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Research article

Variation in *FABP4* gene associated with the morphometric traits in Albanian sheep

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Abstract: Fatty acid-binding protein 4 (FABP4) participates in fatty-acid transportation and variation in the gene has been reported to affect meat quality and growth traits in sheep. The study aimed to identify the association of variation in the intron 3 of the FABP4 gene in an Albanian sheep population with morphometric traits. The analysis covered a total of 70 individuals randomly selected. The molecular analysis detected two alleles A and G with frequencies 0.707 and 0.293 respectively and three genotypes. All morphometric traits have a significant positive correlation. The fixation index was -13.8% indicating a moderate level of outbreeding. The results indicated no significant effect of this locus on morphometric traits.

Keywords: body measurements; correlation; genetic diversity; General Linear model

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Introduction

Sheep farming is quite common in Albania and with a great interest for the local community. There are different local sheep breeds in Albania, especially with the triple purpose (Hoda and Marsan 2012). Recently, lambs are the most important product of sheep, due to the increased culinary demand of consumers. Local sheep breeds are characterized by the use of different markers like SNP (Hoda et al., 2011), microsatellites (Hoda et al., 2009), AFLP (Hoda et al., 2010) or mtDNA (Hoda et al., 2014). The polymorphism of casein genes in different local sheep breeds is reported previously (Bozgo et al., 2020).

Characterization of genes that are directly or indirectly associated with important productive traits plays an important role in the selection of farm animals. The utilization of candidate genes is one of the primary methods to determine the specific genes related to the economic traits in farm animals (Khalil 2020). Recently, single nucleotide polymorphisms (SNPs) of many candidate genes have been specifically determined (Li et al., 2013). Hu et al. (2018) reported the need to study the DNA markers associated with the productive qualities of sheep for greater efficiency of breeding and increasing the profitability of the sheep industry. *FABP4* was proposed as a potential candidate gene for meat quality and fat metabolism (Q. et al., 2018). Ovine *FABP4* gene harbors substantial diversity (Shafey et al., 2020). The fatty acid-binding protein 4 (*FABP4*) plays a role in lipid metabolism and also in transporting long chain fatty acids in mammals. Variation in the *FABP4* gene has been found to be associated with growth, fat deposition and carcass traits in cattle. Ibrahim et al., (2019) suggested an association between variation in ovine *FABP4* gene and milk production traits in Greek Sfakia sheep (Ibrahim et al., 2019). Yan et al., (2018) concluded that variation in *FABP4* affects growth and meat production in sheep. In the present study, we intend to identify the intron 4

polymorphism in the *FABP4* gene and its association with different morphometric traits of Albanian sheep.

Material and Methods

A total of 70 blood samples were collected from randomly selected unrelated individuals of Albanian sheep breed. Eight morphometric traits were measured from these individuals; wither height (WH), rump height (RH), body length (BL), chest depth (CD), chest width (CW), rump width (RW), chest circumference (CC). The salt out extraction method was used to extract DNA from blood samples. Genotyping of SNPs in the selected marker was performed as described previously (Hoda et al., 2011).

Calculation of genetic diversity indices, including genotypes, alleles frequencies, mean expected and observed heterozygosities, and Chi-square test was performed using PopGene32 (ver. 1.32) (Yeh et al., 1999). Origin software was used to perform statistical analyses. A Pearson's test was conducted to evaluate correlations between the morphometric traits. The normality of trait distribution was analyzed by the Shapiro Wilk test. The Kruskal-Wallis test was used to determine the effect of the genotype on the sheep body traits. The relationship between individual and combined genotypes and morphometric traits were analyzed. The general linear model (GLM) procedure of Minitab (MINITAB, USA, v17.1.0) was used for the assessment of the genotype effect on the morphometric traits.

The general linear model was:

$Yij = \mu + Gi + eij$

Where Yij = observed value; μ = overall mean for each trait; Gi = fixed effect of the genotypes and eij = random error.

Results and Discussions

The molecular analysis revealed the presence of two alleles A and G and three different genotypes. In Table 1 are shown the genotype and allele frequencies in the Albanian sheep population as well as other genetic parameters for the *FABP4* marker under study. The observed and expected heterozygosity values were 0.471 and 0.414 respectively. The fixation index was negative indicating a moderate level of outbreeding. The negative values of the inbreeding coefficient can be explained with the small effective population size and with the flock managements by the farmer where one or two rams are used for breeding (Hoda and Marsan 2012). The population was in Hardy Weinberg Equilibrium for the *FABP4* gene. The values of Ne ranged from 1.014 to 1.95. PIC values were 0.328 therefore this marker were considered as moderately informative (Botstein et al., 1980).

Table 1. Genetic parameters for *FABP4* gene in the Albanian sheep population

Alleles	Allelic Frequencies	Genotype	Frequency	Ho	He	Ne	I	F	PIC
A	0.707	AA	0.472	0.471	0.414	1.707	0.605	-0.138	
G	0.293	GG	0.058						0.328
		AG	0.472						

Na = No. of Different Alleles; Ne = No. of Effective Alleles; I = Shannon's Information Index; Ho = Observed Heterozygosity; He = Expected Heterozygosity; F = Fixation Index

The total phenotypic correlations among all morphometric measures for all animals are presented in Table 2. The correlation analyses indicated that morphometric traits were significantly positively correlated ($p<0.001$) with each other (Table 2). These findings are in concordance with those reported previously for the Bardhoka sheep breed (Hoda and Hajno 2021).There is a complete correlation between CD and CC. The strongest correlations are between CC and BW (0.948) and the smallest correlations are between RW and CD, CC (0.236).

The Least Squares Means (LSM) and their respective SE obtained for each trait and each genotype are shown in Table 3. The polymorphism occurring at this marker does not have a significant impact on morphometric traits. The homozygous genotype AA for the *FABP4* gene has a greater body weight compared to other genotypes. This genotype may provide valuable benefits also for other morphometric traits since there were highly positive correlations between these traits. Maybe this mutation of the *FABP4* gene can be used in the breeding program and selection of homozygous individuals for the wild gene due to the increased performance in body weight. Polymorphism of intron 3 of *FABP4* gene is studied also in other species, like Holstein cattle (Ardicli et al., 2019). Shafey et al., (2020) have suggested the associations of SNP found in intron 1 of *FABP4* gene with meat tenderness and intramuscular fat content in Egyptian

sheep: Meanwhile (Xu et al., 2011) have assessed the association of SNP of *FABP4* gene with longissimus thoracic muscle (LT) meat quality traits in sheep. Yan et al., (2018) have reported that variation in *FABP4* affects growth traits and meat production in New Zealand Romney lambs. An association was found between variation in ovine *FABP4* and milk production traits in Greek Sfakia sheep (Ibrahim et al., 2019). Determination of possible associations between gene polymorphisms and carcass characteristics may be useful in improving meat yield and quality (Ardicli et al., 2019). El-Mansy et al. (2019) found association between polymorphism in *FABP4* gene and live performance traits and concluded that applying the marker assisted selection using the *FABP4* gene will be of considerable economic value in sheep production.

Table 2. Correlation matrix of different growth traits in the Albanian sheep breed

	CD	CW	RL	RW	BL	CC	BW
WH	0.639***	0.486***	0.766***	0.242*	0.827***	0.639***	0.773***
CD		0.435***	0.557***	0.236***	0.526***	1***	0.948***
CW			0.516***	0.621*	0.515***	0.435***	0.524***
RL				0.403***	0.741*	0.557*	0.666*
RW					0.287***	0.236***	0.278***
BL						0.526***	0.75***
CC							0.948***

*** ($p < 0.01$); * ($p<0.05$)

Table 3. Relationship between the FABP4 polymorphism with LSM of body measurement in Albanian sheep breed.

Morphometric traits	Symbol	Genotypes			*P* Values
		AA	AG	GG	
Wither height	WH	70.285±0.821	71.181±0.883	68.268±0.457	0.502
Chest depth	CD	36.382±0.764	36.931±0.793	34.541±0.305	0.614
Chest width	CW	19.712±0.738	19.434±0.84	20.4±2.54	0.92
Rump Height	RH	70.623±0.963	72.699±0.948	69.93±1.74	0.265
Rump width	RW	18.332±0.641	18.422±0.625	19.33±1.76	0.893
Body length	BL	72.38±1.17	74.59±1.21	72.83±2.58	0.415
Chest circumference	CC	90.95±1.91	92.33±1.98	86.352±0.762	0.614
Body Weight	BW	56.77±3.04	60.02±3.12	49.75±1.56	0.514

No significant association of the *FABP4* gene and fattening performance and carcass trait was observed on Simmental bulls (Ardicli et al., 2017). The authors (Ardicli et al., 2017) suggested that this might be due to the breed genetic composition or polygenic effects associated with the expression of this trait.

The results indicated no significant effect of this locus on morphometric traits. In addition, the Kruskal_Wallis test showed no significant effect of the genotype on morphometric traits.

In this study, we intended to assess the potential association of SNP detected within intron 3 of the *FABP4* gene with morphometric traits in the Albanian sheep population. The selected gene is shown in literature linked with economic traits in sheep (Khalil, 2020). To our knowledge, this is the first effort to study the association between genotypes and morphometric traits in sheep. This study brings some preliminary data regarding the association of the *FABP4* gene as a candidate marker with morphometric traits. Therefore, further studies are necessary to throw light on possible associations between the marker and phenotypic traits in larger sheep populations.

Ethical Approval

All applicable international, national, and/or institutional guidelines for the care and use of animals followed.

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Conflict of Interest

The authors declare that they have no conflict of interest.

References

Ardicli, S., Dincel, D., Samli, H., & Balci, F. (2017). Effects of polymorphisms at LEP, CAST, CAPN1, GHR, FABP4 and DGAT1 genes on fattening performance and carcass traits in Simmental bulls. *Archives Animal Breeding*, 60(2), 61-70.

Ardicli, S., Samli, H., Vatansever, B., Soyudal, B., Dincel, D., & Balci, F. (2019). Comprehensive assessment of candidate genes associated with fattening performance in Holstein–Friesian bulls. *Archives Animal Breeding*, 62(1), 9-32.

Botstein, D., White, R. L., Skolnick, M., & Davis, R. W. (1980). Construction of a genetic linkage map in man using restriction fragment length polymorphisms. *American Journal of Human Genetics*, 32(3), 314.

Bozgo, V., Hoda, A., Hysi, L., & Papa, L. (2020). Genetic polymorphism detection of Casein genes in Albanian local sheep breeds. *Livestock Research for Rural Development*, 32 (5), Article #71.

El-Mansy, S. A., Peris, S. I. E. M., Ibrahim, A. H. M., & Nasr, A. E. (2019). Genetic variation in the ovine fatty acid binding protein-4 (fabp4) gene and its association with live performance and carcass traits in egyptian ossimi lambs. *Zagazig Journal of Agricultural Research*, 46(6), 2371-2383.

Hoda, A., & Hajno, L. (2021). Body measurements of Bardhoka sheep breed from Albania. *Acta Biologica Turcica*, 34(3), 122-127.

Hoda, A., & Marsan, P. A. (2012). Genetic characterization of Albanian sheep breeds by microsatellite markers. *Analysis of genetic variation in animals*, 3-27. IntechOpen.

Hoda, A., Ajmone-Marsan, P., Dobi, P., & Bozgo, V. (2010). Genetic diversity in Albanian sheep breeds estimated by AFLP markers. *Albanian Journal of Agricultural Sciences*, 9(2), 23-29.

Hoda, A., Bytyqi, H., & Bozgo, V. (2014). Genetic diversity of sheep breeds from Albania and Kosovo revealed by mtDNA. *Journal of Endocytobiosis and Cell Research*, 25, 66-69.

Hoda, A., Bytyqi, H., Dobi, P., & Mehmeti, H. (2009). Genetic diversity of Bardhoka breed in Albania and Kosova analyzed by microsatellite markers. *Research Journal of Agricultural Science*, 41(2), 218-223.

Hoda, A., Hykaj, G., Sena, L., & Delia, E. (2011). Population structure in three Albanian sheep breeds using 36 single nucleotide polymorphisms. *Acta Agriculturae Scand Section A*, 61(1), 12-20.

Hu, Z. L., Park, C. A., & Reecy, J. M. (2019). Building a livestock genetic and genomic information knowledgebase through integrative developments of Animal QTLdb and CorrDB. *Nucleic Acids Research*, 47(D1), D701-D710.

Ibrahim, A. H., Tzanidakis, N., Sotiraki, S., Zhou, H., & Hickford, J. G. (2019). Identification of the association between FABP4 gene polymorphisms and milk production traits in Sfakia sheep. *Archives Animal Breeding*, 62(2), 413-422.

Khalil, M. H. (2020). Molecular applications of candidate genes in genetic improvement programs in livestock. *Egyptian Journal of Animal Production*, 57(Suppl. Issue), 1-23.

Li, X., Ekerljung, M., Lundström, K., & Lundén, A. (2013). Association of polymorphisms at DGAT1, leptin, SCD1, CAPN1 and CAST genes with color, marbling and water holding capacity in meat from beef cattle populations in Sweden. *Meat Science*, 94(2), 153-158.

Shafey, H. I., Mahrous, K. F., Hassan, A. A. M., Rushdi, H. E., & Ibrahim, M. A. El-A. M. (2020). Single-nucleotide polymorphisms in FABP4 gene associated with growth traits in Egyptian sheep. *Veterinary World*, 13(6), 1126.

Xu, Q. L., Tang, G. W., Zhang, Q. L., Huang, Y. K., Liu, Y. X., Quan, K., ... & Zhang, C. X. (2011). The FABP4 gene polymorphism is associated with meat tenderness in three Chinese native sheep breeds. *Czech Journal of Animal Science*, 56(1), 1-6.

Xu, Q. L., Tang, G. W., Zhang, Q. L., Huang, Y. K., Liu, Y. X., Quan, K., ... & Zhang, C. X. (2011). The FABP4 gene polymorphism is associated with meat tenderness in three Chinese native sheep breeds. *Czech Journal of Animal Science*, 56(1), 1-6.

Yan, W., Zhou, H., Hu, J., Luo, Y., & Hickford, J. G. (2018). Variation in the FABP4 gene affects carcass and growth traits in sheep. *Meat Science*, 145, 334-339.

Yeh, F. C., Yang, R. C., & Boyle, T. (1999). POPGENE version 1.32: Microsoft Windows–based freeware for population genetic analysis, quick user guide. *Center for International Forestry Research, University of Alberta, Edmonton, Alberta, Canada*, 1-29.