ABSTRACT

Gene C/EBPa is a gene associated with lipid composition and distribution, the gene therefore contributes in enhancing of meat quality. There is no information of sequence variations of C/EBPa gene investigation in local and exotic beef cattle in Indonesia. This research was aimed to detect the presence of sequence variations of C/EBPa gene in local beef and exotic cattle. Twelve samples were used in this study such as Bali cattle (2), Ongole grade (2), Pasundan (2), Friesian Holstein (FH) (1), Angus (1), Limousin (1), Simmental (2), and Banteng (1). Several steps of the research were DNA extraction and quantification, PCR, electrophoresis, sequencing and analysis of sequence gen C/EBPa. Koleksi DNA pada penelitian ini bersumber dari darah dan rambut. Penentuan basa homolog gen C/EBPa berdasarkan hasil BLAST dan variasi sekuens basis diaanalisis dengan MEGA 6. Hasil penelitian ini menunjukkan bahwa semua sapi yang diteliti telah terkonfirmasi memiliki gen C/EBPa yang teramplifikasi (1.339 pb) pada suhu annealing 54°C. Ditemukan tiga variasi basa posiisi 870 (G→A), 931 (A→G) dan 1196 (G→A) pada sapi Bali dan Banteng, tetapi tidak diperoleh pada sapi lokal lain, eksotik dan referensi sekuens Bos taurus (Japanese Black/Acc No. DQ068270.1, Hanwoo/Acc No. DM82984.1, Qinchuan/Acc No. NM_176784.2). Variasi sekuens tersebut menyebabkan perubahan pada asam amino (Alanine → Threonine/931 dan Asparagine → Serine/1196). Mutasi pada posisi 271 menyebabkan perubahan basa sitosin menjadi adenine (C271A) yang ditemukan di sapi PO dan Simmental. Diperlukan penelitian lebih lanjut tentang perubahan asam amino gen C/EBPa pada sapi Bali, sapi PO dan Simmental terhadap kualitas daging.

Kata kunci: Gen C/EBPa, Variasi sekuens, Kualitas daging, Sapi potong lokal, Sapi eksotik

Sequence Variations of CCAAT/Enhancer Binding Protein α (C/EBPα) gene encoding meat quality in local and exotic cattle breeds

ABSTRACT

Gene of C/EBPα is a gene associated with lipid composition and distribution, the gene therefore contributes in enhancing of meat quality. There is no information of sequence variations of C/EBPα gene investigation in local and exotic beef cattle in Indonesia. This research was aimed to detect the presence of sequence variations of C/EBPα gene in local beef and exotic cattle. Twelve samples were used in this study such as Bali cattle (2), Ongole grade (2), Pasundan (2), Friesian Holstein (FH) (1), Angus (1), Limousin (1), Simmental (2), and Banteng (1). Several steps of the research were DNA extraction and quantification, PCR, electrophoresis, sequencing and sequence analysis of C/EBPα gene. DNA was collected either from blood or hair bulbs. Determination of homolog bases of C/EBPα gene was based on BLAST result while base sequence variation was analyzed by using MEGA 6. The result showed that all cattle samples were confirmed bearing the C/EBPα gene that amplified along of 1,339 bp and annealing temperature of 54°C. It was obtained three nucleotide variations at the position of 870 (G→A), 931 (A→G) and 1196 (G→A) only found in Bali cattle and Banteng, but there were no nucleotide variations in the local beef cattle, exotic cattle and the sequence reference of Bos taurus (Japanese Black/Acc No. DQ068270.1, Hanwoo/Acc No. D82984.1, Qinchuan/Acc No. NM_176784.2). Those nucleotide variations caused changing of amino acid of Alanine to Threonine and Asparagine to Serine at 1196. Mutation at position 271 changed nucleotide Cytosine to Adenine (C271A) that found in PO and Simmental cattle. Further research is needed to confirm the changing of amino acid of C/EBPα gene in Bali cattle might affect to meat quality.

Keywords: C/EBPα gene, Sequence variation, Meat quality, Local beef cattle, Exotic cattle

INTRODUCTION

Meat consumption in Indonesia increases up to 9.89% every year since 1993 to 2015 (Kementerian Pertanian, 2017). The national meat stock was dominantly covered 70% by local beef production (Rusuno, 2015) while, the remain covered by importation. Both Bali cattle (Bos sondaicus) and Ongole Grade (PO) cattle (B. indicus) are more preferable fulfil the national beef demand (Wiyatna, 2007; Yosita et al., 2012). Another local beef cattle such as Pasundan (B. indicus) is one of potential cattle to be used as beef (Sulasmi et al., 2017). Exotic cattle, which haveo tropical climate of Indonesia such as Angus (B. taurus), Simmental (B. taurus), and Friesian Holstein or termed as FH (B. taurus) also contribute to fulfil the national beef meat demand. Although the FH is dairy cattle and they still can be used as beef source when not productive anymore.

Improving of beef meat quality can used molecular selection through detecting genes associated with beef meat quality. There are several candidate
genes associated to the traits of beef meat quality. Those are Micromolar Calcium Activated Neutral Protease (CAPN1) (Page et al., 2002), Leptin (LEP) (Schenkel et al., 2005), Calpastatin (CAST) (Schenkel et al., 2006), Fatty Acid Binding Protein 4 (FABP4) (Barendse et al., 2009), Diacylglycerol O-Acyltransferase (DGAT1) (Yuan et al., 2013) and CCAAT/Enhancer Binding Protein a (C/EBPα) (Shin et al., 2007; Wang et al., 2011). One of those is C/EBPα gene associated to lipid composition and distribution in the body of cattle (Shin et al., 2007; He et al., 2011; Wang et al., 2011). The gene contributes in enhancing of the meat quality (Adoligbe et al., 2015).

The gene of C/EBPα plays an important role during differentiation of preadipocyte into adipocyte. The gene was identified as a Nuclear Factor (NF) that induces at differentiation time and binds specifically to promoter of several genes (Vasseur-Cognet and Lane, 1993). Besides the important role in adipocyte differentiation, the C/EBPα gene plays an important role in lipid deposition. The C/EBPα gene is one of important candidate genes that identified to be associated with carcass and meat quality traits in cattle (Shin et al., 2007).

The C/EBPα gene also regulates metabolism of energy and nutrition with activates several specific genes such as phosphoenolpyruvate carboxykinase and insulin receptor (Park et al., 1990). As like in the rat, the C/EBPα gene in bovine is free of intron. It has an open reading frame (nucleotide 169 – 1,230) encoding a protein of 353 amino acid residues which five amino acid are shorter than in the rat protein (Taniguchi and Sasaki, 1996). Those amino acid express in prior transcription of adipocyte specific gene which has site binding of the C/EBPα gene (Wang et al., 2011).

The data relating to C/EBPα gene in cattle has been confirmed in Japanese Black cattle (Taniguchi and Sasaki, 1996). Qinchuan and Hanwoo (B. taurus) cattle (Wang et al., 2012), C/EBPα gene in Qinchuan cattle has been confirmed in the fragment of 1,062 base pairs (bp). Moreover, the C/EBPα gene in Qinchuan cattle has similarity value of amino acid to Sus scrofa (97%), Homo sapiens (95%), Rattus norvegicus (94%), Ornithogalum cuniculus (94%) and Mus musculus (93%). Jeoung et al. (2004) confirmed that the C/EBPα gene in Hanwoo with a long of 1,059 bp encoded 353 amino acids. The highest expression of C/EBPα gene occurred in adipocyte tissue.

This research was designed to embark the possibility of existing C/EBPα gene in local and exotic cattle in Indonesia, also focused on the sequence variation of the C/EBPα gene. The cattle samples involved were therefore focused on breeds consisting of local beef cattle and exotic cattle breeds. Up to present, there was no information relating to C/EBPα gene either in the local beef or exotic cattle in Indonesia. Finding of this research is expected to contribute in selection of beef cattle with a better meat quality since the C/EBPα gene associating with distribution of lipid in the meat or termed as marbling. This is a first step of molecular selecting for meat quality trait in cattle.

### Table 1. Similarity of C/EBPα gene samples with data from the GenBank

| Sample               | References (Bos taurus) | GenBank Accession | Query Cover | Max Iden |
|----------------------|-------------------------|-------------------|-------------|----------|
| Bali (B. sondaicus)  | Japanese Black          | DQ068270.1        | 100%        | 99%      |
|                      | Korean/Hanwoo           | D82984.1          | 100%        | 99%      |
|                      | Chinese/Qinchuan        | NM_176784.2       | 98%         | 99%      |
| Banteng (B. javanicus) | Japanese Black          | DQ068270.1        | 100%        | 99%      |
|                      | Korean/Hanwoo           | D82984.1          | 100%        | 99%      |
|                      | Chinese/Qinchuan        | NM_176784.2       | 96%         | 99%      |
| Pasundan (B. indicus) | Japanese Black          | DQ068270.1        | 100%        | 99%      |
|                      | Korean/Hanwoo           | D82984.1          | 100%        | 99%      |
|                      | Chinese/Qinchuan        | NM_176784.2       | 96%         | 99%      |
| Ongole Grade (B. indicus) | Japanese Black          | DQ068270.1        | 100%        | 99%      |
|                      | Korean/Hanwoo           | D82984.1          | 100%        | 99%      |
|                      | Chinese/Qinchuan        | NM_176784.2       | 95%         | 99%      |
| Friesians Holstein (B. taurus) | Japanese Black          | DQ068270.1        | 100%        | 99%      |
|                      | Korean/Hanwoo           | D82984.1          | 100%        | 99%      |
|                      | Chinese/Qinchuan        | NM_176784.2       | 98%         | 99%      |
| Angus (B. taurus)    | Japanese Black          | DQ068270.1        | 100%        | 99%      |
|                      | Korean/Hanwoo           | D82984.1          | 100%        | 99%      |
|                      | Chinese/Qinchuan        | NM_176784.2       | 98%         | 99%      |
| Limousin (B. taurus) | Japanese Black          | DQ068270.1        | 100%        | 99%      |
|                      | Korean/Hanwoo           | D82984.1          | 100%        | 99%      |
|                      | Chinese/Qinchuan        | NM_176784.2       | 95%         | 99%      |
| Simmental (B. taurus)| Japanese Black          | DQ068270.1        | 100%        | 99%      |
|                      | Korean/Hanwoo           | D82984.1          | 100%        | 99%      |
|                      | Chinese/Qinchuan        | NM_176784.2       | 99%         | 99%      |
MATERIALS AND METHODS

Samples
Amount of twelve (12) samples were involved in this study. Those were three breeds of local beef cattle i.e Bali (2), Ongole Grade (PO) (2), Pasundan (2) and three exotic breeds i.e Angus (1), Simmental (2), Limousin (1), and Frisian Holstein/FH (1). In this study used Banteng (Bos sondaicus) as comparing breed. Bali cattle was from Nusa Penida, Bali, PO cattle was from Grobogan and Pasundan cattle was from BPPT (Balai Pengembangan dan Perbibitan Ternak) at Ciamis of West Java, while those exotic breed cattle were from BET (Balai Embio Ternak) at Cipelang of West Java which. Banteng from Prigen Malang.

DNA Extraction
DNA was extracted from blood samples (Bali, PO and Pasundan cattle) using high salt method (Montgomery and Sise, 1990) and follicle of tail hair (Angus, Simmental, Limousin, FH and Banteng) using DNA extraction kit (gSYSC™ DNA Extraction Kit). The blood samples were taken from vena caudalis and collected at vacutainer tube containing EDTA (anticoagulant). The DNA was quantified by using a spectrophotometer (GeneQuant).

Amplification of C/EBPa Gene
The gene of C/EBPa (1,339 bp) was amplified using PCR with a pair of primer according to Shin et al. (2007). The forward primer was 5'-ACAAACGGGTATAAATGCTG-3' and reverse primer was 5'-AATCTCCTGGTCCTGATTAC-3'. Total volume of PCR reaction was 12.5 µL consisting of 6.25 µL PCR master mix kit (KAPA2G Robust), primer of forward and reverse (100pmol/µl) was 0.5 µL of each primer, 4.75 µL DDW free nuclease and 0.5 µL DNA template. The PCR was conducted in a thermocycler machine (Eppendorf) and set up as follows: pre denaturation 94°C for 5 minutes; denaturation 94°C for 30 second); annealing 54°C for 1 minute 20 second); extension 72°C for 1 minute15 seconds; final extension 72°C for 5 minutes. The PCR

![Figure 1. Visualization PCR product of C/EBPa gene.](image)

Table 3. Changing of Amino Acid in the position of Base Variation in Bali Cattle

| Sample                                      | Triplet Codon | Amino Acid |
|---------------------------------------------|---------------|------------|
| Base Variation at Position 870              |               |            |
| Bali cattle                                 | 868 869 870*  | Alanine    |
| Angus, FH, Simmental, Pasundan, Ongole Grade| G C A         | Alanine    |
| Base Variation at Position 931              |               |            |
| Bali cattle                                 | 931 932 933   | Alanine    |
| Angus, FH, Simmental, Pasundan, Ongole Grade| G C G         | Threonine  |
| Base Variation at Position 1196             |               |            |
| Bali cattle                                 | 1195 1196* 1197| Asparagine |
| Angus, FH, Simmental, Pasundan, Ongole Grade| A A* C       | Serine     |

Remarks: *nucleotide variation position of Bali cattle

Table 2. Position of Base Sequence Variation of C/EBPa Gene with Reference of GenBank Data

| Species _ Breed | Base Sequence Position |
|-----------------|------------------------|
| B. taurus Japanese Black(DQ068270.1) | G T C C A A A C G G T A C A C T G |
| B. taurus Korean Hanwoo(D42984.1) | . . . A . . . C . . . C . . . . . . . |
| B. taurus Qinchuan(NM_176784.2) | A . . T . . . C G T . . . C . . . T C . |
| B. taurus Friesians Holstein | . . . . . . . C . . . . . . . |
| B. taurus Simmental indv_1 | . . . . . . . C . . . . . . . |
| B. taurus Simmental indv_2 | . . . . . . . A . . . C . . . C . . . |
| B. indicus Pasundan indv_1 | . . . . . . . C . . . . . . . |
| B. indicus Pasundan indv_2 | . . . . . . . C . . . . . . . |
| B. indicus Peranakan Ongole indv_1 | . . . . . . . C . . . . . . . |
| B. indicus Peranakan Ongole indv_2 | . . . . . . . A . . . C . . . C . . . |
| B. sondaicus Bali indv_1 | . . . . . . . C . . . A . . . C G . . . A |
| B. sondaicus Bali indv_2 | . . . . . . . C . . . A . . . C G . . . A |
| B. javanicus Banteng | . . . . . . . C . . . A . . . C G . . . A |

Remarks: Numbering of base sequence based on Japanese Black (DQ068270.1)

Dot (.) Same base to Japanese Black (DQ068270.1)
was run for 40 cycles. The PCR products were visualized in 1.0% agarose gel by Electrophoresis (110 V; 30 minutes) with a 1 kb DNA ladder as molecular marker size. The gel was stained with Ethidium Bromide (Bio-Rad, USA) and documented with UV-Illuminator and Gel Doc.

**Sequence Analysis of C/EBPα Gene**

Sequencing of individual PCR product, in total of 12 animal samples was performed by 1st BASE Laboratory services, Malaysia. Total of 30 μl PCR product of each sample were used for sequence analysis. Electropherogram of C/EBPα gene was checked through (http://asparagin.cenargen.embrapa.br/phph/) to see their quality. The sequence results were assembly from two directions by using ChormasPro 1.5. The C/EBPα gene sequences were aligned and compared with B. taurus (Japanese Black cattle/Acc. No. DQ068270.1, Hanwoo cattle/Acc. No. D82984.1, Qinhuang cattle/Acc. No. NM_176784.2) using Multiple Sequence Alignment (MUSCLE) of MEGA ver. 6.0 program (Tamura et al., 2013). Sequence similarity was analysed using BLAST through NCBI (https://blast.ncbi.nlm.nih.gov/Blast.cgi).

**Figure 2.** Base variations of C/EBPα gene in six cattle breed

**Figure 3.** Nucleotide variations (heterozygous detect) of C/EBPα gene in Simmental and PO cattle.
RESULTS AND DISCUSSION

Size and Similarity of C/EBPa Gene

The C/EBPa gene was successfully amplified with the right size of about 1,339 bp (Figure 1) and it was successfully read 1,173 nt in all samples after alignment. Similarity sequences of all samples reached 99% comparing with all references (B. taurus) such as Japanese Black (DQ068270.1), Korean/Hanwoo (D82984.1), Chinese/Qinchuan (NM_176784.2) (Table 1). C/EBPa gene has only one exon that spread 169 to 1,230 in Japanese Black cattle (DQ068270.1) (Taniguchi and Sasaki, 1996; Shin et al., 2007).

Nucleotide variation of C/EBPa gene was successfully obtained in 17 different positions (Table 2). Interestingly, three different nucleotides were found only in Bali (Bos sondaicus) and Banteng cattle at position 870 (G→A), 931 (A→G) and 1196 (G→A) based on Japanese Black (DQ068270.1) (Bos taurus). Those nucleotides were clearly different with the other samples (sharp and clear peak) (Figure 2). Furthermore, heterozygote was detected in PO and Simmental cattle at position 271 where cytosine changed to Adenine (C271A) but did not change of amino acid. This mutation could be detected using restriction enzyme Smal (CCC*GGG) and produced three of genotype variant (AA, AC and CC). In Korean Hanwoo cattle (Bos taurus), three genotypes were successfully detected with allele frequencies A 0.374 and C 0.626. Genotype AA has the highest of carcass weight and meat quality compared with genotype AC and CC (Shin et al., 2007). The two of three variants nucleotides at in Bali cattle caused changing of amino acid. Those changes were Alanine (GCG), Threonine (ACG), and Asparagine (AAC) to Serine (AGC) (Table 3).

Bali cattle (B. sondaicus) is domestication of wild Banteng (Tanib, 2002; Purwantara et al., 2012). Bali cattle has adapted with the tropical climate for hundred years it could contribute the genetic variation in Bali cattle. Bali cattle have been selected naturally and adapted to the tropical climate with lower forage quality (Margawati, 2012) and internal parasite, local disease so creates a specific genetic diversity (Sutarno and Setyawati, 2015). Bali cattle has characteristics of smaller body size compared to B. indicus (tropic) and B. taurus (sub tropic) (Soares and Dryden, 2011). Environment condition influences directly to the metabolism process of muscles and organs (Gregory, 2010). Bali cattle has higher percentage of carcass, 53.26% (Yosita et al., 2012), 54.0% (Wiyatna, 2007), than Madura, PO and Australian Commercial Cattle (ACC).

In quality meat trait, Marbling Score (MS) of Bali cattle in one years old reached 1.92 (Jakaria et al., 2017). Measuring of MS based on AUS-Meat Standard. Sumba Ongole (Bos indicus) has Marbling Score 2-3 (Priyanto et al., 2015) while Limousin and Shorthorn (Bos taurus) have 3 and 4 of MS, respectively (Cundiff et al., 1993). Marbling Score is intramuscular fat area devided by longissimus dorsi muscle area on 12th -13th ribs.

Tropical environment is predicted influencing the lipid composition of beef meat in Bali cattle. This opinion was supported by a research report of Yosita et al. (2012) that lipid composition in subcutan of sub tropic cattle are more excess compared to the cattle from tropical environment. In sub tropic, lipid in sub cutan plays an important role in protecting the body from low temperature environment. Composition of intramuscular lipid correlates to meat quality such as meat tenderness (Reverter et al., 2003). Intramuscular lipid content and internal lipid B. taurus in sub tropic is more excess compared to the cattle from B. indicus (Yosita et al., 2012).

The four nucleotide variations of Bali cattle, PO cattle, Simmental cattle and Banteng in C/EBPa gene are need deep exploration to get more information about association with quality and quantity meat trait. Those information could be used to build of breeding strategy to improve genetic quality of Bali cattle and other Indonesia local cattle next.

CONCLUSIONS

The C/EBPa gene was successfully confirmed with size of 1,173 bp both in local beef cattle (PO, Bali and Pasundan cattle), in exotic cattle (FH, Angus, Limousin, and Simmental) and Banteng. Specific sequence variations was found in Bali cattle (Bos sondaicus) and Banteng, at the positions of 870 (G→A), 931 (A→G) and 1196 (G→A). Those mutations caused amino acid changing Alanine to Threonine (at 931) and Asparagine to Serine (at 1196). Heterozygote genotypes were detected in PO and Simmental at position 271 of C/EBPa gene ytosine changed to Adenine (C271A).

ACKNOWLEDGEMENT

Authors thank to some colleagues at the laboratory of Animal Molecular Genetics who helped during the research was conducted. Extended thank was delivered to the BET Institution which allowed us to collect the tail hair for the exotic cattle samples.

DAFTAR PUSTAKA

Adolige, C., Y.F. Huangfu, L.S. Zan and H. Wang. 2015. C/EBPa gene as a genetic marker for beef quality improvement. Genetics and Molecular Research 14(3): 9370-9383.

Barendse, W., R.J. Bunch, M.B. Thomas and B.E. Harrison. 2009. A splice site single nucleotide polymorphism of the fatty acid binding protein 4 gene appears to be associated with intramuscular fat deposition in longissimus muscle in Australian cattle. Animal Genetics 40: 770-773.
Cundiff, L.V., F. Szabo, K.E. Gregory, R.M. Koch, M.E. Dikeman and J.D. Crouse. 1993. Breed comparisons in the germplasm evaluation program at MARC. Proceeding of Beef Improvement Federation 25th Annual Research Symposium and Annual Meeting. Asheville, North Carolina. pp. 124–136.

Gregory, N.G. 2010. How climatic changes could affect meat quality. Food Research International 43(7): 1866-1873.

He, H., X. Liu, Y. Gu and Y. Liu. 2011. Effect of genetic variation of C/EBPα gene on body measurement and carcass traits of Qinchuan cattle. Molecular Biology Reports 38(22): 4965-4969.

Jakaria, H. Khasanah, R. Priyanto, M. Baihaqi and M.F. Ulum. 2017. Prediction of meat quality in Bali cattle using ultrasound imaging. Journal of the Indonesian Tropical Animal Agriculture. 42(2): 59-65.

Kementerian Pertanian. 2017. Outlook Daging Sapi Komoditas Pertanian Subsektor Peternakan. Pusat Data dan Sistem Informasi Pertanian. Kementrian Pertanian. Jakarta.

Margawati, E.T. 2012. A global strategy of using molecular genetic information to improve genetics in livestock. Reproduction in Domestic Animals 47(Suppl. 1): 7-9.

Montgomery, G.W. and J.A. Sise. 1990. Extraction of DNA from sheep white blood cells. New Zealand Journal of Agricultural Research 33(3): 437-441.

Page, B.T., E. Casas, M.P. Heaton, N.G. Cullen, D.L. Hyndman, C.A. Morris, A.M. Crawford, T.L. Wheeler, M. Koohmaraie, J.W. Keele and T.P.L. Smith. 2002. Evaluation of single-nucleotide polymorphisms in CAPN1 for association with meat tenderness in cattle. Journal of Animal Science. 80:3077-3085.

Park, E.A., J. W.T. Roesler, J. Liu, D.J. Klemm, A.L. Gurney, J.D. Thatcher, J. Shuman, A. Friedman and R.W. Hanson. 1990. The role of the CCAAT/Enhancer-Binding Protein in the transcriptional regulation of the gene for phosphoenolpyruvate Carboxykinase (GTP). Molecular and Cellular Biology 10(12): 6264-6272.

Priyanto, R., M.F. Asnath, L.A. Edit, M. Baihaqi and M. Ismail. 2015. Peningkatan produksi dan kualitas daging sapi lokal melalui pengemukan berbasis serealia pada taraf energi yang berbeda. Jurnal Ilmu Pertanian Indonesia 20: 108-114.

Purwanta, B., R.R. Noor, G. Andersson and H. Rodriguez-Martinez. 2012. Banteng and Bali cattle in Indonesia: status and forecasts. Reproduction in Domestic Animals 47(1): 2-6.

Reverter, A., D.J. Johnston, D.M. Ferguson, D. Perry, M.E. Goddard, H.M. Burrow, V.H. Oddy, J.M. Thompson and B.M. Bindon. 2003. Genetic and phenotypic characterisation of animal, carcass, and meat quality traits from temperate and tropically adapted beef breeds. 4. Correlations among animal, carcass, and meat quality traits. Australian Journal of Agricultural Research 54: 149-158.

Rusono, N. 2015. Peningkatan Produksi Daging Sapi untuk Mewujudkan Kedaulatan Pangan Hewani. In: Prosiding Seminar Nasional Teknologi Peternakan dan Veteriner. Malang. pp. 12-21.

Schenkel, F.S., S.P. Miller, X. Ye, S.S. Moore, J.D. Nkrumah, C. Li, J. Yu, I.B. Mandell, J.W. Wilton and J.L. Williams. 2005. Association of single nucleotide polymorphisms in the leptin gene with carcass and meat quality traits of beef cattle. Journal of Animal Science 83(9): 2009-2020.

Schenkel, F.S., S.P. Miller, Z. Jiang, I.B. Mandell, X. Ye, H. Li and J.W. Wilton. 2006. Association of a single nucleotide polymorphism in the Calpastatin gene with carcass and meat quality traits of beef cattle. Journal of Animal Science 84: 291-299.

Shin, S. C., M.J. Kang and E.R. Chung. 2007. Identification of a novel SNP associated with meat quality in C/EBPα gene of Korean cattle. Asian Australian Journal Animal Science 20(4): 466-470.

Soares, F.S. and G. McL. Dryden. 2011. A body condition scoring system for Bali cattle. Asian-Australasian Journal of Animal Sciences 24(11): 1587-1594.

Sulasmi, A. Gunawan, R. Priyanto, C. Sunantri and J. Arifin. 2017. Keseragaman dan kedekatan morfometrik ukuran tubuh sapi pasundan. Jurnal Veteriner 18(2): 263-273.

Sutarno and A.D. Setyawan. 2015. Review: Genetic diversity of local and exotic cattle and their crossbreeding impact on the quality of Indonesian cattle. Biodiversitas 16(2): 327-354.

Talib, C. 2002. Bali cattle in the breeding stock areas and their future development. Wartazoa 12(3): 100-107.

Tamura, K., G. Stecher, D. Peterson, A. Filipski and S. Kumar. 2013. MEGA6: molecular evolutionary genetics analysis version 6.0. Molecular Biology and Evolution 30(12): 2725-2729.

Taniguchi, Y. and Y. Sasaki. 1996. Rapid communication: nucleotide sequence of bovine C/EBPα gene. Journal Animal Science 74(10): 2554.

Vasseur-Cognet, M. and M.D. Lane. 1993. CCAAT/Enhancer Binding Protein alpha (C/EBP Alpha) undifferentiated protein: a developmentally regulated nuclear protein that binds to The C/EBP Alpha gene promoter. Proceedings of the National Academy of Sciences of the United States of America 90(15): 7312-7316.

Wang, H., L.S. Zan, H.B. Wang, C. Gong and C.Z. Fu. 2012. Cloning, expression analysis and sequence prediction of the CCAAT/Enhancer-Binding Protein Alpha gene of Qinchuan cattle. Genetics and Molecular Research 11(2): 1651-1661.
Wang, H., L.S. Zan, H.B. Wang and F.B. Song. 2011. A novel SNP of the C/EBPα gene associated with superior meat quality in indigenous Chinese cattle. Genetics and Molecular Research 10(3): 2069-2077.

Wiyatna, M. F. 2007. The ratio of meat index of Indonesian cattle (Bali, Madura PO) with Australian cattle (Australian Commercial Cross). Jurnal Ilmu Ternak 7(1): 22-25.

Yosita, M., U. Santosa and E.Y. Setyowati. 2012. Persentase karkas, tebal lemak punggung dan indeks perdagingan Sapi Bali, Peranakan Ongole dan Australian Commercial Cross. Students E-Journal 1(1): 15-20.

Yuan, Z., J. Li, J. Li, X. Gao, H. Gao and S. Xu. 2013. Effects of DGAT1 gene on meat and carcass fatness quality in Chinese commercial cattle. Molecular Biology Reports 40(2): 1947-1954.