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# A Novel SNPs of the *SREBF1* and *SCARB1* Genes and the Association with Fatty Acid Profile in Bali Cattle

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## ABSTRACT

**This study aimed to investigate the genetic impact of single nucleotide polymorphisms (SNPs) of the sterol regulating element binding factor 1 (*SREBF1*) and scavenger receptor class B member 1 (*SCARB1*) genes on carcass and meat characteristics, as well as fatty acid composition, in the Bali cattle. The blood and beef samples used for DNA sequencing, physical assessment, and fatty acid analysis were collected from 95 male Bali cattle. The ultrasound images were analyzed using the Image-J NIH software. A total of 4 SNPs were identified in the *SREBF1* gene and 5 SNPs in the *SCARB1* gene. The results showed that the 4 SNPs in the *SREBF1* gene, namely g.12629T>C, g.12731T>C, g.12881A>G, and g.12986C>T, were associated with heptadecanoic acid (C17:0) and cis-11-eicosanoic acid (C20:1). The SNPs g.12731T>C of the *SREBF1* gene was associated with fat content, palmitoleic acid (C16:1), stearic acid (C18:0), cis-11-eicosanoic acid (C20:1), and total fatty acids. Furthermore, 4 SNPs in the *SCARB1* gene, including g.72219C>T, g.72380C>A, g.72517G>A, and g.72607C>T correlated with *longissimus dorsi* thickness (LDT). All SNPs in the *SCARB1* gene showed significant associations with cis-10 heptadecanoic acid (C17:1) and cis 8,11,14-eicosatrienoic acid (C20:3n6). The SNP g.72400A>G of the *SCARB1* gene was related to caprylic acid (C8:0), lauric acid (C12:0), arachidonic acid (C20:4n6), monounsaturated fatty acids (MUFA), and unsaturated fatty acids (UFA). These results suggested that the identified polymorphisms in the *SREBF1* and *SCARB1* genes could serve as valuable references for investigating similar genes in other cattle breeds, particularly concerning fatty acids.**

***Keywords: association study; Bali cattle; meat quality; SCARB1; SREBF1***

## INTRODUCTION

In Indonesia, four cattle-breeds significantly contribute to the diverse livestock production. These breeds encompass Bali (*Bos javanicus*), Zebu (*Bos indicus*), Taurine (*Bos taurus*), and crossbred (*Bos indicus* x *Bos javanicus* and *Bos indicus* x *Bos taurus*), each serving a distinct role in meat, milk, and working purposes. Bali cattle, originating from the banteng (*Bibos banteng*), are the native cattle of Indonesia, with excellent adaptability to the tropical environment (Talib, 2022). Compared to *Bos taurus* and *Bos indicus*, Bali cattle possess unique morphological traits characterized by compact carcasses and harmonious body shapes, which are considered ideal for meat production. This unique morphological trait makes Bali cattle significantly have a high carcass percentage, with an average of 52.76% (Suryanto *et al*., 2014) compared to Ongole cattle (50%) (Sutarno & Setyawan, 2016), indicating their potential to yield superior-quality meat (Tahuk *et al*., 2018).

Bali cattle have a small frame but can be efficiently fattened and mature young (Littler, 2007). The practice of fattening at a young age accelerates intramuscular fat development compared to larger-framed cattle with a slower fattening period. The presence of fat plays a crucial role in the flavor profile of meat (Iida *et al*., 2015), as it is stored in various body locations, including organ, subcutaneous (under the skin), intermuscular (between the muscles), and intramuscular (marbling within the muscles) (Hall *et al*., 2016). As cattle age and receive proper nutrition, there is an increase in fat content, leading to the production of enhanced flavor (Sakowski *et al*., 2022).

At 3 years of age, Bali cattle possess an intramuscular fat percentage of 4.50% (Jakaria *et al*., 2017), which is particularly rich in monounsaturated fatty acids (MUFA), constituting 46% of its composition and contains 10% polyunsaturated fatty acids (PUFA) (Ladeira *et al*., 2018). These fatty acids are classified as saturated or unsaturated fats based on their structural and chemical properties. Moreover, flavor is a multifaceted aspect of meat delicacies, influenced by various factors such as the diet of cattle. This affects meat texture and flavor by influencing intramuscular fat levels and fatty acid com-

position (Schumacher *et al.,* 2022). For example, linolenic acid is a fatty acid that contributes to meat flavor (Dinh *et al.,* 2021). Cattle fed with a grain-based diet typically exhibit lower levels of linolenic acid than those raised on a grass-fed diet (Arshad *et al.,* 2018).

Several molecular-based approaches have been explored to improve the quality of Bali cattle by investigating their genetic diversity and association with meat quality traits. Dairoh *et al.* (2021) stated that SNPs in the *CAPN1* gene were associated with a marbling score in Bali cattle. Dairoh *et al.* (2022) also stated that the 3'UTR region of the *CAPN1* gene was found in 8 base deletions in Bali cattle. Furthermore, two specific SNPs in the *ADIPOQ* genes (c.-399C>T and c.-273C>G) showed potential as genetic markers of marbling score for Bali cattle (Sutikno *et al.,* 2018).

The sterol regulating element binding factor 1 (*SREBF1)* gene plays a crucial role in regulating animal lipid metabolism and fatty acid synthesis. This gene regulates lipogenesis and gene expression in fat accumulation and composition within muscle tissue (Liang *et al.,* 2020). The *SREBF1* gene is also involved in fatty acid biosynthesis, which is essential for synthesizing fatty acids (Kanehisa *et al.,* 2019). The scavenger receptor class B member 1 (*SCARB1)* gene is critical in lipid metabolism and cholesterol transportation, as it regulates cholesterol absorption and transfer within the body (Chinetti *et al.,* 2000). According to pathway analysis, the *SCARB1* gene is associated with the marbling trait and plays a crucial role in lipid metabolism, export, transport, catabolism, and storage regulation (Park *et al.,* 2012). Furthermore, it is regulated by 17 genes, including insulin, peroxisome proliferator-activated receptors (*PPARs*), *APOA1,* and *FABP1*.

In cattle, the *SREBF1* gene is located on chromosome 19 and has a length of 29.408 base pairs, while the *SCARB1* gene is found on chromosome 17 with a length of 46.860 base pairs (Ensemble database: ENSBTAG00000007884 and ENSBTAG00000014269). Recent studies show that the variations in these genes are related to meat quality in cattle. Gao *et al.* (2022) stated that the 84 bp indel variation in *SREBF1* was associated with intramuscular fat, carcass, and body size of Chinese Qinchuan cattle. Genetic variations in the *SREBF1* gene are associated with meat quality characteristics, including marbling levels (Li *et al.,* 2014). The expression of the *SCARB1* gene can influence meat characteristics such as marbling (intramuscular fat patterns), tenderness, and meat flavor (Li *et al.,* 2018). Consequently, the *SREBF1* and *SCARB1* genes were selected as candidates to determine the associations between single nucleotide polymorphisms (SNPs) and meat quality traits in Bali cattle. During the investigation, there was limited information on the presence of polymorphisms in these genes, specifically in Bali cattle (*Bos javanicus*). Therefore, this study aimed to investigate the genetic effects of SNPs in the *SREBF1* and *SCARB1* genes on carcass and meat characteristics, as well as fatty acid composition, in the Bali cattle population.

## MATERIALS AND METHODS

### Animals and Blood Samples

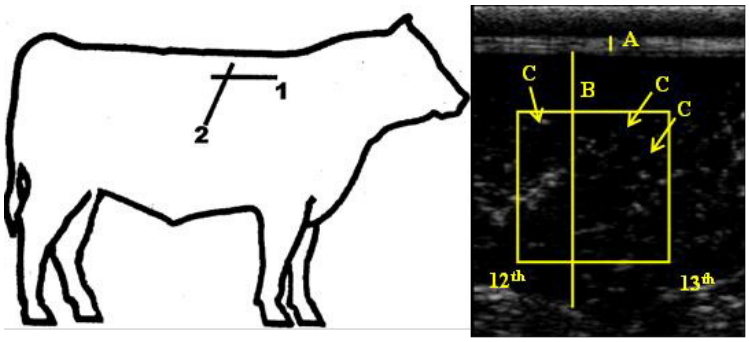
This study used 95 blood samples for DNA isolation (Koshy *et al.,* 2017), 91 ultrasound data, and 44 meat samples. Bali cattle used were male with a body weight of 250-350 kg and ages ranging from 18-36 months. Subsequently, blood samples were collected from the *jugular vein* using project tubes containing 1.5 mL EDTA. This experimental procedure was approved by Animal Ethics Committees from the Department of Food Security, Agriculture, and Fisheries of Banjarmasin City (approval number: 520/624/DKP3/X11/2021). A 250 g meat sample from the tenderloin part of Bali cattle was collected for fatty acid analysis.

### Carcass and Meat Characteristics Data Collection

An ultrasonography device was used to record the carcass and meat characteristics of live Bali cattle (Silva *et al.,* 2012). The ultrasound image data were collected on live cattle using the retrieval technique described by the Beef Improvement Federation (BIF, 2016). Portable ultrasound (SIUI CTS-800, China) was used in brightness mode, with a linear transducer at 7.1 MHz and a depth of 80 mm. Subsequently, back fat thickness (BFT), *longissimus dorsi* thickness (LDT), marbling score (MS), and percentage of intramuscular fat (IMF) were carried out transversally and longitudinally between $12^{th}$-$13^{th}$ thoracic vertebrae following the methods of Jakaria *et al.* (2017), as shown in Figure 1a. This was followed by analyzing the images using Image-J NIH software (ImageJ, NIH, USA). The Image-J software was used to calibrate the scale from units to millimeters. Ultrasound measurements were collected at three points of live Bali cattle following the methods of Crews *et al.* (2016), including back fat thickness, longissimus dorsi thickness, and intramuscular fat. Back fat thickness (A) was measured by drawing a vertical line in the middle, from the fat layer under the skin to the lower back. *Longissimus dorsi* thickness (B) was measured from the bottom of the back fat to the top of the bone. A 30 mm x 30 mm square guideline (C) was employed for measuring intramuscular fat, as shown in Figure 1b. The measurement values of intramuscular fat were subjected to simple linear regression analysis to establish an equation for calculating the intramuscular fat percentage. The marbling was scored according to AUSTRALIAN MEAT and MSA (marbling reference standard). The AUS-MEAT marbling score ranges from 0 to 9 (AUS-MEAT, 2018).

### Fatty Acid Profile

The fatty acid composition was analyzed using the Association of Official Analytical Chemists (AOAC, 2019). In this process, 40 g of meat was subjected to lipid extraction using a chloroform-methanol solution. Transesterification was used to convert the extracted lipids into fatty acid methyl esters (FAMEs). FAMEs were extracted using a hexane solution, centrifugation, and drying. The resulting FAMEs products were dissolved




(a) (b)

Figure 1. (a) Position of ultrasound image scan (USG) on live cattle, 1= longitudinal; 2= transverse (BIF 2016), (b) ultrasound image analysis using Image-NIH at a transverse viewpoint (A= backfat thickness; B= longissimus dorsi thickness; C= intramuscular fat).

in a chloroform solution and filtered to remove unwanted compounds using solid-phase extraction. FAMEs obtained were injected into the gas chromatography machine at 1 µL. The separated fatty acids were detected and measured using a mass spectrometer. Subsequently, each fatty acid component's retention time and peak measurements were recorded. Information about each fatty acid component was obtained by comparing the retention time with standards. The gas chromatography analysis effectively identified and categorized various fatty acids into saturated, monounsaturated, and polyunsaturated fatty.

### Primer Design and DNA Amplification

The *SREBF1* and *SCARB1* primer sequences were designed using an Ensemble genome browser with access codes ENSBTAG00000007884 and ENSBTAG00000014269. The length of primer sequences was determined using the Pimer3, BLAST primer websites (Ye *et al.,* 2012), Multiple Primary Analyzer, and Primary Stats (Hung & Weng, 2016). The primer sequences included forward and reverse as follows F: 5'–TTACCTGAAAACCCCTCACC–3', R: 5'–GTTGCCATCCACGAAGAAAC–3' for SREBF1 gene and F: 5'–TCTTTGAGCCAGCATCTTCT–3', R: 5'–CCAGGTTCTTGTCGGTATCT–3' for *SCARB1* gene, producing PCR products of 783 bp for both genes. The *SREBF1* and *SCARB1* genes were amplified using a thermocycler AB System machine, following the PCR conditions, encompassing 1-minute pre-denaturation at 95 °C, 35 cycles of denaturation at 95 °C for 15 seconds, annealing at 57 °C (*SCARB1*) and 56°C (*SREBF*1) for 15 seconds, extension at 72 °C for 10 seconds, and 3 minutes final extension at 72 °C. In the electrophoresis process, PCR products were separated by 1% agarose gel and examined with a UV Transilluminator (BioradTM, California, USA).

### DNA Sequencing and Genotyping

The DNA sequencing analysis was performed on a 20 µL PCR product placed in a 96-well PCR tray and sealed with a film to be analyzed by the 1st Base Laboratory Services in Selangor, Malaysia. The DNA sequencing was analyzed using Sanger techniques following the methods of Crossley *et al.* (2020). The SNPs identification and genotype determination from the DNA sequencing results were analyzed using FinchTV (Draper, 2008) and Molecular Evolutionary Genetic Analysis (MEGA10) software (Long *et al.,* 2018). The SNPs positions were named starting from the first base of the complete gene sequence.

### Data Analysis

The allele frequency, genotype frequencies, observed and expected heterozygosity, and Hardy-Weinberg equilibrium were calculated using the methods of Webb *et al.* (2021) with PopGen 1.32 software. Allele and genotype frequency values were calculated with the formula of Nei & Kumar (2000) expressed below:

$$x_i = \frac{(2n_{ii} + \sum_{i \neq j} n_{ij})}{2N} \qquad x_i = \frac{n_{ii}}{\mathrm{N}}$$

Where *xi* is allele frequency, *xii* is the genotype frequency, it is the number of individuals with genotypes *ii*, *nij* is the number of individuals with genotypes *ij*, and *N* is the sample number of individuals.

Heterozygosity (Ho) and expected heterozygosity (He) values were calculated with the formula of Nei & Kumar (2000) expressed below:

$$H_0 = \sum_{i \neq j} \frac{N_{1ij}}{N} \qquad H_e = 1 - \sum_{i=1}^{q} x_i^2$$

Where *Ho* is the heterozygosity observation value, *N1ij* is the number of individuals with heterozygous, *N* is the observed number of individuals, *He* is the heterozygosity expectations value, *xi* is the frequency of allele, and *q* is the alleles number.

The following formula from Nei & Kumar (2000) was used to calculate Hardy-Weinberg equilibrium by chi-Square ($\chi 2$) as follows:

$$x^2 = \sum \frac{(O - E)^2}{E}$$

Where $x^2$ is the *chi*-square, *O* is the observed value, and *E* is the expected value.

The association of *SREBF1* and *SCARB1* genotypes with ultrasound back fat thickness, longissimus dorsi thickness, marbling score, intramuscular fat content, and fatty acids were analyzed following the methods of Castelloe (2018) using the General Linear Model (GLM) with SAS 9.4 (SAS Institute, USA). The mathematical model for the GLM is as follows:

$$Yij = \mu + Gi + eij$$

Where Yij is the phenotypic observation, μ is the total mean, Gi is the genotype effect, and eij is the random error. Moreover, carcass and meat characteristics, including fatty acid composition, were also corrected to 36 months of age and similar environment maintenance by Salamena & Papilaja (2010) formula, as follows:

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

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

### Novel of Single Nucleotide Polymorphism

The sequence alignments of 95 samples detected 4 SNPs in the *SREBF1* gene and 5 SNPs in the *SCARB1* gene. These genetic variations were distributed across both the coding and non-coding regions of Bali cattle. SNPs in the *SREBF1* and *SCARB1* genes were detected through the analysis of chromatogram images, indicating double peaks, as shown in Figures 2 and 3. The analysis of the *SREBF1* gene sequence encompassed the span from exon 13 to 15 regions. The 4 SNPs were found between the regions of exon 13 until intron 14 only, which included g.12629T>C on exon 13, g.12731T>C on intron 13, g.12881A>G on exon 14, and g.12986C>T on intron 14, as shown in Table 1.

The *SCARB1* gene sequence was covered from intron 7 to intron 8 regions. The 5 SNPs of the *SCARB1* gene found in Bali cattle were g.72219C>T on intron 7, g.72380C>A and g.72400A>G on exon 8, g.72517G>A, and g.72607C>T on intron 8. Among these SNPs, 4 SNPs, namely g.72219C>T, g.72400A>G, g.72517G>A, and g.72607C>T, of the *SCARB1* gene represented transition mutations, while 1 SNPs (g.72380C>A) was a transversion mutation. The SNPs g.72380C>A and g.72400A>G of the *SCARB1* gene were non-synonymous, causing amino acid changes, as shown in Table 1.

### Polymorphism of SREBF1 and SCARB1 Genes

All SNPs in the *SREBF1* and *SCARB1* genes were polymorphic based on genetic diversity analyses. The number of homozygous BB for SNPs g.12629T>C, g.12731T>C, and g.12881A>G was more than 90% in the SREBF1 gene. In contrast, for SNPs g.12986C>T, AA genotypes were higher compared to the BB genotype. In the case of the *SCARB1* gene, the BB genotype had the highest values for most SNPs, except for SNPs g.72400A>G. Table 2 shows the genotype, allele frequencies, heterozygosity, and Hardy-Weinberg equilibrium of the *SREBF1* and *SCARB1* genes. All SNPs identified

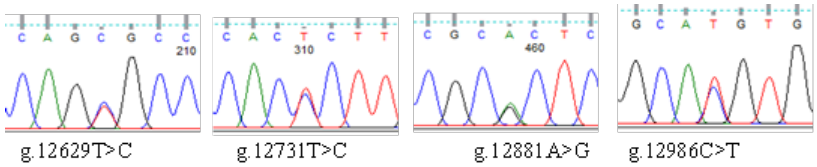



Figure 2. Double band chromatogram of the *SREBF1* genes in Bali cattle

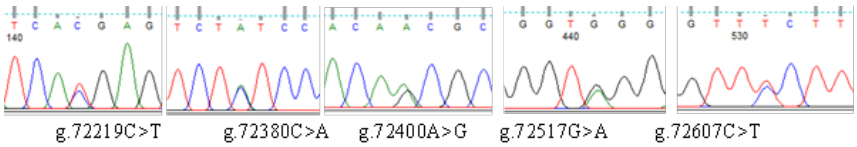



Figure 3. Double band chromatogram of the *SCARB1* genes in Bali cattle

Table 1. SNPs information on the *SREBF1* and *SCARB1* genes in Bali cattle

| Gene | SNPs | Location | Variation type | dbSNP | Amino acids |
|---|---|---|---|---|---|
| *SREBF1* | g.12629T>C | Exon 13 | Transition | Novel | Ser/Ser |
| | g.12731T>C | Intron 13 | Transition | Novel | - |
| | g.12881A>G | Exon 14 | Transition | rs464587519 | Ala/Ala |
| | g.12986C>T | Intron 14 | Transition | rs109146406 | - |
| *SCARB1* | g.72219C>T | Intron 7 | Transition | Novel | - |
| | g.72380C>A | Exon 8 | Transverse | Novel | Ser/Tyr |
| | g.72400A>G | Exon 8 | Transition | Novel | Thr/Ala |
| | g.72517G>A | Intron 8 | Transition | Novel | - |
| | g.72607C>T | Intron 8 | Transition | Novel | - |

Note: Ala= Alanine; Ser= Serine; Tyr= Tyrosine; Thr= Threonine.

in the *SREBF1* gene and 1 specific SNPs (g.72400A>G) in the *SCARB1* gene exhibited Ho values lower than their respective He values, indicating a deviation from the Hardy-Weinberg equilibrium. However, 4 SNPs in the *SCARB1* gene (g.72219C>T, g.72380C>A, g.72517G>A, and g.72607C>T) were found to be in Hardy-Weinberg equilibrium.

### Association Analysis

This study indicated that all SNPs within the *SREBF1* gene showed no significant association (p>0.05) with carcass and meat characteristics of Bali cattle. However, SNPs in the *SCARB1* gene were significantly associated (p<0.05) with carcass characteristics, as presented in Table 3. Significant associations (p<0.05) were found among the 4 SNPs in the *SCARB1* gene (g.72219C>T, g.72380C>A, g.72517G>A, and g.72607C>T) and *longissimus dorsi* thickness (LDT).

In the association analyses, the polymorphism within the *SRBEF1* and *SCARB1* genes significantly affected the fatty acid composition in Bali cattle. Based on statistical analysis, 4 SNPs (g.12629T>C, g.12731T>C, g.12881A>G, and g.12986C>T) of the *SREBF1* gene showed significant associations (P<0.05) with heptadecanoic acid (C17:0) and cis-11-eicosenoic acid (C20:1). As shown in Table 4, SNPs g.12731T>C in the *SREBF1* was directly influenced by the fat content, palmitoleic acid (C16:1), stearic acid (C18:0), cis-11-eicosenoic acid (C20:1), and total fatty acids. All SNPs (g.72219C>T, g.72380C>A, g.72400A>G, g.72517G>A, and g.72607C>T) in the *SCARB1* gene showed significant associations

Table 2. SNPs diversity values of the *SREBF1* and *SCARB1* genes in Bali cattle

| Gene | SNPs | N | Genotypic frequencies | | | Allelic frequencies | | $H_o$ | $H_e$ | Test |
|---|---|---|---|---|---|---|---|---|---|---|
| | | | AA | AB | BB | A | B | | | |
| *SREBF1* | g.12629T>C | 95 | 0.01 | 0.07 | 0.92 | 0.05 | 0.95 | 0.074 | 0.091 | * |
| | g.12731T>C | 95 | 0.01 | 0.06 | 0.93 | 0.04 | 0.96 | 0.063 | 0.081 | * |
| | g.12881A>G | 95 | 0.01 | 0.07 | 0.92 | 0.05 | 0.95 | 0.074 | 0.091 | * |
| | g.12986C>T | 95 | 0.92 | 0.07 | 0.01 | 0.95 | 0.05 | 0.074 | 0.091 | * |
| *SCARB1* | g.72219C>T | 95 | 0.00 | 0.04 | 0.96 | 0.02 | 0.98 | 0.042 | 0.041 | 0.033ns |
| | g.72380C>A | 95 | 0.00 | 0.05 | 0.96 | 0.02 | 0.98 | 0.042 | 0.041 | 0.033ns |
| | g.72400A>G | 95 | 0.92 | 0.06 | 0.02 | 0.95 | 0.05 | 0.063 | 0.100 | * |
| | g.72517G>A | 95 | 0.00 | 0.04 | 0.96 | 0.02 | 0.98 | 0.042 | 0.041 | 0.033ns |
| | g.72607C>T | 95 | 0.00 | 0.04 | 0.96 | 0.02 | 0.98 | 0.042 | 0.041 | 0.033ns |

Note: AA= reference genotype (wildtype); AB= heterozygous genotype; BB= mutant genotype; Ho= observed heterozygosity; He= expected heterozygosity; ns= not significant; * means significantly= chi square values ($\chi^2$ test) > chi-square table (3.84: $\alpha$0.05 db 2).

Table 3. Association of SNPs *SREBF1* and *SCARB1* genes with carcass and meat characteristics in Bali cattle

| Gene | SNPs | Genotype (N) | LDT | BFT | MS | IMF |
|---|---|---|---|---|---|---|
| *SREBF1* | g.12629T>C | CC (83) | 46.99 ± 5.87 | 1.89 ± 0.32 | 1.51 ± 0.56 | 2.57 ± 1.39 |
| | | CT (7) | 49.42 ± 4.68 | 1.83 ± 0.20 | 1.36 ± 0.22 | 2.19 ± 0.55 |
| | | TT (1) | 46.59 ± nc | 1.58 ± nc | 1.87 ± nc | 3.46 ± nc |
| | g.12731T>C | TT (1) | 46.59 ± nc | 1.58 ± nc | 1.87 ± nc | 3.46 ± nc |
| | | TC (6) | 50.61 ± 3.79 | 1.83 ± 0.22 | 1.41 ± 0.19 | 2.32 ± 0.46 |
| | | CC (84) | 46.93 ± 5.86 | 1.89 ± 0.32 | 1.50 ± 0.56 | 2.56 ± 1.39 |
| | g.12881A>G | AA (1) | 46.59 ± nc | 1.58 ± nc | 1.87 ± nc | 3.46 ± nc |
| | | AG (7) | 49.42 ± 4.68 | 1.83 ± 0.20 | 1.36 ± 0.22 | 2.19 ± 0.55 |
| | | GG (83) | 46.99 ± 5.87 | 1.89 ± 0.32 | 1.51 ± 0.56 | 2.57 ± 1.39 |
| | g.12986C>T | CC (83) | 46.99 ± 5.87 | 1.89 ± 0.32 | 1.51 ± 0.56 | 2.57 ± 1.39 |
| | | CT (7) | 49.42 ± 4.68 | 1.83 ± 0.20 | 1.36 ± 0.22 | 2.19 ± 0.55 |
| | | TT (1) | 46.59 ± nc | 1.58 ± nc | 1.87 ± nc | 3.46 ± nc |
| *SCARB1* | g.72219C>T | CT (4) | 53.37 ± 8.64[a] | 1.80 ± 0.29 | 1.66 ± 0.55 | 2.95 ± 1.37 |
| | | TT (87) | 46.89 ± 5.51[b] | 1.88 ± 0.31 | 1.49 ± 0.54 | 2.53 ± 1.35 |
| | g.72380C>A | CA (4) | 53.37 ± 8.64[a] | 1.80 ± 0.29 | 1.66 ± 0.55 | 2.95 ± 1.37 |
| | | AA (87) | 46.89 ± 5.51[b] | 1.88 ± 0.31 | 1.49 ± 0.54 | 2.53 ± 1.35 |
| | g.72400A>G | AA (84) | 47.09 ± 5.49 | 1.87 ± 0.31 | 1.49 ± 0.55 | 2.52 ± 1.36 |
| | | AG (6) | 49.60 ± 8.98 | 1.93 ± 0.30 | 1.67 ± 0.46 | 2.98 ± 1.15 |
| | | GG (1) | 39.64 ± nc | 2.11 ± nc | 1.47 ± 0.00 | 2.47 ± nc |
| | g.72517G>A | GA (4) | 53.37 ± 8.64[a] | 1.80 ± 0.29 | 1.66 ± 0.55 | 2.95 ± 1.37 |
| | | AA (87) | 46.89 ± 5.51[b] | 1.88 ± 0.31 | 1.49 ± 0.54 | 2.53 ± 1.35 |
| | g.72607C>T | CT (4) | 53.37 ± 8.64[a] | 1.80 ± 0.29 | 1.66 ± 0.55 | 2.95 ± 1.37 |
| | | TT (87) | 46.89 ± 5.51[b] | 1.88 ± 0.31 | 1.49 ± 0.54 | 2.53 ± 1.35 |

Note: N= number of samples; LDT= longissimus dorsi thickness; BFT= back fat thickness; MS= marbling score; IMF= intramuscular fat. Means in the same column with different superscripts differ significantly (p<0.05); nc= not counted.

Table 4. Association of SNPs *SREBF1* genes with fatty acids composition in Bali cattle

| Fatty acid composition | g.12629T>C | | | g.12731T>C | | | g.12881A>G | | | g.12986C>T | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | CC (40) | CT (3) | TT (1) | TT (1) | TC (2) | CC (41) | AA (1) | AG (3) | GG (40) | CC (40) | CT (3) | TT (1) |
| Fat content | 3.14 ± 1.16 | 2.99 ± 1.47 | 5.76 ± nc | 5.76 ± nc | 3.81 ± 0.44$^{ab}$ | **3.10 ± 1.18$^{b}$** | 5.76 ± nc | 2.99 ± 1.47 | 3.14 ± 1.16 | 3.14 ± 1.16 | 2.99 ± 1.47 | 5.76 ± nc |
| C8:0 | 0.06 ± 0.20 | 0.01 ± 0.02 | 0.00 ± nc | 0.00 ± nc | 0.02 ± 0.03 | 0.06 ± 0.20 | nc ± nc | 0.01 ± 0.02 | 0.06 ± 0.20 | 0.06 ± 0.20 | 0.01 ± 0.02 | 0.00 ± nc |
| C12:0 | 0.07 ± 0.03 | 0.06 ± 0.03 | 0.05 ± nc | 0.05 ± nc | 0.07 ± 0.01 | 0.07 ± 0.03 | 0.05 ± nc | 0.06 ± 0.03 | 0.07 ± 0.03 | 0.07 ± 0.03 | 0.06 ± 0.03 | 0.05 ± nc |
| C13:0 | 0.03 ± 0.02 | 0.02 ± 0.01 | 0.00 ± nc | 0.00 ± nc | 0.02 ± 0.01 | 0.03 ± 0.02 | 0.00 ± nc | 0.02 ± 0.01 | 0.03 ± 0.02 | 0.03 ± 0.02 | 0.02 ± 0.01 | 0.00 ± nc |
| C14:0 | 2.18 ± 0.58 | 2.18 ± 0.18 | 2.67 ± nc | 2.67 ± nc | 2.29 ± 0.04 | 2.18 ± 0.57 | 2.67 ± nc | 2.18 ± 0.18 | 2.18 ± 0.58 | 2.18 ± 0.58 | 2.18 ± 0.18 | 2.67 ± nc |
| C14:1 | 0.30 ± 0.38 | 0.04 ± 0.01 | 0.07 ± nc | 0.07 ± nc | 0.04 ± 0.02 | 0.30 ± 0.37 | 0.07 ± nc | 0.04 ± 0.01 | 0.30 ± 0.38 | 0.30 ± 0.38 | 0.04 ± 0.01 | 0.07 ± nc |
| C15:0 | 0.60 ± 0.37 | 0.59 ± 0.17 | 0.53 ± nc | 0.53 ± nc | 0.62 ± 0.23 | 0.60 ± 0.36 | 0.53 ± nc | 0.59 ± 0.17 | 0.60 ± 0.37 | 0.60 ± 0.37 | 0.59 ± 0.17 | 0.53 ± nc |
| C16:0 | 21.04 ± 4.12 | 21.66 ± 3.57 | 21.44 ± nc | 21.44 ± nc | 23.64 ± 1.37 | 20.96 ± 4.11 | 21.44 ± nc | 21.66 ± 3.57 | 21.04 ± 4.12 | 21.04 ± 4.12 | 21.66 ± 3.57 | 21.44 ± nc |
| C16:1 | 1.29 ± 0.28 | 1.68 ± 1.03 | 1.20 ± nc | 1.20 ± nc | 2.09 ± 1.06$^{a}$ | **1.28 ± 0.28$^{b}$** | 1.20 ± nc | 1.68 ± 1.03 | 1.29 ± 0.28 | 1.29 ± 0.28 | 1.68 ± 1.03 | 1.20 ± nc |
| C17:0 | 1.94 ± 0.44$^{ab}$ | 3.00 ± 2.62$^{a}$ | 1.35 ± nc | 1.35 ± nc | 3.91 ± 2.96$^{a}$ | 1.92 ± 0.45$^{b}$ | 1.35 ± nc | 3.00 ± 2.62$^{a}$ | 1.94 ± 0.44$^{ab}$ | 1.94 ± 0.44$^{ab}$ | 3.00 ± 2.62$^{a}$ | 1.35 ± nc |
| C17:1 | 0.26 ± 0.14 | 0.37 ± 0.28 | 0.24 ± nc | 0.24 ± nc | 0.47 ± 0.30 | 0.26 ± 0.14 | 0.24 ± nc | 0.37 ± 0.28 | 0.26 ± 0.14 | 0.26 ± 0.14 | 0.37 ± 0.28 | 0.24 ± nc |
| C18:0 | 32.25 ± 3.46 | 31.10 ± 5.60 | 25.33 ± nc | 25.33 ± nc | 34.23 ± 1.98$^{a}$ | **32.07 ± 3.61$^{ab}$** | 25.33 ± nc | 31.10 ± 5.60 | 32.25 ± 3.46 | 32.25 ± 3.46 | 31.10 ± 5.60 | 25.33 ± nc |
| C18:1n9t | 3.00 ± 1.27 | 3.42 ± 1.20 | 1.71 ± nc | 1.71 ± nc | 4.11 ± 0.15 | 2.98 ± 1.26 | 1.71 ± nc | 3.42 ± 1.20 | 3.00 ± 1.27 | 3.00 ± 1.27 | 3.42 ± 1.20 | 1.71 ± nc |
| C18:1n9c | 11.98 ± 4.99 | 13.48 ± 2.61 | 15.55 ± nc | 15.55 ± nc | 14.97 ± 0.44 | 11.95 ± 4.93 | 15.55 ± nc | 13.48 ± 2.61 | 11.98 ± 4.99 | 11.98 ± 4.99 | 13.48 ± 2.61 | 15.55 ± nc |
| C18:2n6c | 1.86 ± 0.83 | 1.34 ± 0.27 | 0.82 ± nc | 0.82 ± nc | 1.19 ± 0.15 | 1.85 ± 0.82 | 0.82 ± nc | 1.34 ± 0.27 | 1.86 ± 0.83 | 1.86 ± 0.83 | 1.34 ± 0.27 | 0.82 ± nc |
| C18:2n9t | 0.12 ± 0.21 | 0.02 ± 0.02 | 0.10 ± nc | 0.10 ± nc | 0.03 ± 0.02 | 0.11 ± 0.21 | 0.10 ± nc | 0.02 ± 0.02 | 0.12 ± 0.21 | 0.12 ± 0.21 | 0.02 ± 0.02 | 0.10 ± nc |
| C18:3n3 | 0.24 ± 0.27 | 0.00 ± 0.00 | 0.42 ± nc | 0.42 ± nc | 0.00 ± 0.00 | 0.23 ± 0.27 | 0.42 ± nc | 0.00 ± 0.00 | 0.24 ± 0.27 | 0.24 ± 0.27 | 0.00 ± 0.00 | 0.42 ± nc |
| C18:3n6 | 0.01 ± 0.02 | 0.01 ± 0.01 | 0.00 ± nc | 0.00 ± nc | 0.01 ± 0.01 | 0.01 ± 0.02 | 0.00 ± nc | 0.01 ± 0.01 | 0.01 ± 0.02 | 0.01 ± 0.02 | 0.01 ± 0.01 | 0.00 ± nc |
| C20:0 | 0.44 ± 0.20 | 0.28 ± 0.07 | 0.26 ± nc | 0.26 ± nc | 0.31 ± 0.07 | 0.43 ± 0.20 | 0.26 ± nc | 0.28 ± 0.07 | 0.44 ± 0.20 | 0.44 ± 0.20 | 0.28 ± 0.07 | 0.26 ± nc |
| C20:1 | 0.30 ± 0.27 | 0.55 ± 0.12 | 0.00 ± nc | 0.00 ± nc | 0.57 ± 0.16$^{a}$ | 0.31 ± 0.27$^{ab}$ | 0.00 ± nc | 0.55 ± 0.12$^{a}$ | 0.30 ± 0.27$^{ab}$ | 0.30 ± 0.27$^{ab}$ | 0.55 ± 0.12$^{a}$ | 0.00 ± nc |
| C20:2 | 0.10 ± 0.16 | 0.04 ± 0.04 | 0.09 ± nc | 0.09 ± nc | 0.04 ± 0.05 | 0.10 ± 0.16 | 0.09 ± nc | 0.04 ± 0.04 | 0.10 ± 0.16 | 0.10 ± 0.16 | 0.04 ± 0.04 | 0.09 ± nc |
| C20:3n3 | 0.04 ± 0.15 | 0.03 ± 0.05 | 0.00 ± nc | 0.00 ± nc | 0.05 ± 0.06 | 0.04 ± 0.15 | 0.00 ± nc | 0.03 ± 0.05 | 0.04 ± 0.15 | 0.04 ± 0.15 | 0.03 ± 0.05 | 0.00 ± nc |
| C20:3n6 | 0.11 ± 0.10 | 0.10 ± 0.10 | 0.00 ± nc | 0.00 ± nc | 0.06 ± 0.08 | 0.11 ± 0.10 | 0.00 ± nc | 0.10 ± 0.10 | 0.11 ± 0.10 | 0.11 ± 0.10 | 0.10 ± 0.10 | 0.00 ± nc |
| C20:4n6 | 0.19 ± 0.14 | 0.02 ± 0.03 | 0.04 ± nc | 0.04 ± nc | 0.03 ± 0.04 | 0.19 ± 0.14 | 0.04 ± nc | 0.02 ± 0.03 | 0.19 ± 0.14 | 0.19 ± 0.14 | 0.02 ± 0.03 | 0.04 ± nc |
| C20:5n3 | 0.16 ± 0.17 | 0.11 ± 0.11 | 0.00 ± nc | 0.00 ± nc | 0.07 ± 0.09 | 0.16 ± 0.17 | 0.00 ± nc | 0.11 ± 0.11 | 0.16 ± 0.17 | 0.16 ± 0.17 | 0.11 ± 0.11 | 0.00 ± nc |
| C21:0 | 0.11 ± 0.05 | 0.05 ± 0.05 | 0.08 ± nc | 0.08 ± nc | 0.05 ± 0.07 | 0.10 ± 0.05 | 0.08 ± nc | 0.05 ± 0.05 | 0.11 ± 0.05 | 0.11 ± 0.05 | 0.05 ± 0.05 | 0.08 ± nc |
| C22:0 | 0.19 ± 0.12 | 0.12 ± 0.03 | 0.06 ± nc | 0.06 ± nc | 0.11 ± 0.04 | 0.19 ± 0.11 | 0.06 ± nc | 0.12 ± 0.03 | 0.19 ± 0.12 | 0.19 ± 0.12 | 0.12 ± 0.03 | 0.06 ± nc |
| C23:0 | 0.10 ± 0.06 | 0.03 ± 0.05 | 0.00 ± nc | 0.00 ± nc | 0.00 ± 0.00 | 0.10 ± 0.06 | 0.00 ± nc | 0.03 ± 0.05 | 0.10 ± 0.06 | 0.10 ± 0.06 | 0.03 ± 0.05 | 0.00 ± nc |
| C24:0 | 0.04 ± 0.04 | 0.04 ± 0.02 | 0.02 ± nc | 0.02 ± nc | 0.04 ± 0.03 | 0.04 ± 0.04 | 0.02 ± nc | 0.04 ± 0.02 | 0.04 ± 0.04 | 0.04 ± 0.04 | 0.04 ± 0.02 | 0.02 ± nc |
| C24:1 | 0.02 ± 0.02 | 0.00 ± 0.00 | 0.00 ± nc | 0.00 ± nc | 0.00 ± 0.00 | 0.02 ± 0.02 | 0.00 ± nc | 0.00 ± 0.00 | 0.02 ± 0.02 | 0.02 ± 0.02 | 0.00 ± 0.00 | 0.00 ± nc |
| SFA | 59.14 ± 6.25 | 59.23 ± 11.55 | 51.79 ± nc | 51.79 ± nc | 65.42 ± 6.05 | 58.84 ± 6.47 | 51.79 ± nc | 59.23 ± 11.55 | 59.14 ± 6.25 | 59.14 ± 6.25 | 59.23 ± 11.55 | 51.79 ± nc |
| MUFA | 17.17 ± 5.15 | 19.54 ± 4.82 | 18.77 ± nc | 18.77 ± nc | 22.25 ± 1.48 | 17.09 ± 5.11 | 18.77 ± nc | 19.54 ± 4.82 | 17.17 ± 5.15 | 17.17 ± 5.15 | 19.54 ± 4.82 | 18.77 ± nc |
| PUFA | 2.83 ± 1.14 | 1.66 ± 0.47 | 1.47 ± nc | 1.47 ± nc | 1.46 ± 0.43 | 2.81 ± 1.13 | 1.47 ± nc | 1.66 ± 0.47 | 2.83 ± 1.14 | 2.83 ± 1.14 | 1.66 ± 0.47 | 1.47 ± nc |
| UFA | 20.00 ± 4.64 | 21.20 ± 4.41 | 20.23 ± nc | 20.23 ± nc | 23.71 ± 1.05 | 19.91 ± 4.62 | 20.23 ± nc | 21.20 ± 4.41 | 20.00 ± 4.64 | 20.00 ± 4.64 | 21.20 ± 4.41 | 20.23 ± nc |
| T. Fatty Acid | 78.99 ± 6.49 | 79.71 ± 15.00 | 71.24 ± nc | 71.24 ± nc | 88.06 ± 5.59$^{a}$ | **78.60 ± 6.87$^{ab}$** | 71.24 ± nc | 79.71 ± 15.00 | 78.99 ± 6.49 | 78.99 ± 6.49 | 79.71 ± 15.00 | 71.24 ± nc |

Note: SFA= Saturated fatty acid; MUFA= Monounsaturated fatty acid; UFA= Unsaturated fatty acid; T. fatty acid= total fatty acid; Means in the same row with different superscripts differ significantly ($p<0.05$); nc= Not counted.

($p<0.05$) with cis-10 heptadecenoic acid (C17:1) and cis-8,11,14-eicosatrienoic acid (C20:3n6). The results of the association analysis between polymorphisms in the *SCARB1* gene and the fatty acid composition of Bali cattle are presented in Table 5. Moreover, the association analysis showed that SNP g.72400A>G in the *SCARB1* gene was significantly associated ($p<0.05$) with caprylic acid (C8:0), lauric acid (C12:0), arachidonic acid (C20:4n6), total monounsaturated fatty acids (MUFA), and total unsaturated fatty acids (UFA).

## DISCUSSION

The molecular method is a technology used in genetic studies to obtain information about genetic diversity at the DNA level. By utilizing this technique, significant information can be obtained on genetic polymorphism and mutations in the population. Ellegren & Galtier (2016) stated that genetic polymorphism varies between species and within genomes, significantly affecting species' evolution and conservation. Molecular technology is an important key in uncovering and analyzing genetic diversity at a more detailed and precise level. In the present study, the DNA Sequencing technique was applied, which is a powerful tool in modern genetics, fast, easy, and clear in disclosing complex and important genetic information (Heather & Chain, 2016).

Sterol regulatory element binding proteins (*SREBF1*) are transcription factors that are fundamentally helix-loop-helix-leucine zippers. This transcription factor is an essential regulator of fatty acid biosynthesis and cholesterol homeostasis by binding to the DNA sequence TCACNCCAC for the sterol regulatory element (Felder *et al.,* 2005). In this study, the *SREBF1* gene was amplified from blood DNA using a primer pair from exon 13 to exon 15. The result of DNA sequencing in Bali cattle revealed that 4 SNPs (g.12629T>C, g.12731T>C, g.12881A>G, and g.12986C>T) were identified in the exon 13 and intron 14 regions of the *SREBF1* gene. The SNPs in the *SREBF1* gene were synonymous mutations. These synonymous mutations did not result in any changes, while non-synonymous mutations were genetic variations that altered amino acid components (Berg *et al.,* 2015). The genetic diversity within a population can be measured by predicting the heterozygosity value. In this study, all SNPs of the *SREBF1* gene exhibited a lower observed heterozygosity (Ho) value compared to expected heterozygosity (He). This indicated a deviation from the proportions of heterozygous genotypes as predicted by the Hardy-Weinberg equilibrium (Hartl & Clark, 2007). This test instrument assessed whether the proportions of genotypes within a population remained consistent across generations (Waples, 2015) and was evaluated based on the $X^2$ value.

The influences of SNPs in the *SREBF1* gene on carcass and meat characteristics were analyzed in 91 live Bali cattle. Based on statistical analysis, all SNPs in the *SREBF1* gene showed no significant differences ($p>0.05$) among the genotypes investigated. The *SREBF* is a transcription factor gene that affects marbling (Lee *et al.,* 2013; Siachos *et al.,* 2021). According to Lee *et al.* (2013), SNPs in the *SREBF1* gene were significantly associated with marbling scores in the exon 9 region. The 84 bp indels in intron 5 illustrated three different genotypes significantly associated with stearic acid (C18:0) (Gamarra *et al.,* 2021). According to Bhuiyan *et al.* (2009), Korean Hanwoo bulls with the LL genotype have more stearic acids (18:0) in their muscle fats than those with the LS and SS genotypes. Stachowiak *et al.* (2013) reported two unique SNPs in the SREBF1 gene were promising markers for pig carcass and performance attributes, but not effective markers for fatty acid content. In the present study, 4 SNPs (g.12629T>C, g.12731T>C, g.12881A>G, and g.12986C>T) in the *SREBF1* gene were significantly associated ($p<0.05$) with stearic acid (C18:0). The TC genotype at SNPs g.12731T>C exhibited the highest average stearic acid (C18:0) of 34.23%. Shramko *et al.* (2020) identified stearic acid as one of the saturated fatty acids that did not significantly increase low-density lipoprotein (LDL) and the risk of cardiovascular disease (CVD). Based on association analysis, 1 SNP in the *SREBF1* gene (g.12731T>C) was associated ($p<0.05$) with fatty acid composition, including caprylic (C8:0), heptadecanoic (C17:0), palmitoleic (C16:1), and eicosanoic acids (C20:1). Palmitoleic acid (C16:1) had been identified as a potential therapeutic agent for metabolic syndrome, insulin resistance, and diabetes (Cruz *et al.,* 2020; Bergman *et al.,* 2013), while eicosanoic acid (C20:1) exhibited an anti-inflammatory or inflammation regulator in the blood.

In the current study, 5 SNPs in the *SCARB1* gene (g.72219C>T, g.72380C>A, g.72400A>G, g.72517G>A, and g.72607C>T) were discovered in the intron 7 and intron 8 regions. The 2 novel SNPs (g.72380C>A and g.72400A>G) in the *SCARB1* gene were non-synonymous mutations. These non-synonymous mutations were genetic variations that altered amino acid components. The SNPs g.72380C>A resulted in an amino acid change from serine to tyrosine, while the SNPs g.72400A>G caused a change from threonine to alanine, potentially affecting the structure and function. This indicated that the SNPs influenced genes' enzymatic activity and structural stability (Ng & Henikoff, 2006). Based on the results, 4 SNPs of the *SCARB1* gene (g.72219C>T, g.72380C>A, g.72517G>A, and g.72607C>T) were found in Hardy-Weinberg equilibrium. The Bali cattle used for the analysis were obtained from breeders with an uncontrolled mating system, which increased the likelihood of inbreeding and deviated from the Hardy-Weinberg equilibrium (Garnier-Gere *et al.,* 2013; Graffelman *et al.,* 2017).

Based on statistical analysis, the 4 novel SNPs in the *SCARB1* gene were significantly associated ($p>0.05$) with backfat thickness (BFT) and *longissimus dorsi* muscle thickness (LDT). Previous studies indicated that BFT and LDT measurements using ultrasonography could be used to estimate Body Condition Scoring (Hussein *et al.,* 2013). Based on the study conducted by Siachos *et al.* (2021), BFT was found to positively correlate with LDT ($r=0.69$), indicating that a rise in BFT increased LDT values. This study showed a significant association of the *SCARB1* genes with fatty acid composition in Bali cattle. The results indicated that the SNP g.72400A>G in the *SCARB1* gene was associated with saturated

Table 5. Association of SNP *SCARB1* genes with fatty acids composition in Bali cattle

| Fatty acid composition | g.72219C>T | | g.72380C>A | | g.72400A>G | | | g.72517G>A | | g.72607C>T | |
|---|---|---|---|---|---|---|---|---|---|---|---|
| | CT (2) | TT (42) | CA (2) | AA (42) | AA (39) | AG (3) | GG (2) | GA (2) | AA (42) | CT (2) | TT (42) |
| Fat content | 2.67 ± 0.73 | 3.21 ± 1.23 | 2.67 ± 0.73 | 3.21 ± 1.23 | 3.27 ± 1.23 | 2.97 ± 0.73 | 1.94 ± 1.21 | 2.67 ± 0.73 | 3.21 ± 1.23 | 2.67 ± 0.73 | 3.21 ± 1.23 |
| C8:0 | 0.00 ± 0.00 | 0.06 ± 0.20 | 0.00 ± 0.00 | 0.06 ± 0.20 | 0.04 ± 0.17$^{b}$ | 0.00 ± 0.00$^{b}$ | **0.40 ± 0.56$^{a}$** | 0.00 ± 0.00 | 0.06 ± 0.20 | 0.00 ± 0.00 | 0.06 ± 0.20 |
| C12:0 | 0.07 ± 0.02 | 0.07 ± 0.03 | 0.07 ± 0.02 | 0.07 ± 0.03 | 0.07 ± 0.02$^{b}$ | 0.11 ± 0.07$^{a}$ | **0.13 ± 0.08$^{a}$** | 0.07 ± 0.02 | 0.07 ± 0.03 | 0.07 ± 0.02 | 0.07 ± 0.03 |
| C13:0 | 0.02 ± 0.03 | 0.03 ± 0.02 | 0.02 ± 0.03 | 0.03 ± 0.02 | 0.03 ± 0.02 | 0.03 ± 0.02 | 0.06 ± 0.00 | 0.02 ± 0.03 | 0.03 ± 0.02 | 0.02 ± 0.03 | 0.03 ± 0.02 |
| C14:0 | 2.30 ± 1.54 | 2.19 ± 0.52 | 2.30 ± 1.54 | 2.19 ± 0.52 | 2.13 ± 0.46 | 2.65 ± 1.25 | 2.77 ± 0.94 | 2.30 ± 1.54 | 2.19 ± 0.52 | 2.30 ± 1.54 | 2.19 ± 0.52 |
| C14:1 | 0.33 ± 0.36 | 0.28 ± 0.37 | 0.33 ± 0.36 | 0.28 ± 0.37 | 0.29 ± 0.38 | 0.25 ± 0.29 | 0.06 ± 0.01 | 0.33 ± 0.36 | 0.28 ± 0.37 | 0.33 ± 0.36 | 0.28 ± 0.37 |
| C15:0 | 0.55 ± 0.05 | 0.60 ± 0.36 | 0.55 ± 0.05 | 0.60 ± 0.36 | 0.60 ± 0.37 | 0.63 ± 0.16 | 0.51 ± 0.12 | 0.55 ± 0.05 | 0.60 ± 0.36 | 0.55 ± 0.05 | 0.60 ± 0.36 |
| C16:0 | 18.75 ± 4.09 | 21.20 ± 4.02 | 18.75 ± 4.09 | 21.20 ± 4.02 | 20.72 ± 3.24 | 23.49 ± 8.69 | 24.86 ± 9.45 | 18.75 ± 4.09 | 21.20 ± 4.02 | 18.75 ± 4.09 | 21.20 ± 4.02 |
| C16:1 | 1.25 ± 0.44 | 1.32 ± 0.36 | 1.25 ± 0.44 | 1.32 ± 0.36 | 1.31 ± 0.36 | 1.50 ± 0.53 | 1.29 ± 0.27 | 1.25 ± 0.44 | 1.32 ± 0.36 | 1.25 ± 0.44 | 1.32 ± 0.36 |
| C17:0 | 1.94 ± 0.53 | 2.00 ± 0.77 | 1.94 ± 0.53 | 2.00 ± 0.77 | 1.99 ± 0.79 | 2.01 ± 0.39 | 2.00 ± 0.81 | 1.94 ± 0.53 | 2.00 ± 0.77 | 1.94 ± 0.53 | 2.00 ± 0.77 |
| C17:1 | 0.53 ± 0.29$^{a}$ | 0.26 ± 0.13$^{b}$ | 0.53 ± 0.29$^{a}$ | 0.26 ± 0.13$^{b}$ | 0.25 ± 0.13$^{b}$ | 0.50 ± 0.21$^{a}$ | 0.27 ± 0.04$^{b}$ | 0.53 ± 0.29$^{a}$ | 0.26 ± 0.13$^{b}$ | 0.53 ± 0.29$^{a}$ | 0.26 ± 0.13$^{b}$ |
| C18:0 | 32.69 ± 3.83 | 31.98 ± 3.71 | 32.69 ± 3.83 | 31.98 ± 3.71 | 31.89 ± 3.67 | 33.98 ± 3.50 | 31.42 ± 5.42 | 32.69 ± 3.83 | 31.98 ± 3.71 | 32.69 ± 3.83 | 31.98 ± 3.71 |
| C18:1n9t | 3.90 ± 0.58 | 2.96 ± 1.27 | 3.90 ± 0.58 | 2.96 ± 1.27 | 3.04 ± 1.27 | 3.17 ± 1.32 | 1.94 ± 0.56 | 3.90 ± 0.58 | 2.96 ± 1.27 | 3.90 ± 0.58 | 2.96 ± 1.27 |
| C18:1n9c | 14.26 ± 6.89 | 12.07 ± 4.80 | 14.26 ± 6.89 | 12.07 ± 4.80 | 12.70 ± 4.12 | 9.54 ± 9.52 | 5.70 ± 7.89 | 14.26 ± 6.89 | 12.07 ± 4.80 | 14.26 ± 6.89 | 12.07 ± 4.80 |
| C18:2n6c | 1.66 ± 0.58 | 1.80 ± 0.83 | 1.66 ± 0.58 | 1.80 ± 0.83 | 1.72 ± 0.76 | 2.40 ± 1.34 | 2.33 ± 1.14 | 1.66 ± 0.58 | 1.80 ± 0.83 | 1.66 ± 0.58 | 1.80 ± 0.83 |
| C18:2n9t | 0.28 ± 0.38 | 0.10 ± 0.19 | 0.28 ± 0.38 | 0.10 ± 0.19 | 0.11 ± 0.20 | 0.20 ± 0.30 | 0.01 ± 0.01 | 0.28 ± 0.38 | 0.10 ± 0.19 | 0.28 ± 0.38 | 0.10 ± 0.19 |
| C18:3n3 | 0.01 ± 0.02 | 0.23 ± 0.27 | 0.01 ± 0.02 | 0.23 ± 0.27 | 0.25 ± 0.27 | 0.01 ± 0.01 | 0.12 ± 0.17 | 0.01 ± 0.02 | 0.23 ± 0.27 | 0.01 ± 0.02 | 0.23 ± 0.27 |
| C18:3n6 | 0.00 ± 0.00 | 0.01 ± 0.02 | 0.00 ± 0.00 | 0.01 ± 0.02 | 0.01 ± 0.02 | 0.00 ± 0.00 | 0.00 ± 0.00 | 0.00 ± 0.00 | 0.01 ± 0.02 | 0.00 ± 0.00 | 0.01 ± 0.02 |
| C20:0 | 0.44 ± 0.11 | 0.42 ± 0.20 | 0.44 ± 0.11 | 0.42 ± 0.20 | 0.42 ± 0.20 | 0.43 ± 0.08 | 0.44 ± 0.08 | 0.44 ± 0.11 | 0.42 ± 0.20 | 0.44 ± 0.11 | 0.42 ± 0.20 |
| C20:1 | 0.28 ± 0.10 | 0.32 ± 0.27 | 0.28 ± 0.10 | 0.32 ± 0.27 | 0.30 ± 0.26 | 0.45 ± 0.30 | 0.37 ± 0.48 | 0.28 ± 0.10 | 0.32 ± 0.27 | 0.28 ± 0.10 | 0.32 ± 0.27 |
| C20:2 | 0.12 ± 0.02 | 0.10 ± 0.16 | 0.12 ± 0.02 | 0.10 ± 0.16 | 0.10 ± 0.16 | 0.11 ± 0.02 | 0.11 ± 0.07 | 0.12 ± 0.02 | 0.10 ± 0.16 | 0.12 ± 0.02 | 0.10 ± 0.16 |
| C20:3n3 | 0.06 ± 0.01 | 0.11 ± 0.10 | 0.06 ± 0.01 | 0.11 ± 0.10 | 0.11 ± 0.10 | 0.13 ± 0.11 | 0.06 ± 0.08 | 0.06 ± 0.01 | 0.11 ± 0.10 | 0.06 ± 0.01 | 0.11 ± 0.10 |
| C20:3n6 | 0.33 ± 0.46$^{a}$ | 0.03 ± 0.11$^{b}$ | 0.33 ± 0.46$^{a}$ | 0.03 ± 0.11$^{b}$ | 0.02 ± 0.08$^{b}$ | 0.40 ± 0.35$^{a}$ | 0.00 ± 0.00$^{b}$ | 0.33 ± 0.46$^{a}$ | 0.03 ± 0.11$^{b}$ | 0.33 ± 0.46$^{a}$ | 0.03 ± 0.11$^{b}$ |
| C20:4n6 | 0.05 ± 0.02 | 0.18 ± 0.14 | 0.05 ± 0.02 | 0.18 ± 0.14 | 0.18 ± 0.14$^{ab}$ | 0.04 ± 0.03$^{b}$ | **0.35 ± 0.06$^{a}$** | 0.05 ± 0.02 | 0.18 ± 0.14 | 0.05 ± 0.02 | 0.18 ± 0.14 |
| C20:5n3 | 0.00 ± 0.00 | 0.16 ± 0.17 | 0.00 ± 0.00 | 0.16 ± 0.17 | 0.15 ± 0.16 | 0.15 ± 0.25 | 0.28 ± 0.28 | 0.00 ± 0.00 | 0.16 ± 0.17 | 0.00 ± 0.00 | 0.16 ± 0.17 |
| C21:0 | 0.12 ± 0.02 | 0.10 ± 0.06 | 0.12 ± 0.02 | 0.10 ± 0.06 | 0.10 ± 0.05 | 0.11 ± 0.02 | 0.07 ± 0.10 | 0.12 ± 0.02 | 0.10 ± 0.06 | 0.12 ± 0.02 | 0.10 ± 0.06 |
| C22:0 | 0.18 ± 0.07 | 0.18 ± 0.12 | 0.18 ± 0.07 | 0.18 ± 0.12 | 0.18 ± 0.12 | 0.20 ± 0.06 | 0.24 ± 0.03 | 0.18 ± 0.07 | 0.18 ± 0.12 | 0.18 ± 0.07 | 0.18 ± 0.12 |
| C23:0 | 0.10 ± 0.00 | 0.09 ± 0.07 | 0.10 ± 0.00 | 0.09 ± 0.07 | 0.09 ± 0.07 | 0.11 ± 0.02 | 0.09 ± 0.12 | 0.10 ± 0.00 | 0.09 ± 0.07 | 0.10 ± 0.00 | 0.09 ± 0.07 |
| C24:0 | 0.02 ± 0.02 | 0.04 ± 0.04 | 0.02 ± 0.02 | 0.04 ± 0.04 | 0.04 ± 0.04 | 0.04 ± 0.03 | 0.04 ± 0.06 | 0.02 ± 0.02 | 0.04 ± 0.04 | 0.02 ± 0.02 | 0.04 ± 0.04 |
| C24:1 | 0.00 ± 0.00 | 0.01 ± 0.02 | 0.00 ± 0.00 | 0.01 ± 0.02 | 0.01 ± 0.02 | 0.00 ± 0.00 | 0.03 ± 0.00 | 0.00 ± 0.00 | 0.01 ± 0.02 | 0.00 ± 0.00 | 0.01 ± 0.02 |
| SFA | 57.30 ± 2.30 | 59.06 ± 6.69 | 57.30 ± 2.30 | 59.06 ± 6.69 | 58.39 ± 5.52 | 63.93 ± 11.60 | 63.17 ± 17.15 | 57.30 ± 2.30 | 59.06 ± 6.69 | 57.30 ± 2.30 | 59.06 ± 6.69 |
| MUFA | 20.54 ± 6.21 | 17.21 ± 5.04 | 20.54 ± 6.21 | 17.21 ± 5.04 | 17.91 ± 4.35$^{a}$ | 15.41 ± 9.92$^{ab}$ | **9.65 ± 6.53$^{b}$** | 20.54 ± 6.21 | 17.21 ± 5.04 | 20.54 ± 6.21 | 17.21 ± 5.04 |
| PUFA | 2.52 ± 0.43 | 2.73 ± 1.17 | 2.52 ± 0.43 | 2.73 ± 1.17 | 2.64 ± 1.11 | 3.43 ± 1.61 | 3.24 ± 1.30 | 2.52 ± 0.43 | 2.73 ± 1.17 | 2.52 ± 0.43 | 2.73 ± 1.17 |
| UFA | 23.06 ± 5.77 | 19.95 ± 4.50 | 23.06 ± 5.77 | 19.95 ± 4.50 | 20.55 ± 3.97$^{a}$ | 18.84 ± 8.37$^{ab}$ | **12.90 ± 5.22$^{b}$** | 23.06 ± 5.77 | 19.95 ± 4.50 | 23.06 ± 5.77 | 19.95 ± 4.50 |
| T. Fatty Acid | 79.41 ± 5.81 | 78.84 ± 7.19 | 79.41 ± 5.81 | 78.84 ± 7.19 | 78.75 ± 7.03 | 82.14 ± 6.26 | 76.25 ± 12.13 | 79.41 ± 5.81 | 78.84 ± 7.19 | 79.41 ± 5.81 | 78.84 ± 7.19 |

Note: SFA= Saturated fatty acid; MUFA= Monounsaturated fatty acid; UFA= Unsaturated fatty acid; T. fatty acid= Total fatty acid; Means in the same row with different superscripts differ significantly ($p<0.05$); nc= Not counted.

fatty acid components, namely caprylic (C8:0) and lauric acids (C12:0), exhibiting negative impacts on health. Previous studies showed that lauric acid increased LDL (low-density lipoprotein) cholesterol levels in the blood, potentially raising the risk of heart disease (Pecina & Ivankovic, 2021). The GG genotype (SNPs g.72400A>G) of the *SCARB1* gene exhibited the highest average lauric acid composition at 0.13%. Furthermore, all SNPs (g.72219C>T, g.72380C>A, g.72400A>G, g.72517G>A, and g.72607C>T) of the *SCARB1* gene were significantly associated with cis-10 heptadecenoic acid (C17:1) and eicosatrienoic acid (C20:3n6). Eicosatrienoic acid (C20:3n6) is classified as an omega-6 fatty acid, constituting essential fats obtainable from food sources. Omega-6 fatty acids can treat neurological problems, alleviate symptoms of inflammatory disorders, enhance metabolism throughout the body, and reduce cardiovascular risk (Calder, 2013; Binia *et al.,* 2017).

This study also found a significant association ($p<0.05$) of the SNP g.72400A>G in the *SCARB1* gene with arachidonic acid (C20:4n6), total MUFA, and UFA. The average value of arachidonic acid was the highest in the GG genotype (0.35), while total MUFA and UFA were found in the AA genotype at 17.91% and 20.55%, respectively. Moreover, unsaturated fatty acids, including omega-3 and omega-6, possessed anti-atherogenic and anti-thrombogenic properties. These characteristics helped reduce the formation of atherosclerotic plaques in blood vessels and inhibit blood clots that could block blood flow (Sakowski *et al.,* 2022; Horcada *et al.,* 2020).

The results indicated that the SREBF1 and *SCARB1* genes exhibited the potential for selection related to fatty acid traits in beef cattle. The new SNP variations in these genes can also serve as valuable information and references to improve meat quality, particularly regarding fatty acids. However, further validation in larger populations and different locations is recommended to confirm the influence of the selected genetic variations.

## CONCLUSION

This study showed that 4 novel SNPs in the *SREBF1* gene and 5 novel SNPs in the *SCARB1* gene significantly influenced the fatty acid composition of Bali cattle. These results suggested that the genetic diversity of the *SREBF1* and *SCARB1* genes exhibited the potential to serve as valuable information or references in the selection of fatty acids in other beef cattle.

## CONFLICT OF INTEREST

Jakaria and Cece Sumantri serve as editor of the Tropical Animal Science Journal but have no role in the decision to publish this article. The authors also declare that there is no conflict of interest with any financial, personal, or other relationships with other people or organizations related to the material discussed in the manuscript.

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