Research Article

Monomorphic pattern for *BMPR1B* gene in prolific Alpine x Beetal and Saanen x Beetal goats

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ABSTRACT

The present study was aimed to delineate genetic variability of Bone Morphogenetic Protein Receptor type 1B (*BMPR1B*) or *FecB* gene in Alpine x Beetal and Saanen x Beetal crossbred goats maintained at ICAR-National Dairy Research Institute, Karnal, Haryana. The mutation present at (Q249R) in *BMPR1B* gene induces early maturity of ovarian follicles by increasing the sensitivity of the follicles to FSH leading to release of more eggs per cycle which increases fecundity and forms the basis of marker assisted selection. Genomic DNA was extracted from blood samples of 88 animals from both the breeds and one set of primers were used to amplify a 140bp fragment of *FecB* gene. The amplified fragments were digested using Ava II enzyme. All the animals under study were found to be non-carriers for *FecB* mutation evidenced by the absence of any polymorphism observed after PCR-RFLP suggesting no apparent association of this mutation with fecundity in Alpine x Beetal and Saanen x Beetal crossbred goats.

Key words: *BMPR1B*, Mutation, PCR-RFLP, Prolificacy

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INTRODUCTION

In India, goat can be visualised as an aid to improving livelihood of small and marginal farmers. Goat also known as "Poor Man's cow" has proved to be the most reliable source of income for livestock rearing community. Increment in goat population by 10.14% (Livestock Census, 2019) in last 10 years can explain the increased demand of goat rearing primarily for meat, milk and fibre. In order to improve meat production, prolificacy is an important factor to count upon. Bone Morphogenetic protein receptor type 1B (*BMPR1B*) gene or *FecB* gene is one of the major genes responsible for prolificacy in case of sheep. Studies have revealed that the effect of *FecB* is due to the mutation present in *BMPR1B* gene (Mulsant *et al.,* 2001; Souza *et al.,* 2001 and Wilson *et al.,* 2001). It is believed that the origin of this mutation is probably from the Garole sheep which carries *FecB* mutation (Davis *et al.,* 2002). This mutation became the basis of marker assisted selection for development of sheep breed Avishaan. Till date, many Indian sheep breeds like Kendrapada (Kumar *et al.,* 2008), Bonpala (Roy *et al.,* 2011) and Nilgiri (Sudhakar *et al.,* 2013) have shown positive association of fecundity with *FecB* mutation. In case of goats, *BMPR1B* gene is present in chromosome number 6 with 448166 bp in length. This gene is autosomal with codominant expression, additive for ovulation rate associated with mutation present at (Q249R) or (A746G). Although some studies were being conducted in goats, success rate seem to be limited. Both the crossbreds Alpine x Beetal (AxB) and Saanen x Beetal (SxB) are prolific and the reason behind their prolificacy is not known yet. So, the aim of the study was to identify genetic variability in *FecB* gene and its association with prolificacy in AxB and SxB crossbred goats.

MATERIALS AND METHODS

88 blood samples were collected (A x B and S x B combined) followed by isolation of DNA using Qiagen mini kit protocol. Quality and quantity check of the DNA was done subsequently by Nanodrop Spectrophotometer. DNA amplification by PCR was done afterwards followed by Restriction Enzyme digestion of PCR products.

Polymerase Chain Reaction (PCR)

The PCR amplification was performed using Verti 96 Well Thermal cycler (Applied Biosystems) with simple programme comprising of 95°C for 5 min, 30 cycles of 95°C for 30 sec, annealing temperature of 60ºC for 30 sec, 72°C for 30 sec and final cycle at 72°C for 5 min with a final volume of 25µl (3µl DNA + 22µl PCR master mix). The primer sequences are given in Table 1. The optimum level of primer concentration used for amplification was 10 pmol/µl. PCR products were later checked in 2% agarose gel at 70-90 V for 30 minutes with molecular marker of 100bp as observed in Fig. 1.

Restriction enzyme digestion

Polymerase Chain Reaction-Restriction Fragment Length Polymorphism (PCR-RFLP) analysis was carried out using *Ava II* enzyme for the present study. After preparation of reaction mixture, it was incubated at

Table 1: Details of primers used for amplification of *BMPR1B* gene

Gene	Primer	Primer Sequence	Product Size
BMPR1B	Forward Primer	5'-GTCGCTATGGGGAAGTTTGGATG-3'	140bp
	Reverse Primer	5'-CAAGATGTTTTCATGCCTCATCAACACGGTC-3'	

37ºC for 12 hours. The Restriction Enzyme digested product was then checked in 3% agarose gel and visualised using Gel Documentation System (Fig. 2).

Fig. 1: *PCR product visualised on 2% agarose gel*

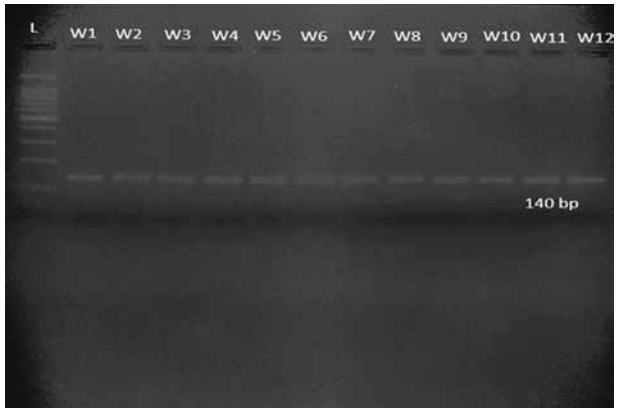

Fig. 2: *Restriction enzyme digested product visualised on 3% agarose gel*

RESULTS AND DISCUSSION

A clear single band of 140 bp fragment of *BMPR1* gene was observed for all the samples in the study (Fig 1). However, PCR-RFLP using *Ava II* revealed a monomorphic pattern of *FecB* gene in A x B and S x B goats. The results of visible bands showed an absence of the enzyme restriction site. Therefore, the genotypes of all the 88 samples were wild type **(++)** which indicated none of them carried *FecB* mutation in the *BMPR1B* gene (Fig 2). This suggested that the prolificacy in Alpine x Beetal and Saanen x Beetal crossbred goats was not linked to *BMPR1B* gene and there might be some other reasons which could explain the prolificacy in these goat breeds.

The results of this study were in agreement with the works of (Hua *et al.,* 2008; Chu *et al.,* 2010; Dutta *et al.,* 2014 and Palai *et al.,* 2013) in different goat breeds. Although two novel mutations in exon 8 of *BMPR1B* gene (775A > G and 777G > A) were observed but there was absence of *FecB* mutation in *BMPR1B* gene in case of Markhoz Goat.

CONCLUSION

Monomorphic patterns observed for studied mutation in both flocks suggests absence of *FecB* polymorphism. So, it can be concluded that the *FecB* mutation plays no role in prolificacy of Alpine x Beetal and Saanen x Beetal goats.

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