al., 1996; Min et al., 2008) and storage duration (Hernández et al., 1999; Alasnier et al., 2000). Variations of the fat content and subsequently the fatty acid composition within the same muscle, as it has been reported by Cameron and Enser (1991) and Lawrie and Ledward (2006) for pork *m. longissimus dorsi*, might also affect fatty acid composition during storage. It should be also mentioned that changes in the fatty acid composition and the percentages in particular of the mono-unsaturated and polyunsaturated fatty acids reflect also individual variations in the animals based on their diets. In the present study, samples were randomly collected from the abattoirs and thus there is no available information about the animals (slaughter age, breed) and their diets.

Conclusions

Differences in lipid oxidation and fatty acid composition were observed during refrigerated and frozen storage of pork and chicken meat. The development of lipid oxidation was rather limited in all meat types and under both storage conditions. Chicken breast skin was most susceptible to lipid oxidation compared to pork and chicken meat. During refrigerated storage constant changes in the fatty acid composition were observed only in chicken muscle samples. Fatty acid composition was fairly stable in pork muscle and chicken skin samples. Frozen storage influenced fatty acid composition at various degrees in all tissues. Fatty acid composition was fairly stable for the first 90 days of frozen storage and any changes became apparent at 180 days of storage.

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Address for correspondence:

Dr. Anastasia Eleftheriadou
A.T.E.I. of Thessaloniki
School of Agricultural Technology
Department of Animal Production
P.O BOX 141
GR-574 00 Thessaloniki, Greece
anelef@ap.teithe.gr