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Effect of *FecB* Gene on Body Weight in Black Bengal Goat

Vivek Kumar[1*], Nandani Kumari[2], Lal Babu Singh[3] and Subodh Kumar[4]

[1]Regional Institute of Rural Development, Kalakankar, Pratapgarh, U.P., INDIA
[2,3]Department of Animal Breeding & Genetics, Ranchi Veterinary College, Kanke, Ranchi, INDIA
[4]Touring Veterinary Officer, Mau, Tekari, Gaya, Bihar, INDIA

**Corresponding Author: V Kumar; Email:* drvivekkr.vet@gmail.com

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ABSTRACT

FecB gene is first described gene which has been found to increase ovulation rate and litter size in sheep. But, work related to genetic mechanism and genetic markers for caprine proliferation has not been done so much. The present study was aimed to screen Black Bengal goat population for polymorphism of *FecB* gene and to study its effect on body weight at different growth stages. DNA samples from 96 animals were isolated and subjected to PCR. Amplified fragments obtained were allowed for polyacrylamide gel electrophoresis for the detection of single strand conformational polymorphism (SSCP) variants. Among all samples, three different SSCP variants were found which were marked as AA, AB and BB. The highest genotype frequency was observed for AB (0.38), which was followed by BB (0.33) and AA (0.29). Least-square analysis of variance showed significant ($P<0.01$) effect of genotype on body weight at birth only. The least square mean of body weights at birth for genotype BB (01.17±00.03 kg) was significantly lower than that of genotype AA (01.43±00.03 kg) and genotype AB (01.36±00.02 kg). It was also observed that genotype had non-significant effect on body weight at 4-week, 8-week, 12-week, 24-week, 36-week and 48-week of age. These results indicate that effect of *FecB* on body weight was limited at birth only. No significant effects were found on growth rate at later stage of life. *FecB* gene was hitherto linked to prolificacy. So, the effect of *FecB* gene on other traits has important bearing if the research is continued further with more number of species specific primers at other loci.

Keywords: FecB, Black Bengal Goat, PCR-SSCP, body weight

Black Bengal goat is considered one of the important meat breed of India. Increase in reproductive efficiency of this breed can cause increased litter size and not only litter size but growth and development of kids are very important for production of the goat meat (chevon).

The first described gene which affects ovulation rate and proliferation in sheep was *FecB* (Booroola fecundity) gene. It is a single autosomal gene, which increases ovulation rate and litter size in sheep [co-dominant for ovulation rate & partially dominant for litter size (Piper *et al.* 1985; Montgomery *et al.* 1992)]. Piper *et al.* (1985) and Piper and Bindon (1996) found that the effect of *FecB* mutation is additive for ovulation rate and each copy increases ovulation rate by about 1.6. This mutation is characterized by production of large numbers of ovulatory follicles that are smaller in diameter than wild-type follicles (Souza *et al.* 2001). Many aspects of the *FecB* gene affecting litter size, organ development and body mass have been studied by Smith *et al.* (1993) who found that along with additive effect on litter size and ovulation rate, this gene has negative effects on fetal growth and development and body mass during gestation.

Growth or body development along with litter size, are important economic traits in goat breeding as well as reproduction. Having impact on genetic correlation with body weight, identification of variation exists within *FecB* is pertinent. Considering the facts, the present work was designed to investigate the *FecB* gene polymorphism and their effects on body weight at growth stages in Black Bengal Goat.

MATERIALS AND METHODS

A total of 96 blood samples (5 ml each) of Black Bengal goats were collected from jugular vein in vaccutainer tubes under sterile condition. Genomic DNA was isolated and purified from white blood cells using proteinase-K digestion and standard phenol-chloroform extraction earlier described by Sambrook *et al*. (1989). Quality of isolated genomic DNA samples were checked by agarose gel electrophoresis which were visualized under UV trans-illuminator.

In the present study, the primers with the sequence of F: 5'-CCAGAGGACAATAGCAA AGCAAA-3' and R: 5'-CAAGATGTTTTCAT GCCTCATCAACAGGTC-3' were utilized as earlier described by Supakorn *et al.* 2010. Primers were synthesized by Xcelris Lab, Ahmedabad.

Polymerase chain reaction was carried out in 10 µL reaction volume with about 1.5 µl (50-100 µg/µl) of genomic DNA. The reaction mixture consisted of 0.5 µl (10 mM) dNTPs, 0.5 µl (25 mM) $MgCl_2$, 0.5 µl (20 ng/µl) each forward and reverse primer, 0.5 µl (5 units/µl) *Taq* polymerase and 1.5 µl 10X PCR buffer. PCR was conducted with the initial denaturation at 94°C for 3 min, followed by 33 cycles of 94°C for 30 sec, 58°C for 30 sec, 72°C for 30 sec and final extension at 72°C for 5 min. Amplified PCR products were subjected for single strand conformational polymorphism (SSCP) through polyacrylamide gel electrophoresis performed at 4°C for 4 hours at 200 V (Markoff *et al.* 1997). After running was over, gel was kept for silver staining according to Bassam *et al.* (1991). The gel was visualized for banding pattern under light and documented followed by analysis of data using available computer softwares like SPAB.

RESULTS AND DISCUSSION

In the present investigation, PCR-SSCP studies were carried out on BMPR1B gene for detection of the point mutation of *FecB* gene in Black Bengal Goat. Molecular studies assessed the occurrence of polymorphism of *FecB* gene in Black Bengal goat population studied. Gupta *et al*. (2005) reported that the PCR-SSCP method is one of the best method employed in detection of SNPs and nucleotide base change.

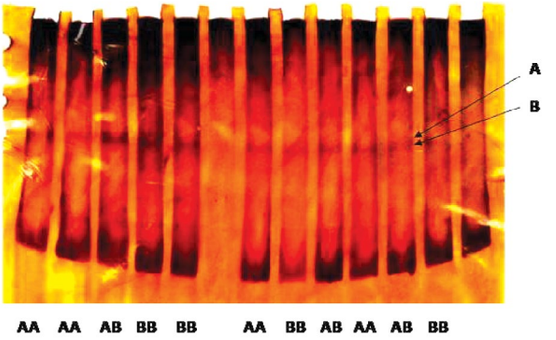

Fig. 1. Different genotypic pattern of FecB gene

Three different SSCP variants were found which were designated as AA, AB and BB (Figure 1). The highest genotype frequency was observed for AB (0.38), which was followed by BB (0.33) and AA (0.29) (Table 1). In concordance to the current study, Polley *et al.* (2009)

Table 2. Least-square means of genotype affecting body weights at birth, 4-week, 8-week, 12-week, 24-week, 36-week and 48-week of age in Black Bengal goats

Genotype	Body weight at						
	Birth	**4-week**	**8-week**	**12-week**	**24-week**	**36-week**	**48-week**
Population mean(µ)	**01.32± 00.02(96)**	**02.37± 00.03(96)**	**03.32± 00.05(96)**	**04.32± 00.06(96)**	**06.42± 00.12(96)**	**08.14± 00.13(96)**	**10.02 ± 00.11(96)**
AA	01.43 ± 00.03[b](28)	02.36 ± 00.06(28)	03.18 ± 00.09(28)	04.17 ± 00.12(28)	06.26 ± 00.23(28)	08.12 ± 00.26(28)	09.91 ± 00.21(28)
AB	01.36 ± 00.02[b](36)	02.37 ± 00.05(36)	03.25 ± 00.07(36)	04.30 ± 00.10(36)	06.35 ± 00.20(36)	08.13 ± 00.21(36)	10.11 ± 00.18(36)
BB	01.17 ± 00.03[a](32)	02.38 ± 00.06(32)	03.53 ± 00.08(32)	04.48 ± 00.11(32)	06.66 ± 00.21(32)	08.16 ± 00.23(32)	10.04 ± 00.19(32)

Means bearing same superscript in a column for a factor did not differ significantly with each other.

Figures in parentheses are number of observations.

worked upon BMPR1B gene (*FecB*) polymorphism in Black Bengal Goat and reported three types of genotype in *FecB* region. Similarly, Chu *et al*. (2010) also found polymorphism of BMPR1B gene and obtained three types of genotypes in Jining Grey goats.

Table 1. *Allelic and genotype frequencies of Black Bengal goat for FecB* gene

Gene	No. of Animal	Allele frequency		Genotype frequency		
		A	B	AA	AB	BB
FecB	96	0.48	0.52	0.29	0.38	0.33

Least-square analysis (Harvey W.R. 1990) of variance showed significant (P<0.01) effect of genotype on body weight at birth. The least square mean of body weights at birth for genotype BB (01.17±00.03 kg) was significantly lower than that of genotype AA (01.43±00.03 kg) and genotype AB (01.36±00.02 kg) (Table 2). Similar findings of Smith *et al.* (1993) supported this fact that body weights were lighter (P<0.05) at most gestational ages in BB/B+ (homozygous carrier/heterozygous carrier) than in ++ (homozygous non-carrier) fetuses. Walling *et al.* (2000) reported that animal carrying a Booroola allele were lighter than non-carrier and observed that Booroola gene is closely linked to a locus affecting early growth and hypothesized that 80% of animals inheriting Booroola allele also inherit the low growth allele. These findings are in consonance to the present results.

However, in contrast to the current study, Visscher *et al.* (2000) found that carriers and non-carriers had same initial and end weights. Also, Kolte *et al.* (2005) observed no significant effect of *FecB* on live weights in any age group of Garole sheep.

It was also observed that genotype had non-significant effect on body weight at 4-week, 8-week, 12-week, 24-week, 36-week and 48-week of age. Important reasons that can be attributed to the effect being non-significant at later ages are that it might be due to small sample size and because at birth, genetic factors have greater effect including *FecB* gene while afterwards, other factors also contribute to the growth traits including management and nutrition.

CONCLUSION

Present findings suggest that *FecB* gene has negative effect on body weight at birth only. At later stages of life, it does not show any effect on growth rate or body development and present research may be continued further on larger sample size and in different species of goat for more conclusive study on effects of *FecB* gene on different growth traits which may be used for marker assisted selection also.

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