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Posted Date: 30 January 2026

doi: 10.20944/preprints202601.2349.v1

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Article

# Optimising Beef Fatty Acid Composition and Lipid Quality through Silage Type and Feeding Intensity during the Finishing Period

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## Simple Summary

Beef fat quality is important for both consumers and the meat industry, as it affects nutritional value and processing properties. The composition of fatty acids in beef can be modified through feeding strategies applied during the finishing period of cattle. In this study, Holstein–Friesian bulls were fed diets differing in silage type (grass or maize silage) and feeding intensity. We examined how these factors influenced the amount of intramuscular fat and the composition of fatty acids in beef. Increasing feeding intensity mainly increased the amount of fat deposited in muscle, while the type of silage primarily affected the fatty acid composition of the fat. Diets based on grass silage resulted in higher proportions of beneficial n–3 fatty acids and a more favourable balance between n–6 and n–3 fatty acids compared with maize silage–based diets. Overall, the results show that combining appropriate silage type with feeding intensity allows producers to influence the nutritional quality of beef fat without markedly changing its technological quality.

## Abstract

The quality of beef fat depends not only on its amount but also on fatty acid composition and lipid quality indices, which are strongly influenced by feeding strategies applied during the finishing period. The aim of this study was to evaluate the effects of silage type and feeding intensity on intramuscular fat deposition, fatty acid composition, desaturase activity indices and lipid quality indices in finishing Holstein–Friesian bulls. Thirty-two bulls were assigned to a 2 × 2 factorial experiment and fed total mixed rations for 120 days. Intramuscular fat content and fatty acid composition of the longissimus lumborum muscle were determined by gas chromatography, and lipid quality indices were calculated from the fatty acid profile. Increasing feeding intensity significantly increased intramuscular fat content and the absolute amounts of most fatty acid groups, whereas silage type mainly affected fatty acid composition by increasing the proportion of n–3 fatty acids and reducing the n–6/n–3 ratio in grass silage–based diets. Significant interactions between silage type and feeding intensity were observed for selected fatty acids, indicating that the response to increased dietary energy supply depended on the forage base of the diet. Despite marked changes in fatty acid composition, lipid quality indices (including AI and TI) were only moderately affected. Overall, these results indicate that feeding intensity primarily controls the extent of lipid deposition in beef, while silage type modulates fatty acid composition, emphasising the importance of combined dietary strategies for improving the nutritional quality of beef fat.

**Keywords:** Holstein–Friesian bulls; finishing period; intramuscular lipid deposition; n–3 fatty acids; n–6/n–3 ratio; desaturase activity; lipid quality indices

## 1. Introduction

In recent years, increasing attention has been paid to the quality of beef fat, not only in terms of sensory attributes but also with regard to its nutritional value and potential implications for human health. The fatty acid composition of beef, including the proportions of saturated (SFA), monounsaturated (MUFA), and polyunsaturated fatty acids (PUFA), as well as the relationships among these fractions, is considered one of the key indicators of final product quality [1,2]. Particular importance is attributed to the content of oleic acid (C18:1 cis-9), which contributes to improved tenderness and juiciness of meat and is also associated with a more favourable lipid profile from a nutritional perspective [3,4].

One of the most important factors shaping the fatty acid profile of beef is the feeding strategy, including the type of forage used and the intensity of fattening. It has been demonstrated that diets based on grass silage promote a higher proportion of n-3 fatty acids and a lower n-6/n-3 ratio, whereas maize silage-based diets and a higher proportion of concentrates are more frequently associated with an increased proportion of MUFA, including C18:1 cis-9 [1,5–7]. These differences result not only from variations in the lipid composition of feeds but also from interactions between diet, ruminal lipid transformations, and tissue metabolism, which together determine the rate of lipogenesis and the composition of triacylglycerol and phospholipid fractions in muscle.

Feeding intensity and finishing strategies play a crucial role in regulating lipogenic processes and shaping the culinary properties of beef. Studies conducted in Holstein–Friesian bulls have shown that less intensive fattening systems favour a higher proportion of PUFA and a reduced n-6/n-3 ratio in meat, despite a concomitant reduction in growth rate [8]. Increasing dietary energy density has been reported to enhance intramuscular fat (IMF) deposition but simultaneously shift fatty acid proportions towards a higher MUFA content at the expense of the relative contribution of PUFA, a phenomenon often described as the “dilution effect” of phospholipids due to the expansion of the triacylglycerol fraction [9]. Consequently, improvements in marbling do not necessarily translate into parallel improvements in the nutritional quality of fat, and the evaluation of feeding effects should encompass both IMF content and the detailed fatty acid profile, as well as lipid quality indices. Moreover, studies comparing different rations during the finishing period have demonstrated that modifications in the proportion of forage components (e.g., lucerne, maize stover, maize silage) may influence selected sensory attributes, such as muscle fibre tenderness and juiciness, as well as the proportions of specific fatty acid fractions [10]. This highlights the need to simultaneously consider sensory quality and lipid quality within the context of feeding systems applied during the finishing period.

The lipid and technological quality of beef is also influenced by factors related to production organisation, feeding level, and slaughter maturity of animals [11,12]. Feeding intensity and diet formulation during the finishing phase determine the rate of fat deposition and the proportions of lipid fractions within muscle tissue, which may translate into changes in technological meat traits, including tenderness, colour, and water-holding capacity [11,13]. It has been shown that increased slaughter weight and higher dietary energy concentration promote intramuscular fat accumulation but may also lead to alterations in the balance between saturated and polyunsaturated fatty acids and changes in the n-6/n-3 ratio [12,13]. From a practical production perspective, this necessitates the development of feeding strategies during the finishing period that balance production efficiency with quality objectives, encompassing both technological properties and the lipid profile and nutritional value of beef [2,11].

Interpretation of changes in the lipid profile requires consideration not only of IMF content but also of fat deposition site and animal-related factors. Previous studies have demonstrated that fatty acid composition differs among fat depots (intramuscular, intermuscular, subcutaneous, and perirenal) and between sexes (bulls vs. steers), and in some experimental systems, feeding intensity was not the dominant factor compared with fat depot location [14]. This further underscores the importance of using indices that describe metabolic processes (e.g., desaturation indices), rather than relying solely on simple proportions of SFA, MUFA, and PUFA.

Increasing attention has also been devoted to desaturase indices, which serve as indirect indicators of the activity of  $\Delta 9$ -desaturase (stearoyl-CoA desaturase). This enzyme catalyses the conversion of saturated fatty acids, such as C16:0 and C18:0, into their monounsaturated counterparts (C16:1 and C18:1) and is closely associated with the intensity of lipogenic processes and energy metabolism [15,16]. Complementary to this approach are lipid quality indices (e.g., AI, TI, and h/H), which integrate the relative importance of individual fatty acids in terms of atherogenic and thrombogenic risk, as well as potential nutritional benefits [2].

In a previous study, Nogalski et al. [17] demonstrated a significant effect of silage type and feeding intensity on carcass traits and intramuscular fat content in fattening cattle. However, that analysis focused primarily on quantitative aspects of fat deposition, without a comprehensive characterisation of lipid quality. Since changes in IMF content do not necessarily reflect changes in fatty acid composition, it is justified to extend the scope of research to include a detailed analysis of individual fatty acids, desaturase indices, and comprehensive lipid quality indices. Existing studies clearly indicate that both silage type and feeding intensity during the finishing period significantly influence the lipid composition of beef [2,5,11,18]. Nevertheless, many of these studies address selected aspects of fat quality without simultaneously considering desaturase indices and a full set of lipid quality indices, thereby limiting insight into metabolic mechanisms and the practical “nutritional cost–benefit” of dietary modifications.

Continuation of the research initiated by Nogalski et al. [17] allows this knowledge gap to be addressed by providing data that improve understanding of how manipulation of silage type and feeding intensity modulates (i) the proportions of key fatty acids, (ii) indirect measures of lipogenic enzyme activity, and (iii) integrated lipid quality indices relevant to consumers. The results obtained may contribute to the current body of knowledge and provide practical guidance for optimising feeding strategies for finishing cattle in order to improve the lipid quality of beef, in line with consumer expectations and contemporary trends in animal-derived food production.

The aim of this study was to evaluate the effect of silage type and feeding intensity on the detailed fatty acid profile, desaturase activity indices, and lipid quality indices of beef, as a continuation of earlier research focusing on carcass traits and intramuscular fat content.

## 2. Materials and Methods

### 2.1. Animals, Housing and Experimental Design

The experiment was conducted in accordance with the feeding and management scheme described by Nogalski et al. [17]. Thirty-two Holstein–Friesian bulls originating from a dairy herd were used. During the rearing period, calves were managed conventionally. From approximately five months of age, animals were fattened semi-intensively using total mixed rations (TMR). At the beginning of the finishing period (approximately 600 days of age; mean body weight about 530 kg), bulls were allocated to experimental treatments using the analogue method to equalise age and body weight among groups. Animals were housed in group pens on deep bedding, with free access to water and mineral salt blocks. Each bull was considered an experimental unit. The finishing period lasted 120 days. Throughout the experiment, animals remained under veterinary supervision, and no health disorders affecting carcass or meat quality were observed.

### 2.2. Feeding Management and Experimental Diets

Feeding management followed the protocol described by Nogalski et al. [17]. Bulls were fed ad libitum total mixed rations (TMR). The experiment was conducted using a  $2 \times 2$  factorial design with silage type (TS; grass silage, GS, or maize silage, MS) and feeding intensity (FI; intensive, I, or semi-intensive, SI) as fixed factors. Four dietary treatments were applied: GS-I, GS-SI, MS-I and MS-SI. The proportion of forage and concentrate in the ration differed according to feeding intensity, whereas silage type determined the forage base of the diet. In intensive diets, the forage-to-concentrate ratio was approximately 50:50 (DM basis), while in semi-intensive diets it was approximately 70:30, as

described by Nogalski et al. [17]. All diets were formulated to meet nutrient requirements of finishing bulls according to INRA [19] recommendations. Chemical composition and fatty acid profiles of silages and concentrate components are shown in Table 1, whereas ingredient composition, chemical composition and nutritive value of experimental diets are presented in Table 2.

**Table 1.** Chemical and fatty acid composition of experimental foders (mean  $\pm$  standard error).

Specification	Grass silage	Maize silage	Triticale	Rapeseed meal
Chemical composition (g·kg <sup>-1</sup> DM) of experimental foders				
Dry matter g·kg <sup>-1</sup>	285	322	875	878
Organic matter	906	967	966	921
Crude protein	121	88.9	122	383
NDF	536	337	162	298
ADF	314	196	41	212
ADL	25.5	12.7	-	-
NFC	194	508	629	237
pH	4.23	3.56		
Lactic acid	43.6	27.5		
Acetic acid	12.5	6.6		
Butyric acid	0.09	0.08		
N-NH <sub>3</sub> (g kg <sup>-1</sup> TN)	75.9	33.9		
UFV	0.86	0.85	1.19	1.03
PDIN	83	50	85	254
PDIE	72	68	98	162
Fatty acid profile (g/100 g fatty acids)				
C14:0	0.16 $\pm$ 0.01	1.77 $\pm$ 0.07		
C16:0	13.99 $\pm$ 0.19	22.65 $\pm$ 0.24		
C18:0	1.98 $\pm$ 0.08	3.09 $\pm$ 0.11		
C18:1 n-9 (OA)	22.65 $\pm$ 0.09	6.11 $\pm$ 0.09		
C18:2 n-6 (LA)	50.78 $\pm$ 0.14	24.32 $\pm$ 0.12		
C18:3 n-3 (LNA)	8.21 $\pm$ 0.09	39.25 $\pm$ 0.15		

NDF—neutral detergent fiber; ADF—acid detergent fiber; ADL—acid detergent lignin; NFC—non-fiber carbohydrates; N-NH<sub>3</sub>—ammonia nitrogen; TN—total nitrogen; UFV—meat production units; PDIN—protein digested in the small intestine depending on rumen-degraded protein; PDIE—protein digested in the small intestine depending on rumen-fermented organic matter; OA—oleic acid; LA—linoleic acid; LNA—linolenic acid.

**Table 2.** Ingredients (% DM) and chemical composition of diets.

Specification	GS-I	GS-SI	MS-I	MS-SI
Grass silage	50	70		
Maize silage			50	70
Triticale grain	47	27	41	18
Rapeseed meal	3	3	9	12
Dry matter (g/kg fresh)	580.09	462.09	598.77	488.26
In g/kg DM				

Organic matter	934.65	922.65	962.45	961.3
Crude protein	129.33	129.13	128.94	130.15
2NDF	353.08	427.88	261.74	300.82
3ADF	182.63	237.23	133.89	170.02
4NFC	399.74	312.74	533.22	497.26
UFV	1.02	0.95	1.01	0.93
PDIN	89.07	88.67	82.71	80.78
PDIE	86.92	82.71	87.76	84.68

GS-I—grass silage intensive; GS-SI—grass silage semi-intensive; MS-I—maize silage intensive; MS-SI—maize silage semi-intensive; NDF—neutral detergent fiber; ADF—acid detergent fiber; NFC—non-fiber carbohydrates; UFV—meat production units; PDIN—protein digested in the small intestine depending on rumen-degraded protein; PDIE—protein digested in the small intestine depending on rumen-fermented organic matter.

### 2.3. Feed Sampling and Chemical Analyses

Representative samples of silages and concentrates were collected at regular intervals throughout the experiment. Dry matter, crude protein, ether extract and ash were determined according to AOAC [20] procedures. Neutral detergent fibre (NDF), acid detergent fibre (ADF) and acid detergent lignin (ADL) were analysed using the detergent method described by Van Soest et al. [21]. Silage pH was measured potentiometrically. Concentrations of lactic, acetic and butyric acids were determined in aqueous extracts according to the principles described by McDonald et al. [22]. Fatty acid composition of feeds was analysed by gas chromatography after preparation of fatty acid methyl esters (FAME) using the modified Peisker method [23].

### 2.4. Slaughter Procedure and Meat Sampling

At the end of the finishing period, bulls were transported to a commercial abattoir and rested for 15–20 h with access to water. All slaughter and post-mortem procedures complied with Council Regulation (EC) No 1099/2009. Samples of the longissimus lumborum muscle were excised from chilled carcasses 96 h post mortem. Muscle samples were vacuum-packed, aged at  $4 \pm 1$  °C until day 14 post mortem, and then frozen at  $-20$  °C until analysis.

### 2.5. Intramuscular Fat Content and Fatty Acid Profile

Chemical composition of the longissimus lumborum muscle was determined after the ageing period. Intramuscular fat (IMF) content was determined by Soxhlet extraction using a Buechi B-811 extraction system with hexane as the solvent, in accordance with AOAC Method 991.36 9 [20], and the results are presented in Table 3. For fatty acid analysis, muscle samples were ground and homogenised using an Ultra-Turrax homogeniser (Janke & Kunkel), and total lipids were extracted from the homogenised samples using the same Soxhlet procedure. Fatty acid methyl esters (FAME) were prepared according to PN-EN ISO 5509 (2001) using the modified Peisker method [23], involving methylation in a methanol–chloroform–H<sub>2</sub>SO<sub>4</sub> mixture. Fatty acid separation and quantification were performed by gas chromatography with flame ionisation detection (GC-FID) using a Varian CP 3800 gas chromatograph equipped with a split/splitless injector and a CP-Sil 88 capillary column (100 m × 0.25 mm i.d.), in accordance with PN-EN ISO 5508 (1996). Helium was used as the carrier gas. Injector and detector temperatures were set at 260 °C. The oven temperature was programmed from 110 °C to 249 °C. Chromatographic data were processed using the Galaxie Chromatography Data System.

Individual fatty acids were identified by comparing their retention times with those of certified reference standards (Supelco Inc., Bellefonte, PA, USA), analysed under identical chromatographic conditions. The proportion of 31 fatty acids was determined and expressed as a percentage of the total identified fatty acids (Table 4). Fatty acids were grouped into saturated (SFA), monounsaturated

(MUFA) and polyunsaturated (PUFA) fractions. In addition, the ratios PUFA/SFA and n-6/n-3 were calculated to characterise the nutritional quality of intramuscular fat.

Nutritional quality of intramuscular fat was assessed using lipid quality indices calculated from the fatty acid profile (Table 5). The atherogenic index (AI) and thrombogenic index (TI) were calculated according to Ulbricht and Southgate [24]:

$$AI = \frac{C12:0 + 4 \times C14:0 + C16:0}{\Sigma MUFA + \Sigma PUFA}$$

$$TI = \frac{C14:0 + C16:0 + C18:0}{(0.5 \times \Sigma MUFA) + (0.5 \times \Sigma PUFA \text{ n-6}) + (3 \times \Sigma PUFA \text{ n-3}) + (\Sigma PUFA \text{ n-3} / \Sigma PUFA \text{ n-6})}$$

These indices were used to evaluate the potential impact of dietary treatments on the nutritional quality of beef fat.

## 2.6. Statistical Analysis

Data were analysed using a two-factorial least squares model including the fixed effects of silage type (TS), feeding intensity (FI) and their interaction (TS × FI):

$$Y_{ijk} = \mu + TS_i + FI_j + (TS \times FI)_{ij} + e_{ijk}$$

where Y is the analysed trait,  $\mu$  is the overall mean, TS is the effect of silage type, FI is the effect of feeding intensity, and e is the random error. Differences between means were considered statistically significant at  $P < 0.05$  and at  $p < 0.01$ . Statistical analyses were performed using Statistica software (TIBCO Software Inc., USA).

## 3. Results

### 3.1. Growth Performance and Carcass Fatness

Results concerning growth performance and carcass characteristics obtained in the first part of this experiment were reported previously by Nogalski et al. (2023). Briefly, finishing Holstein-Friesian bulls reached final body weights of approximately 620–690 kg, with higher values observed under intensive feeding. Average daily gains ranged from about 0.90 to 1.25 kg day<sup>-1</sup> and were significantly affected by feeding intensity. Carcass fatness differed among dietary treatments, with higher fatness scores observed in intensively fed bulls and in animals receiving maize silage-based diets. These results provide the growth and carcass background for the present evaluation of intramuscular fat content and fatty acid composition.

### 3.2. Intramuscular Fat Content and Individual Fatty Acid Composition

The intramuscular fat (IMF) content of the longissimus lumborum muscle was affected by feeding intensity, whereas silage type had no significant effect (Table 3). Higher IMF levels were observed in intensively fed bulls compared with those fed semi-intensively. No significant TS × FI interaction was detected for IMF content.

The proportions of several individual fatty acids were influenced by silage type, feeding intensity, or their interaction (Table 3). Feeding intensity significantly affected the proportions of C14:0, C15:0, C18:1 n-7, C18:2 n-6, and long-chain n-3 fatty acids, including DPA and DHA. Silage type significantly influenced the proportions of C18:0, C18:1 n-7, EPA, and DPA. Significant TS × FI interactions were observed for C15:0, C17:0, C18:3 n-3, C20:4 n-6, DPA, and DHA, indicating that the response of these fatty acids to feeding intensity depended on the type of silage used.

**Table 3.** Individual fatty acid composition (% of total identified fatty acids).

Fatty acid (% total FA)	Type of silage (TS)		Fattening intensity (FI)		SEM	P-value		
	Grass silage	Maiz silage	Intensive (I)	Semi-intensive (SI)		TS	FI	TSxFI
Intramuscular fat (%)	2.74	3.01	3.50	2.26	0.189	0.405	0.001	0.886
C14:0	2.65	2.56	2.77	2.45	0.076	0.731	0.025	0.272
C15:0	0.37	0.42	0.43	0.37	0.013	0.027	0.018	0.041
C16:0	25.68	25.51	26.00	25.19	0.267	0.754	0.146	0.777
C17:0	0.98	1.08	1.05	1.00	0.033	0.108	0.355	0.027
C18:0	13.86	16.00	15.62	14.22	0.485	0.016	0.111	0.275
C16:1 c9	4.02	3.53	3.73	3.83	0.190	0.191	0.784	0.068
C18:1 c9 (oleic)	38.36	37.95	37.80	38.51	0.602	0.732	0.559	0.102
C18:1 c11	1.82	1.61	1.58	1.85	0.057	0.048	0.013	0.525
C18:1 t10+t11 (vaccenic)	2.00	2.08	2.23	1.83	0.151	0.624	0.162	0.204
C18:2 n-6 (LA)	3.81	3.62	3.26	4.14	0.204	0.429	0.022	0.065
C18:3 n-3 (ALA)	0.55	0.47	0.49	0.53	0.022	0.063	0.283	0.018
C20:4 n-6 (ARA)	0.83	0.60	0.48	0.94	0.085	0.122	0.003	0.063
CLA c9,t11	0.41	0.39	0.41	0.39	0.010	0.470	0.145	0.227
EPA (C20:5 n-3)	0.10	0.07	0.07	0.10	0.008	0.022	0.126	0.236
DPA (C22:5 n-3)	0.23	0.16	0.15	0.24	0.016	0.027	0.003	0.026
DHA (C22:6 n-3)	0.03	0.03	0.02	0.03	0.001	0.141	0.040	0.091

LA—linoleic acid; ALA— $\alpha$ -linolenic acid; ARA—arachidonic acid; CLA—conjugated linoleic acid; EPA—eicosapentaenoic acid; DPA—docosapentaenoic acid; DHA—docosahexaenoic acid.

Fatty acid group composition was affected by both dietary factors (Table 4). Feeding intensity significantly influenced the proportions of  $\Sigma$ SFA,  $\Sigma$ PUFA,  $\Sigma$ n-6,  $\Sigma$ n-3, and the PUFA/SFA ratio. Silage type significantly affected  $\Sigma$ n-3 content and the n-6/n-3 ratio. A significant TS  $\times$  FI interaction was detected for  $\Sigma$ n-3 and PUFA/SFA, indicating differential responses of these parameters to feeding intensity depending on silage type.

**Table 4.** Fatty acid groups and ratios (% of total fatty acids).

Parameter (% total FA)	Type of silage (TS)		Fattening intensity (FI)		SEM	P-value		
	Grass silage	Maiz silage	Intensive (I)	Semi-intensive (SI)		TS	FI	TSxFI
$\Sigma$ SFA	44.15	46.31	46.60	43.88	0.629	0.076	0.026	0.430
$\Sigma$ MUFA	49.36	47.68	47.86	49.19	0.703	0.480	0.636	0.112

ΣPUFA	6.56	6.04	5.55	7.02	0.373	0.389	0.010	0.057
Σn-6	4.57	4.22	3.75	5.01	0.267	0.561	0.015	0.068
Σn-3	1.09	0.88	0.87	1.10	0.050	0.023	0.015	0.029
n-6/n-3	4.19	4.84	4.44	4.57	0.157	0.042	0.694	0.615
PUFA/SFA	0.15	0.13	0.12	0.16	0.007	0.109	0.001	0.056
MUFA/SFA	1.13	1.04	1.04	1.13	0.030	0.270	0.222	0.171
EPA+DHA	0.12	0.11	0.10	0.12	0.009	0.225	0.186	0.447

ΣSFA—sum of saturated fatty acids; ΣMUFA—sum of monounsaturated fatty acids; ΣPUFA—sum of polyunsaturated fatty acids; Σn-6—sum of n-6 polyunsaturated fatty acids; Σn-3—sum of n-3 polyunsaturated fatty acids.

### 3.3. Nutritional Quality and Desaturase Indices

Nutritional quality indices calculated from the fatty acid profile are presented in Table 5. Feeding intensity significantly affected the atherogenic index (AI) and the h/H ratio, whereas silage type influenced the thrombogenic index (TI) and the desaturase index 18. No significant TS × FI interactions were observed for lipid quality indices or desaturase indices.

**Table 5.** Nutritional quality indices (calculated from % FA).

Index	Type of silage (TS)		Fattening intensity (FI)		SEM	P-value		
	Grass silage	Maiz silage	Intensive (I)	Semi-intensive (SI)		TS	FI	TS×FI
AI	0.66	0.67	0.63	0.69	0.016	0.838	0.041	0.191
TI	1.40	1.53	1.41	1.51	0.038	0.043	0.176	0.474
h/H	1.61	1.58	1.52	1.67	0.032	0.602	0.023	0.932
Desaturase index 16	0.14	0.12	0.12	0.14	0.006	0.256	0.539	0.091
Desaturase index 18	0.74	0.70	0.71	0.73	0.009	0.046	0.160	0.214

AI—atherogenic index; TI—thrombogenic index; h/H—hypcholesterolemic to hypercholesterolemic fatty acids ratio.

### 3.4. Fatty Acids Expressed per 100 g of Meat and Interaction Effects

When expressed as g per 100 g of meat, selected fatty acids were influenced primarily by feeding intensity, reflecting differences in IMF content (Table 6). To facilitate interpretation of the significant TS × FI interactions detected in the relative fatty acid composition, selected interaction effects are presented separately in Table 7. These results illustrate that the combined effects of silage type and feeding intensity modulated the deposition of specific fatty acids in intramuscular fat.

**Table 6.** Selected fatty acids expressed as g per 100 g of meat (calculated from IMF and % FA).

Fatty acid (g/100 g meat)	Type of silage (TS)		Fattening intensity (FI)		SEM	P-value		
	Grass silage	Maiz silage	Intensive (I)	Semi-intensive (SI)		TS	FI	TS×FI
SFA	1.12	1.31	1.52	0.92	0.084	0.222	0.000	0.620
MUFA	1.23	1.32	1.56	1.01	0.085	0.599	0.001	0.706
PUFA	0.16	0.17	0.18	0.15	0.012	0.943	0.334	0.183

CLA	0.010	0.011	0.013	0.008	0.001	0.767	0.000	0.752
$\Sigma$ n-3	0.027	0.024	0.028	0.023	0.002	0.296	0.370	0.150
$\Sigma$ n-6	0.011	0.012	0.012	0.011	0.001	0.993	0.673	0.145
EPA+DHA	0.003	0.003	0.003	0.003	0.000	0.892	0.272	0.860

SFA—saturated fatty acids; MUFA—monounsaturated fatty acids; PUFA—polyunsaturated fatty acids; CLA—conjugated linoleic acid;  $\Sigma$ n-3—sum of n-3 polyunsaturated fatty acids;  $\Sigma$ n-6—sum of n-6 polyunsaturated fatty acids; EPA+DHA—sum of eicosapentaenoic and docosahexaenoic acids.

**Table 7.** Effects of silage type and feeding intensity combinations on selected fatty acid traits of the longissimus lumborum muscle.

Fatty acid (% total FA)	Grass silage		Maize silage		SEM	P-value
	Intensive (I)	Semi-intensive (SI)	Intensive (I)	Semi-intensive (SI)		
C15:0	0.38 <sup>b</sup>	0.37 <sup>b</sup>	0.47 <sup>a</sup>	0.38 <sup>b</sup>	0.013	0.041
C17:0	0.94 <sup>b</sup>	1.01 <sup>ab</sup>	1.17 <sup>a</sup>	0.98 <sup>ab</sup>	0.033	0.027
C18:3 n-3 (ALA)	0.48 <sup>ab</sup>	0.61 <sup>a</sup>	0.51 <sup>ab</sup>	0.44 <sup>b</sup>	0.022	0.018
DPA (C22:5 n-3)	0.14 <sup>b</sup>	0.30 <sup>a</sup>	0.15 <sup>b</sup>	0.18 <sup>b</sup>	0.016	0.026
$\Sigma$ n-3	0.88 <sup>b</sup>	1.28 <sup>a</sup>	0.86 <sup>b</sup>	0.90 <sup>b</sup>	0.050	0.029

Values are presented as means  $\pm$  SEM. GS-I, grass silage with intensive feeding; GS-SI, grass silage with semi-intensive feeding; MS-I, maize silage with intensive feeding; MS-SI, maize silage with semi-intensive feeding. Different letters within a row indicate significant differences between treatment combinations at  $P < 0.05$  (Tukey's post hoc test). TS, silage type; FI, feeding intensity; ALA— $\alpha$ -linolenic acid; DPA—docosapentaenoic acid.

#### 4. Discussion

The observed differences in intramuscular fatty acid composition resulted from the combined effects of dietary energy supply, forage type and ruminal metabolism, which jointly regulate lipid deposition and modification in ruminant tissues [25,26]. Feeding intensity primarily affected intramuscular fat content and the absolute deposition of fatty acids, whereas silage type and its interaction with feeding intensity modulated the qualitative fatty acid profile (Tables 3–7).

The higher intramuscular fat content observed under intensive feeding can be explained by increased dietary energy availability exceeding the requirements for maintenance and lean tissue growth, thereby favouring lipogenesis in intramuscular adipocytes [13,27]. In ruminants, de novo synthesis of fatty acids in adipose tissue relies mainly on acetate and  $\beta$ -hydroxybutyrate produced during ruminal fermentation, and diets with a higher concentrate proportion typically enhance insulin secretion and lipogenic enzyme activity [13,28]. Consequently, increased intramuscular fat content under intensive feeding is commonly associated with higher absolute amounts of fatty acids expressed per unit of meat, as observed in the present study (Table 6), and has been reported previously by Morales et al. [2], Nogalski et al. [3] and Dykier et al. [18].

Differences in odd-chain fatty acids (C15:0 and C17:0), together with significant TS  $\times$  FI interactions, likely reflect alterations in ruminal microbial synthesis. Odd-chain fatty acids originate predominantly from microbial cell membranes and are incorporated into animal tissues following intestinal absorption of microbial lipids [26,29]. Changes in forage type and feeding intensity modify ruminal pH, passage rate and microbial community structure, thereby affecting the contribution of odd-chain fatty acids to intramuscular fat [1,2].

Silage type influenced the proportions of long-chain polyunsaturated fatty acids, particularly within the n-3 family, which can be mechanistically linked to differences in precursor supply and ruminal biohydrogenation. Grass silage provides higher concentrations of  $\alpha$ -linolenic acid (C18:3

n-3), whereas maize silage is richer in linoleic acid (C18:2 n-6) [1,5]. Although extensive biohydrogenation of unsaturated fatty acids occurs in the rumen, a fraction of these fatty acids or their intermediates escapes hydrogenation and is absorbed in the small intestine [25,26]. Subsequently,  $\alpha$ -linolenic acid may serve as a substrate for elongation and desaturation processes in animal tissues, leading to the formation of long-chain n-3 fatty acids such as EPA and DPA [15,30]. The observed effects of silage type on n-3 fatty acids and the n-6/n-3 ratio are therefore consistent with established dietary modulation of fatty acid precursors.

The significant interactions between silage type and feeding intensity for selected fatty acids and fatty acid groups indicate that increased energy supply modifies fatty acid deposition differently depending on the forage base of the diet (Tables 3, 4 and 7). Similar interaction patterns have been reported by Morales et al. [2] and Torrecilhas et al. [11], suggesting that feeding intensity does not uniformly affect fatty acid metabolism but interacts with forage-derived lipid supply and ruminal fermentation characteristics. In line with this concept, Momot et al. [8] demonstrated that changes in feeding intensity alone were sufficient to alter the proportions of PUFA and the n-6/n-3 ratio in beef, highlighting the sensitivity of fatty acid deposition to dietary energy supply even under a constant forage base.

Despite changes in individual fatty acids, nutritional quality indices such as AI, TI and h/H were only moderately affected and showed no significant interactions. This indicates that compensatory shifts among different fatty acid classes may stabilise composite lipid quality indices, even when individual fatty acids respond to dietary treatments [4,31].

Overall, the present results confirm that feeding intensity predominantly regulates the extent of intramuscular lipid deposition, whereas silage type determines the qualitative fatty acid profile through differences in precursor supply and ruminal biohydrogenation. The interaction between these factors highlights the complexity of nutritional control over lipid metabolism in beef cattle, as previously described in studies focusing on forage-based and mixed feeding systems [7,25].

## 5. Conclusions

The results of this study demonstrate that both feeding intensity and silage type significantly influence the fatty acid composition of intramuscular fat in finishing Holstein-Friesian bulls. Feeding intensity was the primary factor determining the amount of intramuscular fat and the absolute deposition of fatty acids in beef, whereas silage type mainly shaped the qualitative fatty acid profile, particularly with respect to n-3 fatty acids and the n-6/n-3 ratio.

The interaction between silage type and feeding intensity further modulated selected fatty acids and fatty acid groups, indicating that the response to increased dietary energy supply depended on the forage base of the diet. These findings highlight the importance of considering combined dietary strategies rather than isolated nutritional factors when aiming to modify the lipid composition of beef.

Despite changes observed in individual fatty acids, nutritional quality indices showed relatively limited variation, suggesting a degree of stability in overall lipid quality of beef under different feeding regimes. Overall, the present study confirms that targeted manipulation of feeding intensity and silage type can be used to influence both the quantity and composition of intramuscular fat, providing a nutritional basis for optimising beef fatty acid profiles under practical finishing conditions.

**Author Contributions:** Conceptualization, methodology, software, validation, formal analysis, writing—original draft preparation, writing—review and editing, Z.N.; project administration, investigation, resources, data curation, supervision, funding acquisition, M.M. All authors have read and agreed to the published version of the manuscript.

**Funding:** This work was supported by the Minister of Science under the Regional Initiative of Excellence Program.

**Institutional Review Board Statement:** The experiment was approved by the Local Ethics Committee for Animal Experimentation at the University of Warmia and Mazury in Olsztyn (Decision. No. 08/2020; 28 January 2020).

**Informed Consent Statement:** Not applicable.

**Data Availability Statement:** The data presented in this study are available from the corresponding author upon reasonable request.

**Conflicts of Interest:** The authors declare no conflict of interest.

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