RESEARCH ARTICLE

Association Study of Fecundity Gene BMPR1B with Prolificacy in Surti Goats under Farm and Field Condition

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Abstract

The Surti is a dual purpose goat breed of Gujarat. The bone morphogenetic protein receptor type 1B (BMPR1B) gene of transforming growth factor beta (TGF-β) superfamily ligands is playing a role in ovulation as well as litter size. Mutation in Exon-6 region of BMPR1B gene with base size 190 bp reported increasing litter size. Based on the known mutation information in goat and sheep, PCR primers were designed to screen polymorphism in total 100 Surti goats, 50 Surti goats from University Farm, Navsari and 50 Surti goats from field units of Southern part of Gujarat. During PCR-RFLP study no polymorphic sites were found for Exon-6 region of BMPR1B on Surti goats. Moreover, the twinning rate was 10% in first parity and higher in subsequent second (62.5%) and third (76.8%) parties.

Keywords: Fecundity gene, Polymorphism, BMPR1B, PCR-RFLP, Surti goat.

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Introduction

The Surti breed of goat is known to be a good dairy and meat type breed, especially suited for maintenance under complete confinement and stall-feeding. The breeding tract of this breed is mainly from Vadodara to Valsad districts in the southern part of Gujarat. The landless farmers rear it for income from sale of milk and animals. Identifying and using genes associated with the litter size for future selection procedure are very much important for genetic improvement and fecundity of goat. Studies on the inheritance pattern of ovulation rate and litter size in prolific sheep led to identification of a major gene responsible for prolificacy. Therefore, the identification of the genes responsible for the prolificacy in goats, known as fecundity genes, is of importance to goat farming (Sharma *et al.,* 2016). The bone morphogenetic protein receptor type 1B (BMPR1B) gene also known as FecB gene is one of the genes playing a major role in increasing ovulation rate and litter size. The aim of this study was, therefore, to identify polymorphic sites, if any, in exon-6 region of BMPR1B in Surti goats.

Materials and methods

A total of 100 Surti goats half from field and half from the farm were selected for the study. Records related with kidding and kids born per kidding were collected from farm register in case of 50 goats of LRS, NAU, Navsari and by field survey in case of 50 fields Surti goats. To genotype the Surti goats, blood samples (5 mL) from all 100 goats were collected from the jugular vein in sterilized BD vacutainers and were transported to the laboratory at 4°C.

The DNA was extracted from blood as per the standard phenol-chloroform DNA extraction procedure described by Sambrook and Russell (2001). Quality of extracted DNA was checked using 0.8% agarose gel at 80 volts for 2 hours.

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The quantity of DNA was measured using Nano-Drop Spectrophotometer of thermo fisher. Amplification of DNA by PCR was done by using BMPR1B gene-specific primers given in Table 1. PCR reaction was performed using 20 µL total reaction mixture which included 10.0 µl master mix, 0.8 µl forward primer, 0.8 µL reverse primer, 0.3 µl DNA and 5.4 µl Mili-Q-water. PCR protocol was carried out to amplify a specific region of the gene with initial denaturation at 94°C for 1 min followed by 94°C for 45 sec, annealing at 57°C for 1 min and 72°C for 1 min for 35 cycles and final extension at 72°C for 1 min. After amplification, the PCR product was again checked using 2% agarose gel for amplification of BMPR1B gene-specific region and to confirm the size of the amplified region.

Amplified PCR product was digested using restriction enzyme (RE) *AvaII* (Table 1). The digestion mixture was prepared using 0.3 µl RE, 5 µL PCR product, 1.5 µL buffer, and 8.2 µL Mili-Q-water. The digestion mixture was kept at 60°C for 15 minutes in a water bath and subjected to run on

Table 1: The primer pairs, expected product size and restriction enzyme of the BMPR1B gene

Candidate Gene	*Region of Gene*	*Primers (5′ – 3′)*	*Product Size*	*Restriction Enzyme*
BMPR1B	Exon 6	F-5′- CAGAGGACAATAGCAAAGCAAA–3′ R-5′- CAAGATGTTTTCATGCCTCATCAACACGGTC-3′	190 bp	AvaII

Table 2: Parity wise classification, total kids born up to specific parity and twining percentage in Surti Goat

	Parity and kids born					
Total No. of animal	*Parity 1*	*Kids*	*Up to parity 2*	Kids	Up to parity 3	Kids
50 (LRS, NAU)	8	9	7	24	35	152
50 (Field)	2	2	1	3	47	157
Total	10	11	8	27	82	309
Twining	10%		62.5%		76.8%	

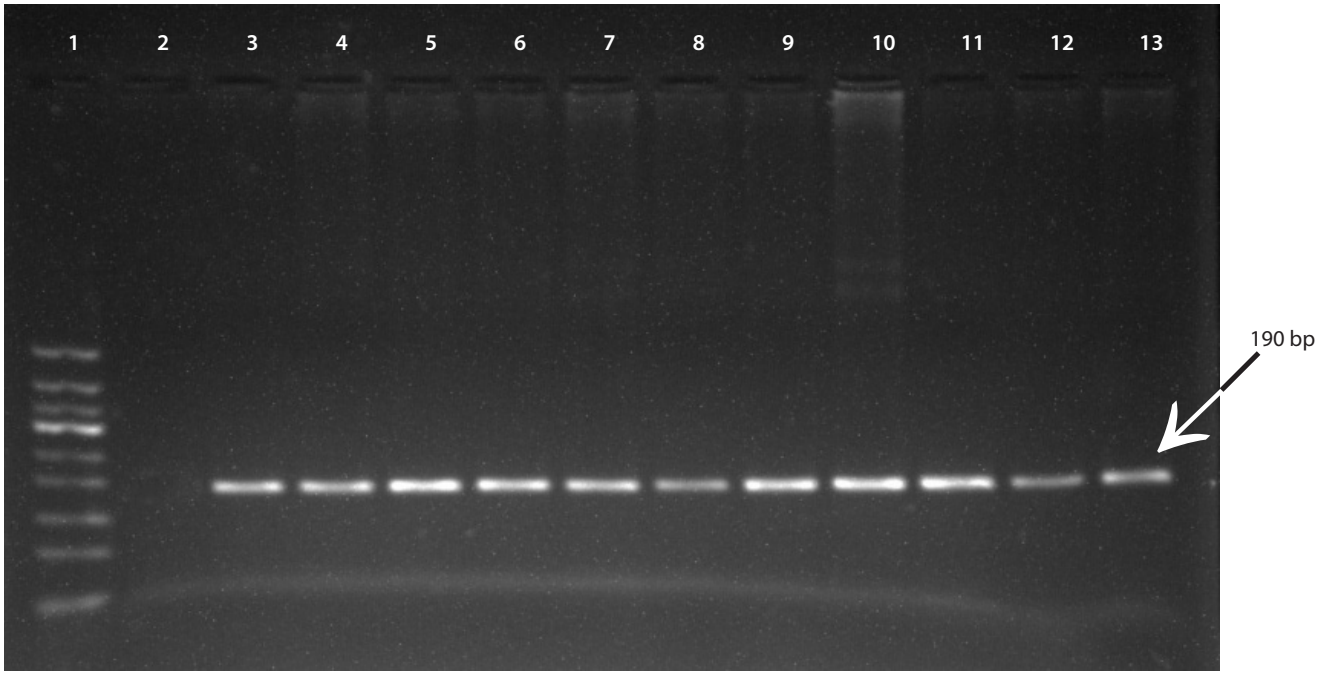

Fig. 1: Agarose gel electrophoresis (2%) of allele specific BMPR1B gene for PCR product. Lane 1: DNA molecular weight marker (50 bp DNA Ladder) Lanes 2: PCR without genomic DNA (Template negative control). Lane 3-13: PCR amplified product of BMPR1B gene (size 190 bp)

2% agarose gel electrophoresis at 80 volts for 2 hours and image was captured using gel doc system.

Results and discussion

Surti goat with first parity is less likely to produce twins as only 10% twining rate was observed in the present study, while during second and third parties the twining percentage were found very high as 62.5% and 76.8%, respectively (Table 2). Some of the second and third parity animals have delivered twins during their first parity also. Animals under the present study had also delivered triplets which were considered in animals having twins but an actual number of kids presented in the table. This is the normal tendency found in Surti goat that in first parity mostly produce single kid and twining start after second parity and continue for subsequent parties. Lesser twining rate as compared to present study was reported by Kuralkar *et al*. (2013) as 57% in Berari goats, while higher twining rates of 69.50% and 64.81% were observed in the same breed by Sharma *et al*. (2016) and Panhale *et al*. (2018).

Genotyping of Surti goat was done by a collection of blood samples from each individual goat under study and DNA was extracted. Extracted DNA was checked for quality (Fig. 1) and quantity. The PCR product size of BMPR1B gene was found as 190 bp in the present study as reported by others.

Restriction digestion of BMPR1B gene PCR product was done using AvaII RE and no polymorphism was found in Surti goats (Fig. 2). The restriction digestion patterns found in the present study were an undigested or intact band of exon-6 region of BMPR1B gene with 190 bp size. A similar finding was reported by Palai *et al*. (2013) in the prolific Raighar

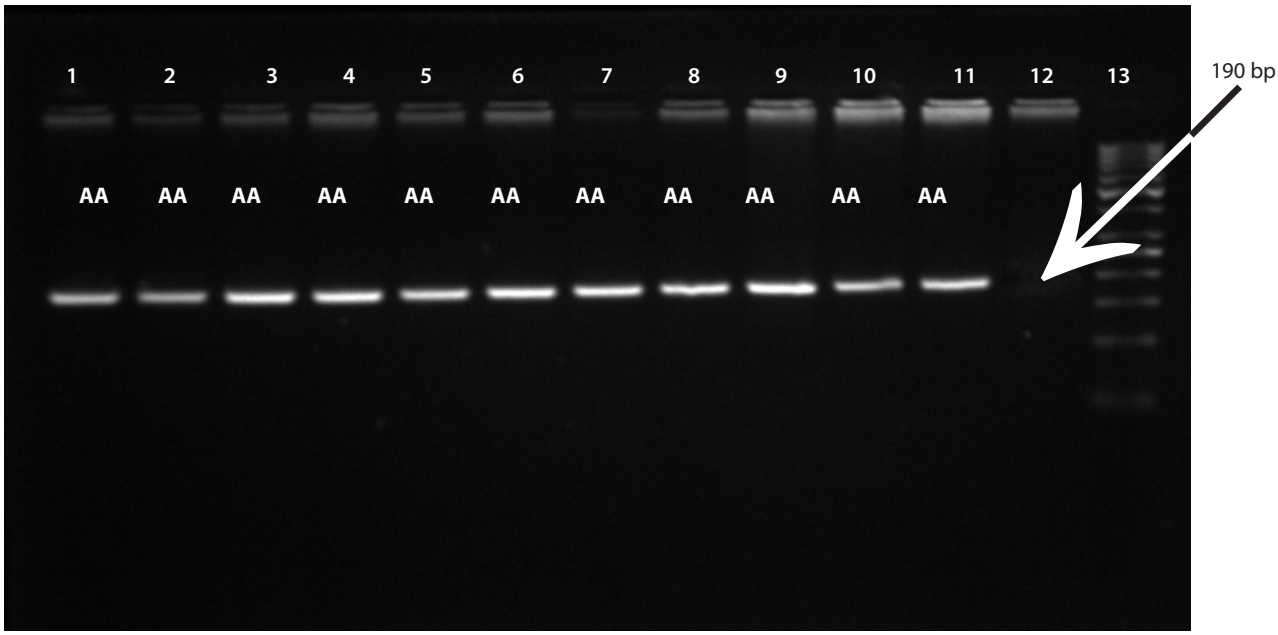

Fig. 2: Restriction pattern of PCR-RFLP product for BMPR1B gene. Lane 1-11: Digested PCR product of BMPR1B gene. Lanes 12: PCR without genomic DNA (Template negative control). Lane 13: DNA molecular weight marker (50 bp DNA Ladder).

Table 3: Genotype wise average kids born in Surti Goat

Particulars	*AA Genotype (50 Goats of LRS, NAU, Navsari)*	*AA Genotype (50 Goats of Field of South Gujarat)*	*Total of 100 goats*
Parity 1	8	2	10
Kids	9	2	11
Parity 2	7	1	8
Kids	24	3	27
Parity 3	35	47	82
Kids	152	157	309
Total Animals	50	50	100
Total Parity	128	154	282
Total Kids Born	185	162	347
Avg. Kids Born/Parity	1.45	1.05	1.23
Avg. Kids Born/Ani.	3.70	3.24	3.47

goats. However, Polley *et al*. (2009) reported the presence of mutant type (G) nucleotide in Black Bengal goats. The wild homozygotes for BMPR-1B on AvaII RE digestion are also reported in goat breeds of China viz., Boer, Huangghuai, Haimen, Nubi and Matou (Hua *et al.,* 2007) and the Rayini goats of Iran (Gazooei *et al.,* 2013).

Out of total 100 Surti goats, all female goats were found with AA genotype. Association study with kidding rate could not be made due to the monomorphic pattern of gene.

From the present study, it was concluded that BMPR1B gene is monomorphic in female Surti goat. In Surti goat twinning rate is lesser in first parity and higher in subsequent parity. There is no association between Exon-6 region of BMPR1B and kidding rate in Surti goat as it is monomorphic.

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