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ORIGINAL PAPER

HEALTH-PROMOTING PROPERTIES OF MEAT FROM ONCE-CALVED AND MAIDEN HEIFERS*

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Abstract

Beef has a high nutritional value owing to the presence of minerals, vitamins, polyunsaturated fatty acids (PUFA) and selected protein fractions. The objective of this study was to investigate the influence of calving and nursing on the fatty acid profile and the content of minerals and vitamins in the meat of crossbred (Limousine x Polish Holstein-Friesian) once-calved heifers in comparison with the meat of maiden heifers. At 15 months of age, once-calved heifers were inseminated with the semen of Limousine bulls. After calving and lactation, they were slaughtered. Meat samples were collected from m. *longissimus dorsi* (MLD). The fatty acid composition was determined with the use of a gas chromatograph. The content of 5 minerals (potassium, sodium, magnesium, zinc, iron) was determined by atomic absorption spectrometry. The content of vitamins A and E was determined by high-performance liquid chromatography. Gestation and nursing had a negligent effect on the content of functional components in the meat of once-calved heifers, in comparison with the meat of heifers. The meat of heifers contained more vaccenic acid (TVA) (C 18:1 T10+11), arachidonic acid (AA) (C 20:4) and docosahexaenoic acid (DHA) (C 22:6), whereas the meat of once-calved heifers was more abundant in oleic acid, zinc, iron and vitamin E ($P \leq 0.01$). Commercially crossbred heifers can be included in once-bred heifer production systems, which create a possibility of producing additional offspring plus higher body weight of heifers with no adverse effect on the functional properties.

Keywords: once-calved and maiden heifers, beef, fatty acids, minerals, vitamins.

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INTRODUCTION

The sensory properties of beef are significantly influenced by fat deposition (Modzelewska-Kapituła, Nogalski 2014). The quality of fat is determined mainly by the content of fatty acids, which deliver health benefits for consumers. Polyunsaturated fatty acids (PUFA), mainly linoleic acid (LA), arachidonic acid (AA), eicosapentaenoic acid (EPA), docosapentaenoic acid (DPA) and docosahexaenoic acid (DHA), are biologically active substances that stimulate immunity, limit fat deposition, exert antibacterial, anticarcinogenic and antioxidant effects. Many lifestyle diseases are linked to an excessively wide ratio of *n*-6 to *n*-3 PUFA in a human diet. An optimal balance between intake of *n*-6 and *n*-3 fatty acids should be 5-8:1-2 (WHO 2003), whereas contemporary diets are characterized by a ratio of 10:20 (Breslow 2006). Beef should not be evaluated based only on its fat content, because it is also an excellent source of iron, zinc and magnesium (Li et al. 2005). In the group of most highly consumed meats in the United States, beef is characterized by the highest content of available iron and zinc (USDA-ARS 2010). Calcium, magnesium and phosphorus are important to a wide variety of body functions. First of all, they serve for bone mineralization. In addition, they are cofactors of many enzyme systems, sustaining muscle and nerve excitation. Iron is required for maintaining the oxygen carrying capacity of blood (Clarkson, Haymes 1995). Magnesium has a strong relationship with the immune system, in both nonspecific and specific immune responses, which are also known as innate and acquired immune responses (Tam et al. 2003). Vitamins A and E are important natural dietary antioxidants. Vitamin E is an essential nutrient which stabilizes PUFA and is a key determinant of meat quality, in particular in meat from ruminants (Wood et al. 2008).

In Poland, beef production is based mainly on young bulls and culled dairy cows. Beef quality can be improved by commercial crossbreeding of selected cows and heifers of the dairy cattle population to produce beef calves (Nogalski et al. 2013*a*). Heifers account for around 15% of slaughtered cattle, and around 200,000 heifers are slaughtered each year (Integrated System of Agricultural Market Information 2015). The population of calves suitable for fattening has been dwindling, and this problem could be addressed by using once-calved heifers for calf production. This production regime is known as the once-bred heifer (OBH) system. In this system, dairy heifers that are not suitable for further breeding and heifers produced by commercial crossbreeding in dairy herds are inseminated with the semen of beef bulls upon reaching reproductive maturity (Nogalski et al. 2016). After calving, primiparas referred to as once-calved heifers nurse their offspring for several months and are slaughtered after weaning. Earlier studies have demonstrated that gestation increases the body weight gains of heifers (Zembayashi 2001). In comparison with adult suckler cows, young growing heifers use more dietary energy to increase their body weight, therefore the OBH system is more effective than the production of calves in beef cattle herds (Bourdon, Brinks 1987). Meat production based on once-calved heifers, produced by crossbreeding with beef bulls in dairy cattle herds, is characterized by higher economic efficiency, and the production of crossbreeds as byproducts of dairy farms is cheaper than the production of beef cattle breeds in conventional cow-calf systems (Zembayashi 2001). Once-calved heifers should be used in the production of beef calves only if the resulting carcasses are characterized by high meat quality and if they increase the supply of beef on the consumer market (Hoving-Bolink et al. 1999). According to Daszkiewicz et al. (2006), the meat of once-calved heifers is characterized by lower quality than beef from heifers that are not used for reproductive purposes.

The slaughter value, carcass traits and meat quality of crossbred Polish Holstein-Friesian x Limousine bulls, steers and heifers have been analyzed in several studies (Nogalski et al. 2013*a*,*b*, Modzelewska-Kapituła, Nogalski 2014, Sobczuk-Szul et al. 2014), whereas the slaughter value of once-calved heifers, in particular the content of functional components in beef, remains insufficiently researched. The aim of this experiment was to investigate the influence of calving and nursing on the fatty acid profile and the content of minerals and vitamins in the meat of crossbred (Limousine x Polish Holstein-Friesian) once-calved heifers in comparison with the meat of maiden heifers.

MATERIAL AND METHODS

Animals, experimental design, and treatments

The experiment was performed on 33 females, including 16 heifers and 17 once-calved heifers produced by crossing Limousine (LM) bulls with Polish Holstein-Friesian (PHF) cows. The semen of 9 bulls was used for insemination. The feeding regime for calves and heifers was described in a previous study (Nogalski et al. 2016). Sixteen heifers were fattened until 18 months of age, after which they were transported to a meat processing plant and slaughtered. Seventeen once-calved heifers at 466.3 days of age and average body weight of 432.8 kg were inseminated with the semen of Limousine bulls. Heifers calved at 24.5 months of age. During gestation, the average daily gain of heifers was 0.73 kg, and their average body weight before calving was 636 kg. Parturition was unassisted in 10 heifers, and assistance was required in 7 animals. One heifer died after difficult parturition. One calf died shortly after birth. The average birth weight of calves (25% PHF and 75% LM) was 39.2 kg. Calves remained with their mothers for 102 days on average, and they achieved average body weight of 147.8 kg over the nursing period. Weaned calves were moved to fattening pens. After weaning, once-calved heifers were transported to a slaughterhouse. There they were kept individually for 15 to 20 h with access to fresh water. The animals were weighed before slaughter. All slaughter and post-slaughter procedures were executed according to meat industry rules (Council Regulation (EC) No 1099/2009). The animals were stunned, dressed and halved along the spine into two half-carcasses, which were chilled for 96 h at 4°C. Electric stimulation was not applied to the carcasses. The protocol for animal research was approved by the Ethics Committee of the University of Warmia and Mazury. The *longissimus dorsi* muscle (MLD) was removed from the right half-carcasses 96 h post mortem, and weighed. Meat specimens were collected from m. *longissimus dorsi* between the 11^{th}-13^{th} thoracic vertebrae. The samples were transported to the laboratory under refrigeration.

Fatty acid profile

According to the Soxhlet method, intramuscular fat (IMF) was extracted from ground beef speciments using a Büchi B-811 instrument with n-hexane as solvent. Fatty acid methyl esters were obtained by dissolving the extracted fat in a methanol-chloroform-H_2SO_4 mixture, followed by methylation according to the modified Peisker method (Żegarska et al. 1991). The profile of 33 fatty acids was assayed with the use of a gas chromatograph (Varian CP 3800). The operating conditions of the gas chromatograph are shown in Table 1. Data were calculated in Galaxie software. The following groups of fatty acids and ratios were calculated: SFA – saturated fatty acids, UFA – unsaturated fatty acids, MUFA – monounsaturated fatty acids, PUFA – polyunsaturated fatty acids, *n*-3 and *n*-6 fatty acids MUFA/SFA, PUFA/SFA, and *n*-6/*n*-3 PUFA.

Table 1

Working parameters of the gas chromatograph

Injector	split/splitless
Detector	flame-ionization
Sample volume	1 μL
Stationary phase	CP-sil 88
Length of capillary column	100 m
Inner diameter of capillary column	0.25 mm
Carrier gas	helium
Injector temperature	260°C
Detector temperature	260°C
Initial oven temperature/ raised to	110°C/ 249°C
Time of a single analysis	68 min

Determination of mineral content

Samples of MLD were homogenized, specimens of approximately 0.5 g were placed in Teflon-lined pressure vessels and combined with 7 ml of 65%

spectrally pure nitric acid (Merck). Each sample was analyzed in duplicate. The vessels were sealed, and the samples were mineralized in the MARS Xpress 5 microwave digestion system (Candela, USA). Every mineralization procedure involved 2 blank samples and 2 samples of certified reference material. Mineralized samples were cooled and transferred to 25 ml volumetric flasks. They were analyzed with an atomic absorption spectrometer (Candela, USA) equipped with lamps for different elements (potassium, sodium, magnesium, zinc, iron).

Determination of vitamin A and E content

The content of vitamins A and E was determined based on the applicable standards (PN-EN 12822; PN-EN 12823-1), which were slightly modified for the needs of this study. Samples of MLD obtained from the loin were ground, and 2 g specimens were placed in 25 ml amber glass flasks. The samples were saponified at a temperature of 80°C for 30 minutes in 1.2 ml of 20% ascorbic acid, 0.6 ml of methanol and 5 ml of 60% potassium hydroxide. After cooling, fat was extracted twice with n-hexane, and the extract was evaporated to dryness in a stream of nitrogen. The fat remaining in flasks was dissolved in 1 ml of anhydrous ethanol and filtered into amber glass vials. Chemical compounds were separated by high-performance liquid chromatography (HPLC) using a silica column. The separated compounds were identified by two detectors in tandem (UV-visible photodiode array detector and fluorescence detector). Alpha-tocopherol was detected by fluorescence spectroscopy, and retinol was detected with the use of the UV-visible photodiode array detector. Injection volume was 20 μl. The content of total α-tocopherol and retinol in meat was calculated in duplicate for each muscle, based on the external standard, from a standard curve of peak area vs. concentration.

Statistical analysis

The experimental data from two treatments involving heifers (n = 16) and once-calved heifers (n = 15) were processed statistically by one-way ANOVA and the Tukey's HSD test in a Statistica 10 program (StatSoft Inc., Tulsa, USA) at a significance level of 1 and 5%.

RESULTS AND DISCUSSION

Heifers were slaughtered at an average age of 558.9 days and average body weight of 482.4 kg (Table 2). Once-calved heifers were 293.4 days older at slaughter, and their average body weight was 569.7 kg. In a study by Litwińczuk et al. (1991), the average body weight of PHF x LM once-calved heifers slaughtered at 30-32 months of age was 460 kg. In our study, once-calved heifers were 110 kg heavier at slaughter, which resulted from diffe-

Table 2

Age and body weight at slaughter and the characteristics of the m. *longissimus dorsi* (MLD) of heifers and once-calved heifers

Traits	Heifers (n = 16)		Once-calved heifers (n = 15)		Significance
	LSM	SE	LSM	SE	
Age at slaughter, days	558.9	2.16	852.3	4.36	**
Body weight at slaughter (kg)	482.4	12.88	569.7	16.59	**
Weight of MLD (kg)	10.33	0.34	11.01	0.37	*
MLD area (cm^2)	82.8	3.24	89.6	2.65	*
MLD area, cm^2 100 kg^{-1} of slaughter weight	17.2	0.21	15.7	0.24	-
Intramuscular fat content of MLD (%)	2.97	0.31	3.99	1.46	*

MLD – m. *longissimus dorsi* (MLD), LSM – least-squares means, SE – standard error;
Significant differences between means: * $P \leq 0.05$ and ** $P \leq 0.01$.

rent feeding regimes. In the cited experiment (Litwińczuk et al. 1991), once-calved heifers were grazed in summer, whereas in our study, the animals were kept indoors throughout the year and fed grass silage supplemented with concentrate. The *longissimus dorsi* muscle is one of the most valuable cuts of beef. The weight and cross-sectional area of MLD were higher in once-calved heifers, which could be attributed to their higher body weights at slaughter. According to Bureš , Bartoň (2012), the carcasses of older animals are characterized by lower content of high-priced cuts, smaller MLD area per 100 kg of slaughter weight and higher fatness. In a study by Steen, Kilpatrick (1995), higher slaughter weight was also associated with higher carcass fatness and a reduction in meat and bone proportions. In our study, MLD area per 100 kg of slaughter weight was lower and the intramuscular fat (IMF) content of MLD was significantly higher in once-calved heifers than in heifers. Harper, Pethick (2004) were previously identified gender as an important source of variation in the IMF content of beef, and the influence of sex hormones on the growth and development of intramuscular adipocytes. Heifers mature faster and are therefore much more predisposed to IMF deposition. Intramuscular fat from once-calved heifers contained less ($P \leq 0.01$) SFA and more ($P \leq 0.01$) UFA, in particular MUFA (Table 3). A high proportion of SFA and a low proportion of PUFA in meat fat results from the hydrogenation of dietary fat by ruminal microflora (De Smet et al. 2000). For optimal results, beef producers should decrease the concentrations of SFA in fat and/or increase the content of PUFA, in particular n-3 fatty acids (Kołczak 2008). In the present study, gestation and lactation increased the share of MUFA in the total fatty acid pool in once-calved heifers, as compared with heifers. In the work of Zembayashi (2001), pregnancy and 105 days of lactation increased the content of MUFA, whereas gestation alone

Table 3

Fatty acid groups and ratios in intramuscular fat of heifers and once-calved heifers

Item	Heifers (n = 16)		Once-calved heifers (n = 15)		Significance
	LSM	SE	LSM	SE	
SFA (%)	49.35	0.532	46.71	0.799	**
UFA (%)	50.68	0.534	53.29	0.798	**
MUFA (%)	46.60	0.672	49.77	0.826	**
PUFA (%)	4.08	0.299	3.52	0.164	-
MUFA/SFA	0.948	0.022	1.072	0.036	**
PUFA/SFA	0.083	0.025	0.076	0.012	-
n-3 fatty acids (%)	1.19	0.125	0.92	0.048	*
n-6 fatty acids (%)	2.49	0.207	2.18	0.125	-
n-6/n-3 PUFA ratio	2.29	0.149	2.37	0.063	-

SFA – saturated fatty acids, UFA – unsaturated fatty acids, MUFA – monounsaturated fatty acids, PUFA – polyunsaturated fatty acids, LSM – least-squares means, SE – standard error; Significant differences between means: * $P \leq 0.05$ and ** $P \leq 0.01$.

significantly decreased the share of MUFA in once-calved heifers in comparison with heifers. Lengyel et al. (2003) noted that the PUFA content of intramuscular fat in MLD decreased with age, reaching 25.5% at 7 months, 18.4% at 14 months and 13.6% at 19 months. In our study, the share of PUFA in the total fatty acid pool was 0.56% lower in once-calved heifers than in heifers. Sobczuk-Szul et al. (2014) observed a similar PUFA/SFA ratio (0.063), Węglarz (2010) reported a higher PUFA/SFA ratio (0.19) and Aldai et al. (2007) noted a much higher PUFA/SFA ratio (0.45) than in our study. In our research, we noted that the content of omega-3 fatty acids was statistically higher ($P \leq 0.05$) and the n-6/n-3 ratio was lower in IMF of heifers than in the group of one-calved heifers (Table 3). According to the FAO and WHO recommendations, the n-6/n-3 ratio in a human diet should be around 5.0 (Kołczak 2008). In the present research, the beef was characterized by an even lower n-6/n-3 ratio, which may have a positive impact on human health. As regards functional fatty acids, the IMF of once-calved heifers contained 2.93% more ($P \leq 0.01$) oleic acid in comparison with heifers (Table 4). Statistically significant differences were noted in the concentrations of vaccenic acid (TVA), AA and DHA which were higher in the IMF of heifers.

Beef is the most zinc-abundant type of meat, and its zinc content is 2-fold higher in comparison with pork and veal and 5- to 7-fold higher in comparison with poultry meat (Barowicz, Brejta 2000). In our experiment, significant differences in the zinc and iron content of meat were observed between heifers and once-calved heifers (Table 5). In the meat of once-calved heifers, the iron content was 0.56 mg higher and zinc content was 0.63 mg higher per 100 g of fresh meat in comparison with the meat of heifers. Giuf-

Table 4

Fatty acids with functional properties in the intramuscular fat of heifers and once-calved heifers

Fatty acids (%)	Heifers (n = 16)		Once-calved heifers (n = 15)		Significance
	LSM	SE	LSM	SE	
C 18:1 T10+11 vaccenic acid (TVA)	1.082	0.121	0.668	0.082	*
C 18:1 n-9 oleic acid (OA)	38.68	0.700	41.61	0.685	**
C 18:2 linoleic acid (LA)	2.090	0.158	1.930	0.098	-
C 18:2 C9 T11 conjugated linoleic acid (CLA)	0.240	0.023	0.251	0.010	-
C 18:3 linolenic acid (LNA)	0.568	0.042	0.525	0.020	-
C 20:4 arachidonic acid (AA)	0.407	0.053	0.247	0.044	*
C 20:5 eicosapentaenoic acid (EPA)	0.077	0.013	0.064	0.007	-
C 22:5 docosapentaenoic acid (DPA)	0.274	0.048	0.181	0.022	-
C 22:6 docosahexaenoic acid (DHA)	0.116	0.065	0.023	0.005	*

LSM – least-squares means, SE – standard error;
Significant differences between means: * $P \leq 0.05$ and ** $P \leq 0.01$.

Table 5

Content of mineral elements in the meat (mg kg^{-1} of fresh meat)
of heifers and once-calved heifers

Minerals	Heifers (n = 16)		Once-calved heifers (n = 15)		Significance
	LSM	SE	LSM	SE	
Potassium (K)	4832.5	84.30	4765.8	95.00	-
Sodium (Na)	524.0	11.29	538.1	12.42	-
Magnesium (Mg)	196.6	5.83	186.4	4.40	-
Zinc (Zn)	34.5	1.62	40.8	2.18	*
Iron (Fe)	13.2	0.77	18.8	1.05	**

LSM – least-squares means, SE – standard error;
Significant differences between means: * $P \leq 0.05$ and ** $P \leq 0.01$.

FRIDA-MENDOZA et al. (2007) investigated variations in the mineral content of the *longissimus dorsi thoracis* muscle from water buffalo and Zebu-influenced cattle. They found that both zinc and magnesium concentrations increased with the animals' age (evaluated at 17, 19, and 24 months of age). In a study by PURCHAS, BOSBOOM (2005), iron concentrations in the meat of crossbred beef heifers were also influenced by the age at slaughter, but not by a diet or feeding intensity. In the cited study, the meat of heifers slaughtered at 27-28 months of age contained 18.6% more iron than the meat of heifers slaughtered at 16-17 months of age. In our study, where the age difference between the compared groups was similar, the meat of older heifers contained 42% more iron. In addition to age, gestation was probably another factor that contributed to the higher iron content of beef from older heifers.

Vitamins A and E are essential nutrients in a human diet (Wood et al. 2008). The meat of once-calved heifers was more abundant in vitamins A and E than the meat of heifers (Figure 1). The observed differences in the vitamin E content of beef (1806 and 2412 μg kg^{-1}) were statistically significant, and they can be attributed mainly to the higher IMF content of meat from once-calved heifers.

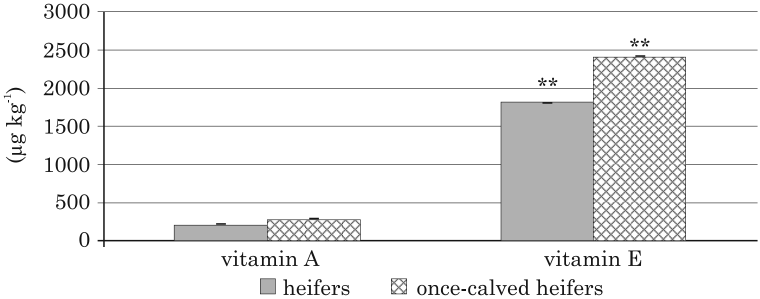

Fig. 1. Content of vitamins A and E in the meat (μg kg^{-1} of fresh meat) of heifers and once-calved heifers.
Significant differences between means: ** $P \leq 0.01$

CONCLUSIONS

The meat of heifers was more abundant in TVA, AA and DHA, whereas the meat of once-calved heifers was characterized by higher concentrations of oleic acid, zinc, iron and vitamin E. These commercially crossbred heifers can be included in once-bred heifer beef production systems, which create a possibility of producing additional offspring plus higher body weight of heifers with no adverse effect on the content of functional components in meat.

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