Buletin Peternakan 48 (1): 64-69, February 2024

ISSTAP
Indonesian Society for Sustainable Tropical Animal Production

Bulletin of Animal Science

ISSN-0126-4400/E-ISSN-2407-876X Accredited: 36a/E/KPT/2016
http://buletinpeternakan.fapet.ugm.ac.id/

Doi: 10.21059/buletinpeternak.v48i1.86454

Fatty Acid Profiling of Bali and Wagyu Cattle using Principal Component Analysis

Dairoh[1], Sutikno[3], Andi Baso Lompengeng Ishak[3], Rudy Priyanto[1], Cece Sumantri[1], Mokhamad Fakhrul Ulum[2], and Jakaria[1]*

[1]Department of Animal Production and Technology, Faculty of Animal Science, IPB University, Bogor, 16680, Indonesia
[2]Division of Reproduction and Obstetrics, School of Veterinary Medicine and Biomedical Sciences, IPB University, Bogor, 16680, Indonesia
[3]Indonesian Agency for Agricultural Instrument Standardization, Ministry of Agriculture, Indonesia

Article history
Submitted: 3 July 2023
Accepted: 12 December 2023

* Corresponding author:
E-mail: jakaria@apps.ipb.ac.id

ABSTRACT

This study aimed to compare the fatty acid profiles of Bali cattle and Wagyu cattle. A total of 50 beef was used in this study, consisting of 44 Bali cattle from Kupang, NTT, and 6 samples of Wagyu cattle from supermarkets. The fatty acid profiles identified are saturated and unsaturated (MUFA and PUFA). The fatty acid analysis used is the Gas Chromatography (GC) method. Descriptive analysis was used to examine data on the fatty acid profile, and T-test analyzed fatty acid composition differences between Bali and Wagyu beef. Differences in fatty acid compositions have been reported based on breeds. The Bali beef had significantly ($p<0.05$) higher saturated fatty acid than Wagyu. Several fatty acids of meat from Bali cattle were significantly different ($p<0.05$) from Wagyu, except for myristoleic (C14:0) and palmitoleic acids (C16:0) did not show significant differences ($p>0.05$). The principal component analysis (PCA) results showed that the first principal component was UFA, MUFA: SFA ratio, oleic acid, omega-9, MUFA, and palmitic acid. In contrast, the second principal component was myristoleic acid, linolenic acid, omega-3, PUFA: SFA ratio, PUFA, omega-6, linoleic acid, stearic acid, SFA, and palmitoleic acid. The study's findings revealed that Bali beef had a much more saturated fatty acid composition of Bali beef was higher than Wagyu beef. This result suggests that Wagyu cattle have a more favorable fatty acid profile, which benefits health.

Keywords: Bali cattle, Fatty acid, Principal component analysis, Wagyu cattle

Introduction

Beef is among the most popular and widely consumed animal protein sources worldwide. The fatty acid composition of beef is important in its nutritional quality and health benefits. One of the important economic properties related to meat quality is the fatty acid composition, both saturated and unsaturated, which is related to flavor and health safety. Fatty acids are the building blocks of fat and are an essential component of the human diet (Simopoulos*,* 2016). Fatty acids regulate body functions, growth, cell development, and other biological processes (Calder*,* 2015).

The fatty acid composition of beef can vary depending on feed type, genetics, and rearing environment. There are two main types of fatty acids in beef: saturated fatty acids and unsaturated fatty acids. Saturated fatty acids are generally found in beef in fairly high amounts. Saturated fatty acids are usually found in high amounts in beef. Some examples of saturated fatty acids that are common in beef are palmitic acid (C16:0) and stearic acid (C18:0). Several previous studies have reported that consumption of excessive amounts of saturated fatty acids can be harmful to consumers' health, such as the increased risk of heart disease, stroke, and other health problems (Zong *et al.,* 2016; Mozaffarian and Wu, 2011). Therefore, it is recommended to limit the consumption of saturated fatty acids in the daily diet (Micha *et al.,* 2012).

The fatty acid content of beef is not all negative as beef also contains unsaturated fatty acids, including omega-3 and omega-6 fatty acids, known to have health benefits. Omega-3 fatty acids, such as eicosapentaenoic acid (EPA) and docosahexaenoic acid (DHA), are associated with reduced risk of heart disease, inflammation, and improved brain function (Calder, 2017). Although the omega-3 content in beef is lower compared to other major sources of omega-3s, such as fatty fish, consumption of beef containing omega-3 fatty acids can still provide benefits in meeting the daily needs for these nutrients. Omega-6 fatty acids are also found in beef and are essential fatty acids the body requires for normal functioning (Simopoulos, 2002). However, it is important to maintain a balance between omega-6 and omega-3 fatty acid intake (Patterson *et al.,* 2012). Excessive imbalance in omega-6 intake can lead to chronic inflammation and contribute to the risk of heart disease, type 2 diabetes, and obesity (Mozaffarian and Wu, 2011). Therefore, it is wise to pay attention to omega-6 intake and strive to achieve a balanced balance with omega-3 fatty acids (Calder, 2016).

Some of the health implications of beef fatty acid components are important to consider not only individual factors but also the combined factors of genetics and environment. Feeding diets such as grass-fed or grain-fed diets have been shown to influence changes in the fatty acid composition of beef. Grass-fed cattle showed lower saturated fatty acid (SFA) content and higher omega-3 and conjugated linoleic acid (CLA) concentrations compared to grain-fed cattle (Nogoy *et al.,* 2022). Daley et al. (2010) also reported that grass-fed cattle tend to increase saturated fatty acids such as stearic acid (C18:0), which is one of the saturated fatty acids that are neutral to cholesterol levels or not harmful to the health of the body (Hunter *et al.,* 2010). Meanwhile, cows fed grains may increase saturated fatty acids such as myristic acid (C14:0) and palmitic acid (C16:0), raising cholesterol levels.

One of Indonesia's indigenous cattle breeds, Bali cattle, is known for its small frame and low-fat content. Small-framed and low-fat cattle tend to have healthier fatty acid profiles, such as high unsaturated fatty acids and omega-3 fatty acids, which are healthier than imported beef (Wood *et al.,* 2008). Based on the information, we know that the information on fatty acid composition in Indonesian cattle, especially Balinese cattle, still needs to be improved. As a result, this study aims to investigate the fatty acid profile of two different cattle breeds, namely Bali and Wagyu cattle.

Materials and Methods

Beef samples

A total of 50 beef samples of the Bali (n=28; males) and Wagyu cattle (n=6) breeds were used in this study. Bali cattle were raised in Kupang, the East Nusa Tenggara Province, and then transported by ship to the slaughterhouse. The Bali cattle were slaughtered in the Basirih slaughterhouse at South Banjarmasin of South Kalimantan Province. Bali cattle that were slaughtered were obtained from ages I1 – I3 (18 – 35 mon). After slaughter, the *longissimus dorsi* muscle was sampled and cut into 250 grams. Wagyu beef was commercial beef imported from Japan and obtained from the same market.

Fatty acid profiling

Fatty acid profile analysis used meat samples between ribs 12-13 (*longissimus dorsi*) for Bali cattle and *semitendinosus* muscle for Wagyu. The analysis of fatty acid profiles, including both saturated and unsaturated fatty acids, was analyzed using the AOAC (2012) (Association of Official Analytical Chemists) extraction method 991.36 and AOAC 969.333. The fatty acid composition was measured using gas chromatography (GC). A total of 40 g of meat was subjected to lipid extraction using a chloroform-methanol solution. The extracted lipids were then converted into fatty acid methyl esters (FAMEs) through transesterification. The FAMEs were extracted using a hexane solution, centrifugation, and drying. The resulting FAME products were dissolved in a chloroform solution and filtered to remove unwanted compounds using solid-phase extraction. FAMEs were injected into the gas chromatography machine in a volume of 1 µL. The separated fatty acids were detected and measured using a mass spectrometer. The retention time and peak measurements are recorded for each fatty acid component. Information about each fatty acid component is obtained by comparing the retention time with standards. GC found saturated, monounsaturated, and polyunsaturated fatty acids.

Data analysis

The unpaired T-test was used in the statistical analysis to determine the difference in fatty acid composition of Bali and Wagyu cattle breeds (Gasperz, 1998). The principal component analysis (PCA) was performed using R Program (Giaretta *et al.,* 2018). Moreover, the fatty acid profile data was also corrected to 36 months of age by Salamena and Papilaja's (2010) formula as follows:

$$\text{Xi corrected} = \left[\frac{\bar{X}_{\text{standard}}}{\bar{X}_{\text{observation}}}\right] \text{ x X observation value i}$$

Where $X_{i\ corrected}$ is corrected data i; $\bar{X}_{standard}$ is standard group average; $\bar{X}_{observation}$ is observation group average; and $X_{observation\ value\ I}$ is observation value i.

Results and Discussion

Fatty acid profile in different breeds

In this study, fatty acid composition analyses of Bali and Wagyu breeds were performed using GC methods. The fatty acid compositions are presented in Table 1. Palmitic acid (C16:0) and stearic acid (C18:0) were beef's most prominent saturated fatty acids. In contrast, oleic acid (C18:1n9c) was the predominant unsaturated fatty acid, a finding consistent with the study of Santana *et al.* (2023). It has been reported that fatty acid compositions are different depending on breed. Bali beef has the highest stearic acid (C18:0) content, while Wagyu beef has the highest palmitic fatty acid (C16:0) content. This discrepancy in the composition of the most abundant saturated fatty acids between the two breeds, namely stearic acid (C18:0) in Balinese cattle and palmitic acid (C16:0) in Wagyu cattle, implies that environmental factors, such as dietary intake, can influence alterations in meat fatty acid composition. Daley *et al.* (2010) reported that grain-fed cattle contained higher palmitic (C16:0) and myristic (C14:0) fatty acids compared to grass-fed. Both of these fatty acids were associated with increased serum cholesterol and low-density lipoprotein (LDL) levels, leading to cardiovascular disease and adversely affecting overall health

(Tarino *et al.,* 2010). High stearic acid (C18:0) in beef does not contribute to increased cholesterol because stearic acid is one of the saturated fatty acids that are neutral to cholesterol levels and are considered harmless to health (Williamson *et al.,* 2005). Stearic acid (C18:0) desaturates to oleic acid (C18:1n9c) during the metabolic process; therefore, consumption of stearic acid as a supplement does not affect blood lipids and one of the main risk factors for cardiovascular disease (Baer *et al.,* 2004).

The predominant unsaturated fatty acid in both Bali and Wagyu beef was oleic acid (C18:1n9c). This study was aligned with previous research that has consistently identified oleic acid (C18:1n9c) as having the highest concentration among unsaturated fatty acids (Missio *et al.,* 2017; Fiorentini *et al.,* 2018). The results showed that the average content of oleic acid (C18:1n9c) in Bali beef was lower than in Wagyu beef but higher than the results of Bain *et al.* (2016) for Bali cattle (6.30%) with a diet consisting of fed 40% grass and 60% concentrate. Among other unsaturated fatty acids are polyunsaturated fatty acids (PUFA), characterized by hydrocarbon chains containing two or more double bonds and often categorized as omega-3 and omega-6. Burillo *et al.* (2012) discovered that omega-3 sourced from polyunsaturated fatty acids (PUFA) can contribute to reducing cholesterol levels and increasing high-density lipoprotein (HDL) serum levels. Linoleic acid (C18:2n6c) is a conjugated fatty acid in the PUFA group. In this study, Bali beef exhibited a higher level of linoleic acid (C18:2n6c) at 1.80% compared to Wagyu content of 0.44%.

Table 1 shows that the fatty acid composition of Bali beef and Wagyu beef were significantly different ($p<0.05$), except for palmitoleic acid (C16:1) and myristoleic acid (C14:1), which were not significantly different ($p>0.05$). Fatty acid composition in Bali beef was higher than Wagyu for stearic acid (C18:0), linoleic (C18:2n6c), linolenic (C18:3n3), PUFA: SFA ratio, omega-3 and omega 6. Stearic acid significantly differed ($p<0.05$) between Bali and Wagyu beef. Bali beef had much more stearic acid than Wagyu beef. In terms of palmitic acid composition, Balinese beef had relatively less palmitic acid than Wagyu beef. In a previous study, palmitic acid, an SFA, had a negative effect on beef meat flavor (Scollan *et al.,* 2014). The research findings indicate that the highest total saturated fatty acid composition was found in Bali beef, reaching 58.18%, whereas Wagyu beef exhibits a lower content of saturated fatty acids at 38.87%. These results underscore a significant difference in the fatty acid profiles between the two beef types. In this context, Bali beef tends to have a higher concentration of saturated fats than Wagyu beef. The tendency for Bali beef to have a higher saturated fat content could be attributed to the feeding system. Bressan *et al.* (2011) reported that grass-fed diets tend to be rich in saturated fatty acids, particularly stearic acid (C18:0). Considering that Bali cattle obtained from Kupang have been reared under a grazing system and primarily rely on grass feed, these findings are consistent with the reported trends.

The average polyunsaturated fatty acid composition derived from omega-3 and omega-6 in Bali beef was higher (2.72%) than in Wagyu beef (0.61%). Bressan *et al.* (2011) reported that *Bos indicus* cattle had higher PUFA values than *Bos taurus* cattle reared on the same feed. That indicates that cattle breed factors can affect the composition of PUFA in meat. In human nutrition, there are two important types of essential fatty acids: linolenic acid (LA), which belongs to the omega-3 fatty acid group, and linoleic acid (LA), which belongs to the omega-6 fatty acid group. Although omega-3 (n-3) and omega-6 (n-6) fatty acids come from different families, they can be synthesized in the body through the same enzymes, mainly delta-5- desaturase and delta-6-desaturase (Ruxton *et al.,* 2004). Generally, a high intake of omega-3 fatty acids is associated with a lower prevalence of age-related memory decrease and a lower risk of developing Alzheimer's disease. In addition, omega-3s may reduce the risk of cardiovascular disease leading to heart attacks (Colussi *et al.,* 2017). Omega-6 fatty acids play an important role in cattle health and can have positive effects. Some positive effects of omega-6 fatty acids in cattle include regulating cell function, growth, development, and hormone balance (Vargas-Bello-Pérez *et al.,* 2018). Omega-6 fatty

Table 1. Fatty acid composition in Bali and Wagyu cattle

Fatty acid composition (% w/w)	Bali (n = 44)	Wagyu (n = 6)
Saturated fatty acid (SFA)	58.18 ± 6.66[a]	38.87 ± 2.64[b]
Palmitic acid (C16:0)	21.09 ± 4.01[b]	28.78 ± 1.29[a]
Stearic acid (C18:0)	32.02 ± 3.67[a]	8.42 ± 1.40[b]
Monounsaturated fatty acid (MUFA)	17.37 ± 5.06[b]	43.48 ± 1.42[a]
Palmitoleic acid (C16:1)	1.32 ± 0.36[a]	0.72 ± 0.67[a]
Oleic acid (C18:1n9c)	12.17 ± 4.83[b]	42.41 ± 0.97[a]
Myristoleic Acid (C14:1)	0.28 ± 0.37[a]	0.20 ± 0.04[a]
Polyunsaturated fatty acid (PUFA)	2.72 ± 1.14[a]	0.61 ±0.11[b]
Linoleic acid (C18:2n6c)	1.80 ± 0.82[a]	0.44 ± 0.10[b]
Linolenic acid (C18:3n3)	0.22 ± 0.26[a]	0.08 ± 0.005[b]
MUFA : SFA	0.30 ± 0.09[b]	1.12 ± 0.09[a]
PUFA : SFA	0.05 ± 0.02[a]	0.02 ± 0.002[b]
Omega-3	0.42 ± 0.73[a]	0.08 ± 0.01[b]
Omega-6	2.09 ± 0.95[a]	0.50 ± 0.11[b]
Omega-9	15.28 ± 5.09[b]	42.44 ± 1.07[a]
Unsaturated fatty acid (UFA)	20.09 ± 4.53[b]	44.09 ± 1.38[a]

acids also contribute to the immune system and play a role in cell membrane formation and hormone synthesis (Bessa *et al.,* 2017).

Bali beef has the potential to produce quality meat with good nutrition for health. These results are the basis of information that there is potential to develop high-quality premium Bali beef in terms of physical properties and good nutrition. Fatty acids are quantitative traits strongly influenced by genetics, environment, and their interactions. Producing Bali beef with good nutritional quality must be supported by a controlled maintenance system such as feeding management, comfortable cages, and cattle that are not stressed. Genetic factors involve several candidate genes that have an important role in fatty acid synthesis, including Diacylglycerol acyltransferase 1 (DGAT1), Thyroglobulin (TG), Fatty acid-binding proteins (FABP4), Fatty acid synthase (FASN), and Stearoyl-CoA desaturase (SCD) genes (Pećina and Ivanković, 2021). The variation of these candidate genes in different cattle breeds will affect the fatty acid composition of meat. Genetic differences can be observed between animals in different breeds or strains, as well as as a result of breed crosses (Rossato *et al.,* 2010). Several studies have shown that in adipose tissue, the *Bos indicus* breed's cattle have lower saturation levels than the *Bos taurus* breed (Rossato *et al.,* 2010). Nutritionally, Nellore beef, which belongs to the *Bos indicus* breed, is known to have an advantage over the Angus breed. That is evident from its lower cholesterol and higher content of n-3 fatty acids, including conjugated linoleic fatty acid (CLA) precursors such as C18:1 trans fatty acid (Bressan *et al.,* 2011).

Fatty acid profile markers in different breeds

Principal component analysis (PCA) of fatty acid profiles of different cattle breeds was presented in Figure 1. The first principal component, the Wagyu cattle breed, has the highest total diversity of 58.4%, while Bali cattle, as the second principal component, has a diversity of 13.3%. Figure 1 shows two right and left clusters characterized by the fatty acid profiles of different cattle breeds. The fatty acid profile characteristic of the first cluster in Wagyu cattle was total unsaturated fatty acids (UFA), MUFA: SFA ratio, oleic acid, omega-9, total monounsaturated fatty acids (MUFA), and palmitic acid, while Bali cattle as the second cluster was characterized by myristoleic acid, linolenic acid, omega-3, PUFA: SFA ratio, total polyunsaturated fatty acids (PUFA), omega-6, linoleic acid, stearic acid, total saturated fatty acids (SFA), and palmitoleic acid. The fatty acid profile characteristics that genetically distinguish the two groups of cattle breeds indicate that fatty acid profiles are influenced by genetic and environmental factors (Hardjosubroto, 1998).

Differences in fatty acid profiles in the two cattle breeds may be due to genetic factors and husbandry management differences. Sakowski *et al.* (2022) stated that in addition to genetic factors,

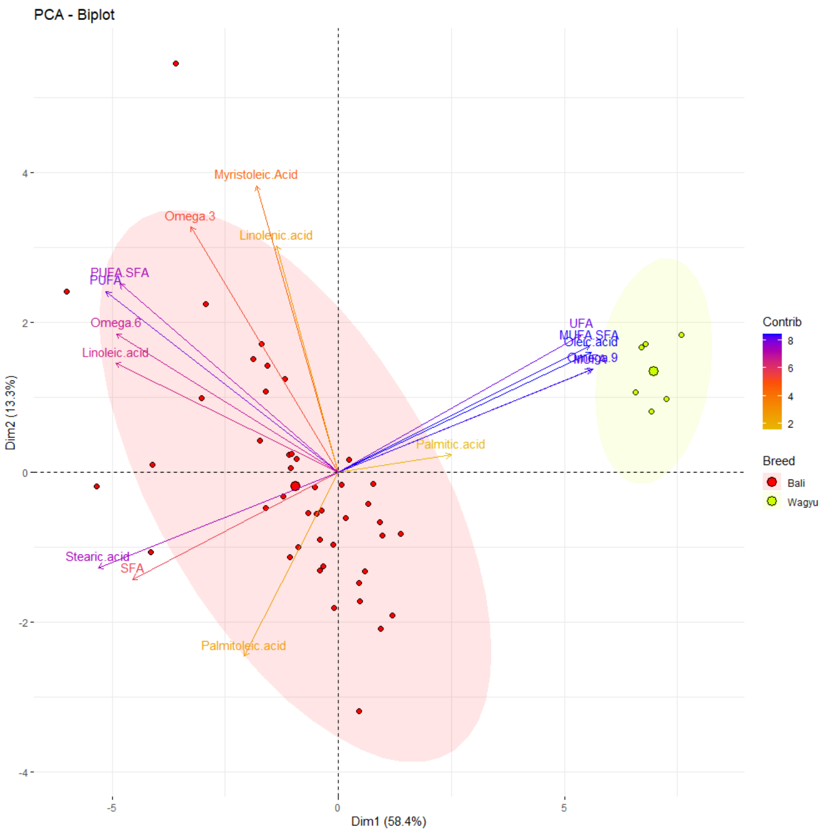

Figure 1. Principal component analysis (PCA) results in different breeds.

differences in fatty acid profiles are caused by differences in environmental factors, including maintenance management, especially feed management. In a study conducted by Sakowski *et al.* (2022), it was found that grass-fed beef had a lower total fat content compared to grain-fed beef. In addition, grass-fed also showed a more favorable fat profile in terms of monounsaturated fatty acids (SFA) and levels of long-chain n-3 polyunsaturated fatty acids (PUFA; EPA, DPA, DHA). The results of this study indicate that Balinese cattle contain fatty acids of equal or better quality than Wagyu beef, which may offer more health benefits to consumers.

Conclusion

Bali beef is superior in the content of polyunsaturated fatty acids (PUFA) of the omega-3 and omega-6 types that are good for consumer health compared to wagyu beef. Bali beef contains more saturated fatty acids than wagyu beef but has a higher unsaturated fatty acid ratio (PUFA: SFA) than wagyu beef. The components of the UFA, MUFA: SFA ratio, oleic acid, omega-9, MUFA, and palmitic acid characterize fatty acids in Wagyu cattle. In contrast, myristoleic acid, linolenic acid, omega-3, PUFA: SFA ratio, PUFA, omega-6, linoleic acid, stearic acid, SFA, and palmitoleic acid characterize the fatty acids in Bali cattle.

Conflict of interest

The authors declare that there are no conflicts of interest concerning financial, personal, or other relationships with other persons or organizations to the subject matter discussed in the manuscript.

Funding statement

This research received was supported by Ministry of Education, Culture, Research, and Technology and Research and Innovation for Advanced Indonesia-Association of Research and Innovation in Indonesia-RIIM (grant number: 4830/IT3.L1/PT.01.03/P/B/2022) and the authors were grateful to the Department of Food Security, Agriculture, and Fisheries, South Banjarmasin, South Kalimantan.

Acknowledgment

The authors gratefully acknowledge the support from National Research and Innovation Agency (BRIN) via Research and Innovation for Advanced Indonesia (RIIM) with contract number: 4830/IT3.L1/PT.01.03/P/B/2022.

Author's contribution

Dairoh was responsible for the experiments, data analysis, and writing the first draft. Sutikno, Andi Baso Lompengeng Ishak, Rudy Priyanto, Cece Sumantri, Mokhamad Fakhrul Ulum, and Jakaria were responsible for study design, data resources, and data validation. The submitted version was read and approved by all authors.

Ethics approval

This experiment had no negative consequences for the animals' welfare. All procedures in this study have been approved by the Banjarmasin City Food Security, Agriculture, and Fisheries Agency's Animal Ethics Committee (approval ID:520/624/DKP3/XII/2021).

References

AOAC. 2012. Officials Methods of Analysis. 19th ed. Association of Official Analytical Chemists, Arlington.

Baer, D. J., T. T. Judd, B. A. Clevidence, and R. P. Tracy. 2004. Dietary fatty acids affect plasma markers of inflammation in healthy men fed controlled diets: a randomized crossover study. Am. J. Clin. Nutr. 79: 969-973.

Bain, A., D. A. Astuti, S. Suharti, C. Arman, and K. G. Wiryawan. 2016. Performance, nutrient digestibility, and meat quality of Bali cattle fed a ration supplemented with soybean oil, calcium soap and cashew fruit flour. Med. Pet. 39: 180-188.

Bessa, R. J., S. P. Alves, J. Santos-Silva, and F. Santos-Silva. 2017. Effect of lipid supplements on ruminal biohydrogenation intermediates and muscle fatty acids in lambs. Anim. 11: 1162-1170.

Bressan, M. C., L. V. Rossato, E. C. Rodrigues, S. P. Alves, R. J. Bessa, E. M. Ramos, and L. T. Gama. 2011. Genotype x environment interactions for fatty acid profiles in Bos indicus and Bos taurus finished on pasture or grain. J. Anim. Sci. 89: 221-232.

Burillo, E., R. Mateo-Gallego, A. Cenarro, S. Fiddyment, A. M. Bea, I. Jorge, J. Vázquez, and F. Civeira. 2012. Beneficial effects of omega-3 fatty acids in the proteome of high-density lipoprotein proteome. Lipids. Health. Dis. 11: 116.

Calder, P. C. 2015. Functional roles of fatty acids and their effects on human health. J. Parent. Ent. Nut. 39: 18S-32S.

Calder, P. C. 2016. Omega-3 fatty acids and inflammatory processes: from molecules to man.Biochem. Soc. Trans. 45: 1105-1115.

Calder, P. C. 2017. Omega-3 fatty acids and inflammatory processes: from molecules to man. Biochem. Soc. Trans. 45: 1105-1115.

Colussi, G., C. Catena, M. Novello, N. Bertin, and L. A. Sechi. 2017. Impact of omega-3 polyunsaturated fatty acids on vascular function and blood pressure: relevance for cardiovascular outcomes. Nutr. Metab. Cardiovasc. Dis. 27: 191–200.

Daley, C. A., A. Amber, P. S. Doyle, G. A. Nader, and S. Larson. 2010. A review of fatty acid profiles and antioxidant content in grass-fed and grain-fed beef. Nutr. J. 9: 10.

Fiorentini, G., M. O. Santana, J. D. Messana, A. L. S. Valente, C. J. Harter, C. H. S. Rabelo, R. P. Barbero, D. P. D. Lanna, R. A. Reis, and T. T. Berchiell. 2018. Effect of lipid sources on the fatty acid profile of meat from pasture-and feedlot-finished Nellore bulls. Lives. Sci. 211: 51-60.

Gasperz, V. 1998. Teknik Analisis dalam Penelitian Percobaan. Penerbit Tarsito, Bandung.

Giaretta, E., A. L. Mordenti, G. Canestrari, N. Brogna, A. Palmonari, and A. Formigoni. 2018. Assessment of muscle longissimus thoracis et lumborum marbling by image analysis and the relationship between meet quality parameters. Plos One. 13: 1-12.

Hardjosubroto, W. 1998. Aplikasi Pemuliabiakan di Lapangan. Gramedia Widiasarana Indonesia, Jakarta.

Hunter, J. E., J. Zhang, and P. M. Kris-Etherton. 2010. Cardiovascular disease risk of dietary stearic acid compared with trans, other saturated, and unsaturated fatty acids: A systematic review. Am. J. Clin. Nutr. 91: 46-63.

Micha, R., G. Michas, and D. Mozaffarian. 2012. Unprocessed red and processed meats and risk of coronary artery disease and type 2 diabetes – An updated review of the evidence. Current Atherosclerosis Reports. 14: 515-524.

Missio, R. L., J. Restle, A. K. Freitas, M. E. Lage, P. S. Pacheo, U. O. Bileg, and J. T. Padua. 2017. Age castration of Nellore males on profile of fatty acids of meat. Semina: Ciências Agrárias, Londrina. 38: 3739-3748.

Mozaffarian, D. and J. H. Wu. 2011. Omega-3 fatty acids and cardiovascular disease: effects on risk factors, molecular pathways, and clinical events. American. Coll. Card. J. 58: 2047-2067.

Nogoy, K. M. C., B. Sun, S. Shin, Y. Lee, X. Z. Li, S. H. Choi, and S. Prak. 2022. Fatty Acid Composition of Grain- and Grass-Fed Beef and Their Nutritional Value and Health Implication. Food. Sci. Anim. Resour. 42: 18-33.

Patterson, E., R. Wall, G. F. Fitzgerald, R. P. Ross, and C. Stanton. 2012. Health implications of high dietary omega-6 polyunsaturated fatty acids. J. Nutr. Metab. 2012: 1-16.

Poćina, M. and A. Ivanković. 2021. Effect of FASN, SCD, and GH genes on carcass fatness and fatty acid composition of intramuscular lipids in F1 Holstein × beef breeds. Agric. 13: 571.

Rossato, L. V., M. C. Bressan, E. C. Rodrigues, L. T. Gama, R. J. B. Bessa, and S. P. A. Alves. 2010. Parâmetros físico-químicos e perfil de ácidos graxos da carne de bovinos Angus e Nelore terminados em pastagem. R. Bras. Zootec. 39: 1127–1134.

Ruxton, C. H. S., S. C. Reed, M. J. A. Simpson, and K. J. Millington. 2004. The health benefits of omega-3 polyunsaturated fatty acids: a review of the evidence. J. Hum. Nut. Diet. 17: 449-459.

Salamena, F. J. and J. Papilaja. 2010. Characterization and genetic relationships analysis of buffalo population in MOA island of South-East West Maluku regency of Maluku Province. J. Indonesian Trop. Anim. Agric. 35: 75-82.

Sakowski, T., G. Grodkowski, M. Gołebiewski, J. Slósarz, P. Kostusiak, P. Solarczyk, and K. Puppel. 2022. Genetic and environmental determinants of beef quality-a review. Front. Vet. Sci. 24: 1-8.

Santana, E. O. C., R. R. Silva, J. I. Simionato, G. T. Junio, T. O. J. Lins, G. D. da Costa, B. M. A. C. Mesquita, H. D. R. Alba, and G. G. P. Calvarho. 2023. Sex effect on the fatty acid profile and chemical composition of meat from beef cattle fed a whole shelled corn diet. Arch. Anim. Breed. 66: 51-60.

Scollan, N. D., D. Dannenberger, K. Nuernberg, I. Richardson, S. Mac Kintosh, J. F. Hocquette, and A. P. Moloney. 2014. Enhancing the nutritional and health value of beef lipids and their relationship with meat quality. Mea Sci. 97: 384–394.

Simopoulos, A. P. 2002. The importance of the ratio of omega-6/omega-3 essential fatty acids. Biomed. Pharma. 56: 365-379.

Simopoulos, A. P. 2016. An increase in the omega-6/omega-3 fatty acid ratio increases the risk for obesity. Nutr. 8: 128.

Tarino, P. W., Q. Sun, F. B. Hu, and R. M. Krauss. 2010. Meta-analysis of prospective cohort studies evaluating the association of saturated fat with cardiovascular disease. Am. J. Clin. Nutr. 91: 535-46.

Vargas-Bello-Pérez, E., N. Wrage, K. Gebauer, T. Von, M. Mielenz, A. Schröder, and G. Jahreis. 2018. Influence of different dietary levels of n-6/n-3 polyunsaturated fatty acids on the performance and fatty acid composition in muscle and subcutaneous fat of beef cattle. Anim. Feed. Sci. Tech. 235: 1-12.

Williamson, C. S., R. K. Foster, S. A. Stanner, and J. L. Buttriss. 2005. Red meat in the diet. Nutr. Bull. 30: 323–355.

Wood, J. D., M. Enser, A. V. Fisher, G. R. Nute, P. R. Sheard, R. I. Richardson, S. I. Hughes, and F. M. Whittington. 2008. Fat deposition, fatty acid composition and meat quality: A review. Meat. Sci. 78: 343–358.

Zong, G., Y. Li, A. J. Wanders, M. Alssema, P. L. Zock, W. C. Willett, F. B. Hu, and Q. Sun. 2016. Intake of individual saturated fatty acids and risk of coronary heart disease in US men and women: Two prospective longitudinal cohort studies. BMJ. 355: 1-11.