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INTRAMUSCULAR FAT CONTENT IN THE MASSETER MUSCLE (*MUSCULUS MASSETER*) OF CATTLE DEPENDING ON SEX AND AGE

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Abstract. The objective of the experiment was to determine the amount of intramuscular fat (IMF) in the masseter muscle (*musculus masseter*) of slaughter cattle depending on sex and age. Subjects were 40 cattle (7 cows older than 5 years, 12 heifers and 21 bulls aged around 24 months). After slaughter, muscle samples were collected and frozen in liquid nitrogen. The frozen samples were cut into 10 μm sections on a cryostat. Next, the sections were placed on a glass slide and stained with Oil Red to determine the intramuscular fat content. The study showed that the masseter muscle in the studied population of slaughter cattle had a relatively low proportion of intramuscular fat (1.84% on average). IMF level was highest in heifers (2.26%) and lowest in bulls (1.48%) ($P \leq 0.05$). The age of the experimental animals had no significant effect on intramuscular adipose tissue content in the masseter muscle.

Key words: slaughter cattle, masseter muscle, intramuscular fat

INTRODUCTION

The nutritive value of beef meat varies mainly depending on the content and composition of protein and fat. Carcass composition, in particular the amount of fat in the carcass depends on the type of animal as well as its genetic potential, sex, age and nutrition. In the case of beef, it is desirable that the carcasses are low in external and intermuscular fat. This does not apply to the intramuscular fat, the amount of which has a significant effect on the quality and sensory attributes of

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meat. Culinary beef has less than 5% intramuscular fat. Intramuscular fat contains about 44% saturated fatty acids (SFA), 46% monounsaturated fatty acids (MUFA) and 10% polyunsaturated fatty acids (PUFA) [Ślusarczyk and Strzetelski 2006, Kołczak 2008].

The masseter muscle is a very strong and massive muscle with a multipennate arrangement of fibres. It covers the outer surface of the ramus of the mandible and the adjoining body of the mandible at the height of molar teeth, reaching from the zygomatic arch and facial crest to the outer surface of the body of the mandible). The masseter consists of two muscle layers – superficial (pars superficialis) and deep (pars profunda) – which differ in muscle fibre orientation (the fibres are placed at an angle of 90°). The function of the masseter muscle is to elevate the mandible and connects it to the upper jaw; in herbivores, including cattle, a unilateral contraction of this muscle allows lateral movements of the mandible, which are important during mastication [Akajewski 1997, Janowicz 1999, Krysiak et al. 2001]. Other than the function played during an animal's lifetime, the masseter is valued as a culinary delicacy. Once considered slaughterhouse waste, today it is hugely popular and is served in many restaurants as beef cheeks, prepared in different ways.

The aim of the study was to analyse the amount of intramuscular fat in the masseter muscle (*musculus masseter*) of slaughter cattle depending on sex and age.

MATERIAL AND METHODS

The experiment used 40 Polish Holstein-Friesian cattle: 12 heifers, 21 bulls and 7 cows. At slaughter, the age of the animals was about 24 months for heifers and bulls, and over 5 years for cows.

The animals, which originated from individual farms, were slaughtered in a meat processing plant located in the Pomorskie province. Immediately after slaughter (within 45 minutes), samples of the masseter muscle were collected, chilled and transported to the laboratory of the Department of Animal Biochemistry and Biotechnology of the University of Technology and Life Sciences in Bydgoszcz. The collected muscle samples were frozen in liquid nitrogen (–196°C) and stored until analysis. Frozen samples were cut into 10 μm sections using a cryostat (Thermo Scientific, Waltham, USA) at –25°C. Next, the sections were mounted on a glass slide and stained with the fat-soluble dye Oil Red to determine the content of intramuscular fat (which stains red).

The microscopic images were saved on a computer disk using an Optek UB-200 microscope equipped with a ToupCamTM digital camera. Multiscan v. 18.03 image analysis software (Computer Scanning Systems II, Warsaw, Poland) was

used to measure the percentage of intramuscular fat in a 3 mm^2 area of the masseter muscle.

The results were subjected to two-way analysis of variance using Statistica PL [2014].

$$Y_{ij} = \mu + a_i + b_j + e_{ij};$$

where:

μ – overall mean,

a_i – effect of i-th sex (female, male),

b_j – effect of j-th age (about 24 months, more than 5 years),

e_{ij} – random error of observation.

Arithmetic mean ($\bar{x}$) and standard deviation (SD) were calculated. Significant differences between the groups were determined with Tukey's HSD test for unequal numbers.

RESULTS AND DISCUSSION

The percentage of intramuscular fat in the masseter muscle of animals from the analsyed groups averaged 1.48% for bulls, 2.14% for cows and 2.26% for heifers (Table 1). The means between the heifer and bull groups differed significantly ($P \leq 0.05$).

Table 1. Intramuscular fat content (%) determined histochemically in masseter muscle of different groups of cattle

Tabela 1. Udział tłuszczu śródmięśniowego oznaczanego histochemicznie (%) w mięśniu żwaczu w poszczególnych grupach bydła

Group – Grupa	Intramuscular fat, % – Tłuszcz śródmięśniowy, %	
Heifers (n = 12) – Jałówki (n = 12)	$\bar{x}$	2.26[a]
	SD	2.21
Bulls (n = 21) – Buhajki (n = 21)	$\bar{x}$	1.48[a]
	SD	1.38
Cows (n = 7) – Krowy (n = 7)	$\bar{x}$	2.14
	SD	1.62
On average – Średnio	$\bar{x}$	1.84
	SD	1.71

[a] – Significance at $P \leq 0.05$.
[a] – Istotność przy $P \leq 0{,}05$.

Trela et al. [2004] point to 1.5% as the minimal level of intramuscular fat content, below which the meat as a raw material becomes unattractive for the consu-

mer. In animals with weight gains of 640–767 g/day, Groth et al. [1999] obtained 1.18% fat in the Black-and-White breed (BW) and 0.87% fat in BW×Charolais crossbreds.

According to Młynek [2011], the proportion of intramuscular fat in bovine muscles should be around 3%. The author compared the quality of meat from BW bulls (less than 25% HF inheritance) and commercial crossbreds obtained from Charolais, Limousin, Blonde d'Aquitaine, Simmental and Piemontese bulls. The essence of the study was to compare the relationships between growth intensity of the animals, their fat content and culinary quality of their meat (based on three muscles: *biceps brachii*, *semimembranosus* and *longissimus lumborum*). The results of meat quality showed a significantly higher content of intramuscular fat in Black-and-White bulls (3.77%) compared to commercial crossbreds (2.42%) in the group of animals with low growth rate (771 g per day). In the groups with high growth intensity (941 g per day), the content of intramuscular adipose tissue in bull muscles was similar at 2.93% (BW) and 2.75% (commercial crossbreds). Similar results were obtained by Zin [1995], who reported that in the bulls with daily gains of 744 to 801 g, the proportion of intramuscular fat ranged from 2.40 to 2.81%.

In turn, Graham et al. [2009] demonstrated that intramuscular fat content in crossbreds varied depending on breed of the sire used for mating. The proportion of intramuscular fat in the longissimus dorsi muscle was 1.5% for animals sired by a Belgian Blue bull, 1.9% for offspring sired by a Limousin bull, 2.8–3% for progeny sired by an Aberdeen Angus bull, and 3.5% for crossbreds from a Japanese Wagyu bull. These results suggest that the different percentages of fat in muscles are due to origin and breed (genotype) of the studied animals.

A study by Gotoh et al. [2009], which compared the composition and quality of carcasses from the Wagyu breed and European breeds (Belgian Blue, Angus, Holstein-Friesian) at the age of 24 months, showed very large differences in the content of intramuscular adipose tissue. In the longissimus dorsi muscle, it was as much as 23.3% in the Japanese cattle and ranged from 0.6 to 4.7% in the European breeds of cattle. The authors attributed these differences to the specific characteristics of each breed and the system of feeding. In relation to the above findings, our results for the percentage of intramuscular fat are similar to those obtained for the European breeds of cattle.

The data in Table 2 indicate that females (heifers and cows) were characterized by a greater proportion of intramuscular fat (2.21%) in comparison with males (1.48%). The effect of sex proved statistically significant ($P \leq 0.05$).

Considering the age of the studied animals (Table 3), it is concluded that older animals (cows) had a greater percentage of intramuscular fat than younger ani-

Table 2. Intramuscular fat content (%) determined histochemically in masseter muscle depending on sex

Tabela 2. Udział tłuszczu śródmięśniowego oznaczanego histochemicznie (%) w mięśniu żwaczu w zależności od płci

Sex – Płeć	Intramuscular fat, % – Tłuszcz śródmięśniowy, %	
Females (n = 19) – Samice (n = 19)	$\bar{x}$	2.21[a]
	SD	1.97
Males (n = 21) – Samce (n = 21)	$\bar{x}$	1.48[a]
	SD	1.38
On average – Średnio	$\bar{x}$	1.84
	SD	1.71

[a] – Significance at $P \leq 0.05$.
[a] – Istotność przy $P \leq 0,05$.

mals (heifers and bulls). The means calculated for this trait were 1.77 and 2.14%, respectively, with a non-significant difference.

Table 3. Intramuscular fat content (%) determined histochemically in masseter muscle depending on age

Tabela 3. Udział tłuszczu śródmięśniowego oznaczanego histochemicznie (%) w mięśniu żwaczu w zależności od wieku

Age – Wiek	Intramuscular fat, % – Tłuszcz śródmięśniowy, %	
24 months (n = 33) – 24 miesiące (n = 33)	$\bar{x}$	1.77
	SD	1.75
5 years (n = 7) – 5 lat (n = 7)	$\bar{x}$	2.14
	SD	1.62
On average – Średnio	$\bar{x}$	1.84
	SD	1.71

[a] – Significance at $P \leq 0.05$.
[a] – Istotność przy $P \leq 0,05$.

Exemplary microscopic images of intramuscular fat in the masseter muscle of the cattle groups under study are shown in photographs (Fig. 1–3; 10×10 magnification).

CONCLUSIONS

The results obtained in our study fall within the intramuscular fat levels reported for European breeds by other authors. Both the percentage and distribution of intramuscular fat are determined by genotype, feeding level, age of the animals, and type of muscle. In our study, the masseter muscle in the analysed population of slaughter cattle had a relatively low proportion of intramuscular fat (1.84% on

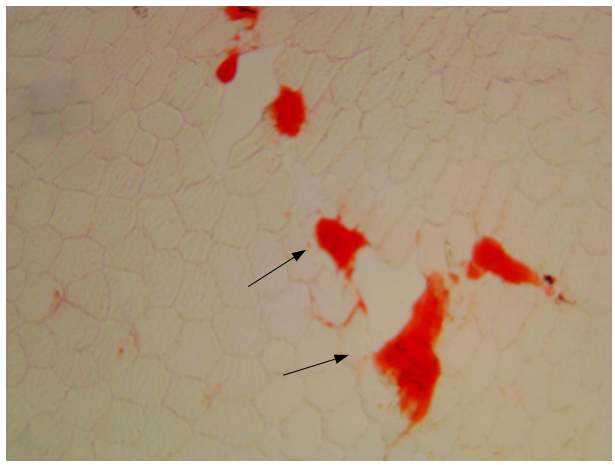

Fig. 1. Cross section of masseter muscle of bull. Intramuscular fat (arrows)

Fot. 1. Przekrój poprzeczny mięśnia żwacza buhajka. Tłuszcz śródmięśniowy (strzałki)

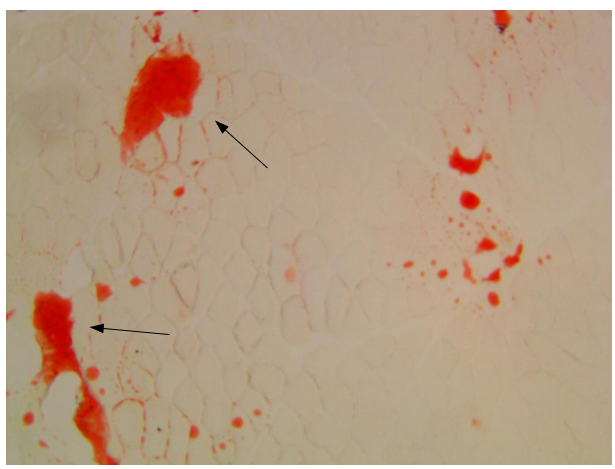

Fig. 2. Cross section of masseter muscle of heifer. Intramuscular fat (arrows)

Fot. 2. Przekrój poprzeczny mięśnia żwacza jałówki. Tłuszcz śródmięśniowy (strzałki)

average). The highest IMF level was found in heifers (2.26%) and the lowest in bulls (1.48%) ($P \leq 0.05$). There was no significant effect of the age of the studied animals on the level of intramuscular adipose tissue in the masseter. There are no reports in the literature on the fat content of this muscle in cattle. The masseter is continuously active and has mainly oxidative metabolism [Picard et al. 1996,

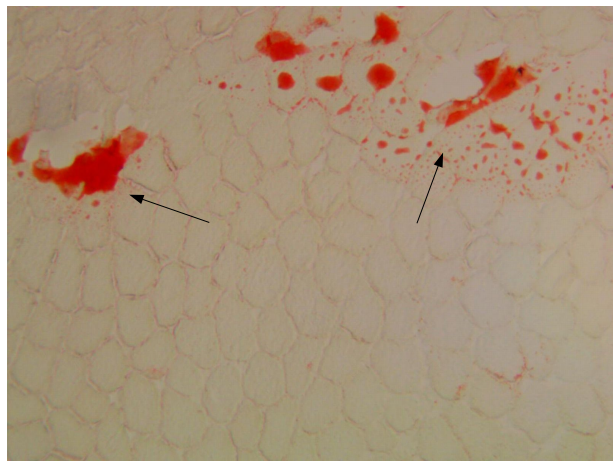

Fig. 3. Cross section of masseter muscle of cow. Intramuscular fat (arrows)

Fot. 3. Przekrój poprzeczny mięśnia żwacza krowy. Tłuszcz śródmięśniowy (strzałki)

Suzuki 1977] and it is likely that the low level of intramuscular adipose tissue is due to the specific characteristics of this muscle.

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ZAWARTOŚĆ TŁUSZCZU ŚRÓDMIĘŚNIOWEGO W MIĘŚNIU ŻWACZU (*MUSCULUS MASSETER*) BYDŁA W ZALEŻNOŚCI OD PŁCI I WIEKU

Streszczenie. Celem pracy było oznaczenie ilości tłuszczu śródmięśniowego w mięśniu żwaczu (*musculus masseter*) bydła rzeźnego w zależności od płci i wieku. Materiał doświadczalny stanowiło 40 osobników bydła (7 krów powyżej 5 roku życia, 12 jałówek i 21 buhajów w wieku około 24 miesięcy). Po uboju pobierano próbki mięśnia i zamrażano w ciekłym azocie. Zamrożone próbki ścinano w kriostacie na 10 µm skrawki. Następnie skrawki umieszczano na szkiełku podstawowym i poddano barwieniu Red-Oil w celu określenia zawartości tłuszczu śródmięśniowego. Z przeprowadzonych badań wynika, że mięsień żwacz (*musculus masseter*) badanej populacji bydła rzeźnego charakteryzował się stosunkowo niskim udziałem tłuszczu śródmięśniowego (średnio 1,84%). Najwyższy jego poziom stwierdzono u jałówek – 2,26%, natomiast najniższy u buhajków – 1,48% ($P \leq 0{,}05$). Nie stwierdzono istotnego wpływu wieku badanych zwierząt na poziom śródmięśniowej tkanki tłuszczowej w mięśniu żwaczu.

Słowa kluczowe: bydło rzeźne, mięsień żwacz, tłuszcz śródmięśniowy

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