J I T A A
Journal of the Indonesian Tropical Animal Agriculture
Accredited by Ditjen Penguatan Risbang No. 60/E/KPT/2016

J. Indonesian Trop. Anim. Agric.
pISSN 2087-8273 eISSN 2460-6278
http://ejournal.undip.ac.id/index.php/jitaa
42(4):213-219, December 2017
DOI: 10.14710/jitaa.42.4.213-219

Genetic polymorphism of kappa-casein gene in Friesian Holstein: a basic selection of dairy cattle superiority

S. D. Volkandari*, Indriawati and E. T. Margawati
Research Center for Biotechnology, Indonesian Institute of Sciences
Jl. Raya Bogor km 46, Cibinong, Bogor, West Java, Indonesia 16911
**Corresponding E-mail : volkandari@gmail.com*

Received April 28, 2017; Accepted September 13, 2017

ABSTRAK

Kasein adalah protein susu yang terbagi dalam empat grup yaitu αS1, αS2, β-casein dan kappa-casein (CSN3). Gen kappa-kasein berpengaruh terhadap pembentukan komponen dalam susu. Tujuan dari penelitian adalah mengidentifikasi polimorfisme gen kappa-kasein pada sapi perah Friesian Holstein (FH). Sebanyak 59 sampel sapi FH yaitu 32 asal Malang, 10 asal Sukahati Bogor dan 17 asal Pusat Penelitian Bioteknologi-Lembaga Ilmu Pengetahuan Indonesia (LIPI) (ternak koleksi) digunakan dalam penelitian. Sampel DNA diekstraksi dengan metode garam pekat dan dikuantifikasi menggunakan Spektrofotometer. Fragmen DNA spesifik gen kappa-kasein sepanjang 379 pb diamplifikasi menggunakan metode *Polymerase Chain Reaction* (PCR) dengan suhu *annealing* 56^{o}C dan 30 siklus. Teknik PCR-RFLP dengan enzim restriksi *Hind*III digunakan untuk analisis genotip. Hasil analisis menunjukkan bahwa terdapat tiga varian genotip yaitu AA, AB dan BB pada populasi Malang dan ternak koleksi Puslit Bioteknologi-LIPI sementara dua varian (AA dan BB) ditemukan pada populasi Sukahati. Rata-rata frekuensi genotip AA sebesar 65,28%, AB 31,72% dan BB 3,00% dengan rata-rata frekuensi alel A sebesar 0,81 dan B sebesar 0,19. Populasi sapi FH dalam keseimbangan genetik. Kesimpulan, polimorfisme gen kappa-kasein ditemukan pada ketiga populasi sapi FH dengan frekuensi alel A lebih tinggi dibanding alel B. Alel B diketahui berhubungan dengan produksi susu, komposisi susu dan *cheese yield*. Peningkatan alel B pada populasi FH berdampak terhadap performans produksi dan komponen susu. Eksplorasi (kuantitatif, kualitatif, dan molekuler) perlu dilakukan untuk aplikasi genetika molekuler pada seleksi sapi perah.

Kata Kunci: gen kappa casein, polimorfisme, sapi perah, Friesian Holstein

ABSTRACT

Caseins are milk protein subdivided into four main groups which are αS1, αS2, β-casein and kappa-casein (CSN3). Kappa-casein gene influences the manufacturing of milk properties. The aim of this study was to identify the kappa-casein gene polymorphism in Friesian Holstein (FH) cattle. Fifty nine (59) samples consisted of 32 (Malang), 10 (Sukahati Bogor) and 17 (Research Center for Biotechnology-Indonesian Institute of Sciences's collections) were applied in this study. DNA samples were extracted by high concentrated NaCl and quantified by spectrophotometer. The kappa-casein gene was amplified at 379 bp fragment by PCR method using a pair primer of kappa-casein at 56^{o}C annealing for 30 cycles. PCR-RFLP technique with *Hind*III was used for genotyping analysis. The result showed that there were three variants of genotypes (AA, AB and BB) in two populations from Malang and RC for Biotechnology-LIPI's collection while cattle from Sukahati had only AA and AB genotypes. The averages of genotype frequencies were 65.28%; 65.28%; and 3.00% for AA, AB and BB genotypes

respectively while frequencies of 0.81 and 0.19 were for A and B alleles, respectively. FH cattle populations were in equilibrium genetics. This finding concludes that polymorphism was found in three of FH populations with A allele was more common in kappa-casein locus. B allele is known having association with milk production, milk component and cheese yield. Increasing of B allele would influence on milk performance of FH cattle. Explorations of quantitative, qualitative and molecular genetics are important to improve dairy cattle performance.

Keywords :kappa casein gene, polymorphism, Friesian Holstein, dairy cattle

INTRODUCTION

Milk yield is a polygenic trait which is affected by environmental factors (Akyuz *et al*., 2013). The yield and quality of milk in modern breeding programs can be expressed through the quantity of milk, fat and protein in milk. Normally, cow's milk contains 3-5% protein, of which 80% is casein and 20% is whey (Patel *et al*., 2007a). The specific proteins of bovine milk are in the form of casein fractions which divided into four fractions of α-1, α-2, β and kappa-casein (insoluble fraction), α-lactalbumin and β-lactoglobulin. Those 4 fractions are classified as soluble fraction (El-Rafey and Darwish, 2008).

Kappa-casein is the most interesting especially for a milk protein polymorphism since it correlates to milk quality and composition (Scheeper *et al*., 2010). The kappa casein molecule is a single chain polypeptide of 169 amino acids with a molecular weight of 19.2 kDa (Rachagani and Gupta, 2008). Kappa casein constitutes approximately 12% of total casein (Azevedo *et al*., 2008). The total length of kappa casein gene is approximately of 13 kb sequence that divided into 5 exons (Alexander *et al*., 1988) and located on chromosome 6p31. There are fourteen (14) polymorphism that were described as A, A1, B, B2, C, D, E, F, F1, F2, G1, G2, H, I and J (Caroli *et al*., 2009). Mutation points in exon IV have determined two allele variants of A and B variants which are differed in the amino acids of 136 and 148. Position of 136 (A variant) threonine (Thr) was replaced by isoleucine (IIe) while position of 148 (B variant) aspartic (Asp) was replaced by alanine (Ala) (Alexander *et al*., 1988). Many researchers have reported that genotype in kappa casein gene associated with milk yield (Vidović *et al*., 2013; Deb *et al*., 2014), milk fat yield (Djedović *et al.,* 2015), and cheese yield (Azevedo *et al*., 2008). In buffalo, kappa casein gene was observed for milk production traits (Patel *et al*., 2007b; Margawati *et al*., 2016).

Quantitative traits (milk yield, fat and protein yield) has been used in the selection of dairy sires and cows. Meanwhile, genetic improvement of quantitative traits is relatively slow (Lukac *et al*., 2013), therefore molecular approach study could be applied in the selection (Marker Assisted Selection/MAS). Identification of milk protein genetic polymorphism has interested many researchers (Contreras *et al.,* 2011; Deb *et al*., 2014; Djedović *et al.,* 2015) because of its possibility in association of milk protein genotypes and economically important traits in dairy cattle (Contreras *et al.,* 2011).

Study of kappa casein gene of Friesian Holstein cattle in Indonesia is needed to improve the genetic quality of milk performance (milk yield and milk content). The purpose of this research was to determine the polymorphism of kappa-casein gene in Friesian Holstein dairy cattle as a basic selection to get superior dairy cattle.

MATERIALS AND METHODS

Blood Samples

Fifty nine (59) fresh blood of Holstein cattle were collected from several locations are presented in Table 1. Five to ten (5-10) milliliter bloods per sample were taken from *vena jugular* into a vacutainer tube containing K_3EDTA (BD Vacutainer Systems, Plymouth, UK). Blood samples were stored at 4^{o}C for DNA extraction.

DNA Extraction and Quantification

DNA extraction was performed using standard procedure of salting out (Montgomery and Sise, 1990). DNA quantity and quality were determined by measuring the absorbance at 260 nm for DNA concentration and 260/280 nm for DNA purity using GeneQuant Pro spectrophotometer (Amersham Bioscience, UK). Working dilution of extracted DNA was prepared for each sample at a concentration of 50 ng /µl. The extracted DNA was appropriately labeled and stored at -20^{o}C until for analysis.

Table 1. Blood samples of FH cattle in this study

Origin	N
Malang, East Java	32
Sukahati Bogor, West Java	10
Research Center for Biotechnology-Indonesian Institutes of Sciences's collection, West Java	17
Total	59

N: Total number of samples

PCR Amplification of Kappa Casein Gene

Primer sequences for Kappa-Casein gene PCR was adopted from Mitra *et al.* (1998). Forward primer KCN-F was 5'-CACGTCACCCACACCCACATTTATC-3' while reverse primer KCN-R was 5'-TAATTAG CCCATTTCGCCTTCTCTGT-3'. Total volume reaction of PCR was 20 µl containing 2 µl genomic DNA (50 ng/ µl), 2 µl each primers (10 pmol/µl), 10 µl PCR Master Mix (DreamTaq Green PCR Master Mix (2x), Thermoscientific) and 6 µl water nuclease-free (Thermo Fisher Scientific, USA). The PCR was carried out in Master cycler Gradient Eppendroft (Germany) thermal cycler. PCR program was set up as follows: 1 min at 95°C, 1 min at 95°C, 1 min at 56°C, 1 min at 72°C, 30 cycles and 5 min at 72°C. The PCR product was visualized using 1% agarose gel. The gel was stained with ethidium bromide and DNA fragments were visualized using a UV transimullator (Major Science, USA). A 100 bp DNA ladder (VC 100 bp Plus_Vivantis, USA) was lined up as molecular size marker.

Genotyping of Kappa-Casein Gene using RFLP technique

Restriction Fragment Length Polymorphism (RFLP) method with *Hind*III restriction enzyme was applied for genotyping analysis of kappa-casein gene (Mitra *et al.,* 1998). Total volume of mixture reaction was consisted of 10 µl containing 5 µl PCR product, 1 µl Buffer R with BSA, 0.2 µl *Hind*III (10 U/µl) enzyme (Fermentas, Germany), and 3.8 µl DDW. The mixture reaction was incubated at 37^{o}C for 4 h. Digested products were analyzed using electrophoresis on 2% agarose gel. The gel was stained with ethidium bromide (Applichem, USA) and visualized using UV Transimullator (MUV21, MajorScience, USA) for specific fragment pattern. Expected fragment sizes are shown in Table 2.

Table 2. Expected Fragment Sizes

PCR Product (bp)	Restriction Fragments Expected Sizes (bp)		
	AA	AB	BB
379	379	379; 225; 154	225; 154

Statistical Analysis

Estimations of genotypes and allele frequencies of Kappa-Casein gene were determined by calculation (Nei and Kumar, 2000) as follows:

$$x_{ii} = \frac{\sum N_{ii}}{N} \qquad x_{ij} = \frac{\sum N_{ij}}{N}$$

$$x_i = \frac{(2N_i + N_j)}{2N} \qquad x_j = \frac{(2N_j + N_{ij})}{2N}$$

Where:

x_{ii} : Genotype frequency of A_iA_i (homozygote)

x_{ij} : Genotype frequency of A_iA_j (heterozygote)

x_i : Allele frequency of A_i

x_j : Allele frequency of A_j

n_{ii} : Genotype of $A_i A_i$

n_{ij} : Genotype of $A_i A_j$

N_i : Allele A_i

N_j : Allele A_j

The probability of Hardy-Weinberg equilibrium associated with the observed genotypic frequencies was obtained using the *Chi-Square* (χ^2) test (Hartl, 1988):

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

Where :

χ^2 : *Chi-Square test* value

O : the number of observed genotypes

E : the number of expected genotypes

RESULTS AND DISCUSSION

The PCR product of the kappa-casein gene using specific set of primers (KCN-F and KCN-R) was a 379 bp fragment (Figure 1) that stretched along from nucleotide position of 197 to 574 based on GenBank Accesion No. AJ841946.1 (Figure 2).

A 379 bp fragment DNA of kappa casein gene after digested by *Hind*III restriction endonuclease (genotyping analysis) generated three fragments of 379, 225 and 154 bp. One fragment of 379 bp represented AA homozygote, and two fragments of 225 and 154 bp represented BB homozygote while three fragments of 379, 225 and 154 bp represented AB heterozygote (Figure 3).

Average of genotype frequencies of kappa-casein gene FH cattle in this study was 65.28%; 31.72% and 3.00% for AA, AB and BB respectively while allele frequencies were 0.81 for A and 0.19 for B allele (Table 3). This result was agreed to the previous study reported by several researchers. Khaizaran and Al-Razem (2014) showed a similar result that A and B genotypes were favorable alleles. A allele of 0.81 was more frequently presented than B allele (0.19) in Palestinian Holstein-Friesian cattle. The observation made by Keating *et al*. (2007) showed that A allele was dominant allele than B allele among various dairy cattle population. In Poland, Sitkowska *et al.* (2008) reported that A allele in

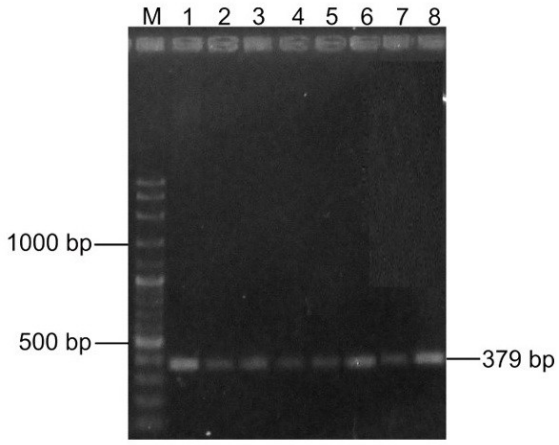

Figure 1. PCR product of Kappa Casein gene in FH cattle (379 bp). M: Ladder 100 kb; lane 1-8: FH samples

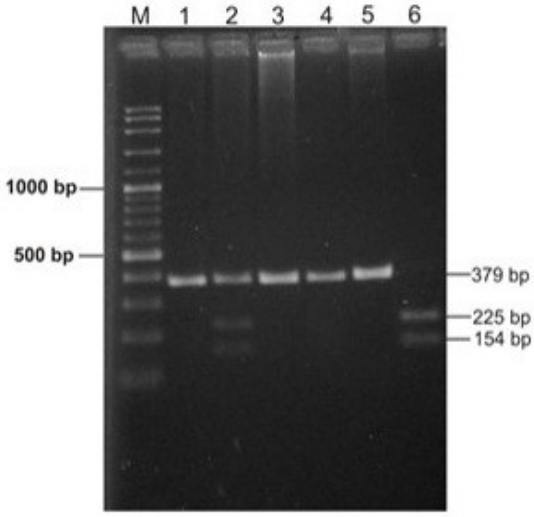

Figure 3. PCR-RFLP product of Kappa Casein gene using *Hind*III enzyme. M: Ladder 100 kb; lanes 1,3,4,5: AA genotype; lane 2: AB genotype; lane 6: BB genotype.

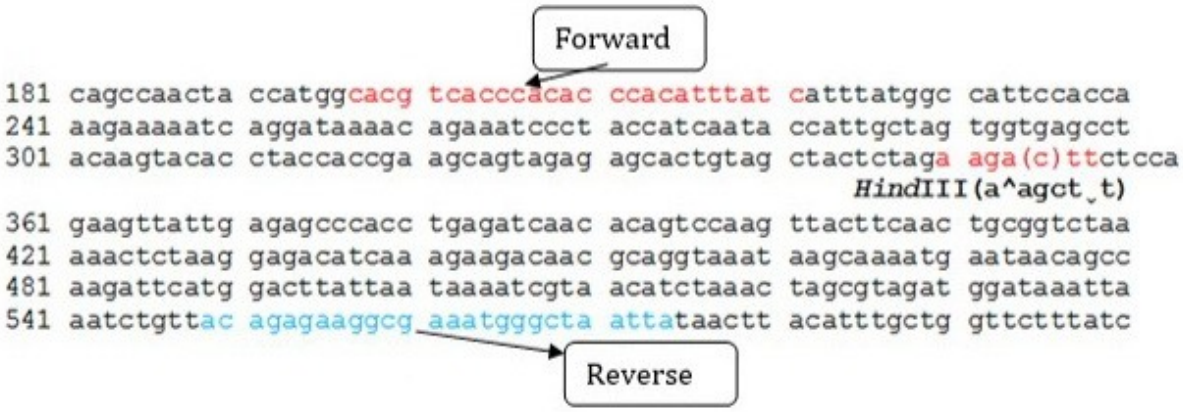

Figure 2. Primer and *Hind*III restriction site of Kappa casein gene based on GenBank Accesion No. AJ841946.1

Table 3. Genotype and allele frequencies, and Hardy Weinberg Equilibrium of Kappa Casein gene in FH cattle population

Samples	N		K-CN Genotype			Allele Frequencies		x^2
			AA	AB	BB	A	B	
Malang	32	Observed	19 (59.38%)	12 (37.50%)	1 (3.12%)	0.78	0.22	0.30
		Expected	19.47	10.98	1.55			
Sukahati, Bogor	10	Observed	6 (60%)	4 (40%)	0 (0%)	0.80	0.20	0.63
		Expected	6.40	3.20	0.40			
RC for Biotech's collection	17	Observed	13 (76.47%)	3 (17.65%)	1 (5.88%)	0.85	0.15	1.45
		Expected	12.28	4.33	0.38			
Total	59							
Average of genotypes freq			65.28	31.72	3.00			
Average of allele freq						0.81	0.19	

x^2 value = 5.99

N: Total number of samples

Holstein-Friesian hwas higher than B allele (0.83 and 0.17, respectively).

Crossbred cattle of meat and dairy cattle showed that A allele was more dominant than B allele. Trakovickà *et al*. (2012) reported that frequency of A allele was more frequent than B allele in crossbred of Simmental and Holstein cattle. In Frieswal cattle (Friesian x Sahiwal) showed that A allele (0.58) was higher than B allele (0.42) (Deb *et al*., 2014).

On the contrary, Jersey cattle had B allele that was more dominant than A allele. Zepeda-Batista *et al*. (2015) found that Mexican Jersey cattle had three alleles (A, B and E). Frequency of B allele (0.69) was higher than that of A allele (0.26) and E allele (0.05). Similar finding of kappa-casein gene in Jersey cattle from China was found by Ren *et al*. (2011) that reported B allele (0.88) more frequent than A allele (0.11). Friesian Holstein populations in this study were in equilibrium genetic (Table 3).

Previous studies reported that B allele had a favorable and significant effect on both milk yield and milk protein contents (Azevedo *et al*., 2008; Caroli *et al*., 2014; Morkūnienė *et al.,* 2016). Vidović *et al*. (2013) reported that Holstein cows with BB had higher percentage of milk fat than AA and AB genotypes. Furthermore, Alipanah *et al*. (2008), indicated that polymorphism of kappa-casein gene affected fat percentage in Black Pied breed (Russian cattle). BB genotype showed a higher fat percentage (4.79±0.23) than AA and BB alleles which were 4.42±0.20% and 4.30±0.19%, respectively. In Sahiwal and Tharparker cattle, Rachagani *et al*. (2008) found that BB genotypes of those cattle had higher Solid Non Fat (SNF) percentage and yield of monthly production than AA and AB genotypes. Another study of Deb *et al.* (2014) reported that in Frieswal cattle, AB genotype had a significant higher total milk yield, peak yield and SNF for 300 days than AA genotypes. Similarly result also reported by Gurses and Yuce (2012) that AB genotype of East Anatolian Red cattle (Turkey cattle) had a higher percentage of SNF and protein content milk than AA genotype.

As reported by Azevedo *et al*. (2008) and Burbano *et al*. (2010) that Allele of B has favorable effect in cheese yield. According to Bittante *et al*. (2012), κ-casein BB variant of milk has superior renneting properties with shorter rennet coagulation time (RCT), faster rates of curd firming and producing a firmer curd compared to other κ-casein variants.

Increasing frequency of B allele and BB genotype in FH population could help to improve milk production traits (milk yield, milk protein content) through selection based on allele and genotype. Selected animals having BB and AB genotypes could be crossed to generate more animals with BB genotypes.

CONCLUSION

All three populations of Friesian Holstein cattle were polymorphic which AA genotypes and A allele at locus κ-casein were commonly found. Increasing of B allele is expected to improve of milk performance in FH cattle. Milk performance (milk yield, milk component) needs to be explored by application of molecular genetic markers for dairy cattle selection.

ACKNOWLEDGMENTS

The research was supported by DIPA-LIPI research grant (079.01.2.450083/2016). Authors thank to Muhammad Ridwan for laboratory assistance and people who involved in blood collection in the field.

REFERENCES

Akyüz, B., K. Arslan, D.M. Bayram and K.M. Iscan. 2013. Allelic Frequency of Kappa-Casein, Growth Hormone and Prolactin Gene in Holstein, Brown Swiss and Simmental Cattle Breeds in Turkey. Kafkas Universitesi Veteriner Fakultesi Dergisi 19:439-444.

Alexander, J.L., A.F. Stewart, A.G. Mackinlay and T.V. Kapelinskaya. 1988. Isolation and characterization of the bovine kappa-casein gene. Eur. J. Biochem. 178:395-401.

Alipanah, M., L. Kalashnikova and G. Rodlonov. 2005. Kappa-casein genotypic frequencies in Russian breeds Black and Red Pied cattle. Iran. J. Biotechnol. 3:191-194.

Alipanah, M., L. A. Kalashnikova and G.V. Rodionov. 2008. Kappa-Casein and PRL-RsaI genotypic frequencies in two Russian cattle breeds. Arch. Zootech. 57(218):131-138.

Azevedo, A.L.S., C.S. Nascimento, R.S. Steinberg, M.R.S. Carvalho, M.G.C.D. Peixoto, R.L. Teodoro, R.S. Verneque, S.E.F. Gulmaràes and M.A. Machado. 2008. Genetic polymorphism of the kappa casein gene in Brazilian cattle. Genet. Mol. Res. 7:623-630.

Bittante, G., M. Penasa and A. Cecchinato. 2012. Invited review: Genetics and modeling of milk coagulation properties. J. Dairy Sci. 95:6843-6870.

Burbano, G.L.Z., Y.M.E. Cabrera, C.E.S. Portilla and C.Y.R.Galindo. 2010. Kappa casein genotypes and curd yield in Holstein cows. Rev. Colomb. Ciene. Pecu. 23:422-428.

Contreras, V.I.P., D.I.L. Jaramillo, G.M.P. Bracamonte, J.C.M. González, and A.M.S. Rincón. 2011. Covenient genotyping of nine bovine K-casein variants. Electron. J. Biotechnol. 14(4):1-6. DOI: 10.2225/vol14-issue4-fulltext-10

Caroli, A.M., S. Chessa and G.J. Erhardt. 2009. Milk protein polymorphisms in cattle: Effect on animal breeding and human nutrition. J. Dairy. Sci. 92:5335-5352.

Deb, R., U. Singh, S. Kumar, R. Singh, G. Sengar and A. Sharma. 2014. Genetic polymorphism and association of kappa-casein gene with milk production traits aamong Frieswal (HF X Sahiwal) cross breed of Indian origin. Iran J. Vet. Res. 15:406-408.

Djedović, R., V. Bogdanović, P. Perišić, D. Stanojević, J. Popović and M. Brka. 2015. Relationship between genetic polymorphism of K-Casein and Quantitative milk yield triats in cattle breeds and crossbreds in Serbia. Genetika. 47:23-32.

El-Rafey, G.A and S.F. Darwish. 2008. A PCR-RFLP assay to detect genetic variants of kappa-casein gene in cattle and buffalo. Arab J. Biotech 11:11-28.

Gurses, M and H. Yuce. 2012. Determination of Kappa Casein gene polymorphisms and their effects on milk composition in some native cattle breeds of Turkey. J. Anim. Vet. Adv. 11(7):1023-1027.

Hartl, D.L. 1988. Principle of Population Genetic. Sunderland: Sinauer Associates, Inc. Publisher.

Keating, A.F., P. Davoren, T.J. Smith, R.P. Ross and M.T. Cairns. 2007. Bovine Kappa-Casein promoter haplotypes with potential implications for milk protein expression. J. Dairy Sci. 90:4092-4099.

Khaizaran, Z.A and F. Al-Razem. 2014. Analysis of selected milk traits in Palestinian Holstein-Friesian cattle in relation to genetic polymorphism. J. Cell Anim. Biol. 8(5):74-85.

Margawati, E.T., S.D. Volkandari, Indriawati and C. Talib. 2016. Genotyping of kappa-casein gene of buffalo in Indonesia. Proceedings, International Conference on Livestock Production and Veterinary Technology, Indonesian Center for Animal Research and

Development, Ministry of Agriculture, Bali, Indonesia, 10-12th August, 2016. P. 37-44.

Mitra, A., P. Schlee, I. Krause, J. Blusch, T. Werner, C.R. Balakrishnan and F. Pirchner. 1998. Kappa-casein polymorphism in Indian dairy cattle and buffalo: a new genetic variant in buffalo. Anim. Biotechnol. 9:81-87.

Montgomery, G.W and J.A. Sise. 1990. Extraction of DNA from sheep white blood cell. New Zeal. J. Agric. Res. 33:437-441.

Morkūnienė, K., L. Baltrėnaitė, A. Puišytė, R. Bižienė, N. Pečiulaitienė, N. Makštutienė, R. Mišeikienė, I. Miceikienė and S. Kerzienė. 2016. Association of Kappa Casein Polymorphism with Milk Yield and Milk Protein Genomic Values in Cows Reared in Lithuania. Vet. Med. Zoot. 74(96):27-32.

Nei, M. and S. Kumar. 2000. Molecular Evolution and Phylogenetics. New York: Oxford University Press.

Patel, R.K., J.B.Chauhan, K.M.Singh and K.J. Soni. 2007a. Allelic frequency of Kappa-Casein and Beta-Lactoglobulin in Indian Crossbred (Bos taurus x Bos indicus) dairy bulls. Turk. J. Vet. Anim. Sci. 31(6):399-402.

Patel, R.K., J.B. Chauhan, K.M. Singh and K.J. Soni. 2007b. Genotype and allele frequencies of κ-casein and β-lactoglobulin in Indian river buffalo bulls (*Bubalus bubalis).* Buffalo Bull. 26(3):63-66.

Rachagani, S and I.D. Gupta. 2008. Bovine kappa-casein gene polymorphism and its association with milk production traits. Genet. Mol. Biol. 31:893-897.

Ren, D.-X., S.-Y. Miao, Y.-L. Chen, C.-X. Zou, X.-W. Liang and J.-X. Liu. 2011. Genotyping of the κ-casein and β-lactoglobulin genes in Chinese Holstein, Jersey and water buffalo by PCR-RFLP. J. Genet. 90:e1-e5.

Scheepers, R.C., E. Marle-Koster and C.Visser. 2010. Genetic variation in the kappa-casein gene of South African goats. Small Rumin. Res. 93:53-56.

Sitlowska B., W. Neja and E. Wiśniewska. 2011. Relation between kappa-casein polymorphism (CSN3) and milk performance traits in heifer cows. J. Cent. Eur. Agr. 9(4):641-644.

Trakovickà, A., N. Moravčikovà and A. Navràtilovà. 2012. Kappa-casein gene polymorphism (CSN3) and its effect on milk production traits. Acta fytotechnica et zootechnica. Nitra, Slovaca Universitas Agriculturae Nitriae. (Suppl 3/2012):61-64.

Vidović, V., Z. Nemes, A. Popović-Vranjes, D. Lucač, D. Cvetanovic, L. Strbac and M. Stupar. 2013. Heritability and correlations of milk traits in the view of kappa-casein genotypes in Vojvodina Holstein-Friesian dairy cattle. Mljekarstvo 63:91-97.

Zepeda-Batista, J.L., B. Alarcón-Zúñiga, A. Ruíz-Flores, R. Núñez-Domínguez and R. Ramírez-Valverde. 2015. Polymorphism of three milk protein genes in Mexican Jersey cattle. Electron. J. Biotechnol. 18:1-4.