

tively (Figure 1b). Further, *A. cf. culicifacies* 'Bluchistan' and *A. dthali* were found to have shared substitution homogeneity that was different from the members of *A. culicifacies* species complex, while all the five members of the sibling species complex shared similar substitution pattern. The results, thus, confirmed that *A. cf. culicifacies* 'Bluchistan' is close to *A. dthali*, than to any member of the *A. culicifacies* species complex.

In conclusion, the DNA sequence analyses of the ribosomal ITS2 region clearly revealed that *A. cf. culicifacies* 'Bluchistan' and *A. dthali* are the same species. Our conclusion is further substantiated by the fact that *A. dthali* is widely distributed in southern parts of Iran including Baluchistan. Current developments in genome analysis and barcode genes have further facilitated the identification of new species in addition to existing methods such as morphological, cytological, isozyme based methods. However, misidentification of disease vectors sometimes leads to the suggestion of wrong vector control strategies. This study, thus, highlights the importance of accurate morphological identification of field collected specimens before applying modern molecular and computational phylogenetic techniques to establish the taxonomic relationships.

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## Increasing prolificacy and ewe efficiency in sheep through *FecB* gene introgression

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The *FecB* gene of Garole (G) was introgressed into non-prolific Malpura (M) sheep and the performance of the GM half-breds is being monitored. DNA samples of Garole, Malpura and Garole × Malpura (GM) crossbreds were screened by PCR-RFLP to determine the presence of *FecB* mutation. The majority of Garole (96%) and GM crossbred (72%) were carriers (BB and B+) for the *FecB* mutation. The *FecB<sup>BB</sup>* and *FecB<sup>B+</sup>* carrier ewes resulted in 81.19 and 69.31% higher prolificacy respectively, as compared to non-carrier Malpura ewes. The viable benefits accrued by

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**gene infusion are reflected by 45.7% increase in the prolificacy and 35.05% in weaning rate of GM compared to that of Malpura. A single mutated copy of *FecB* gene produced 0.70 and weaned 0.58 extra lambs in GM sheep. Likewise, the ewe productivity efficiency and ewe efficiency in terms of lambs' weight produced per kg of ewes' body weight of *FecB* carrier GM ewes showed upward trend during the growth phase as compared to non-carriers. The *FecB* carrier twin lamb bearing ewes weaned 78.38% more lambs compared to non-carrier ewes. The DNA test should be applied in routine as a marker for identifying *FecB* gene in crosses with Garole sheep.**

**Keywords:** *FecB* gene, Garole × Malpura, PCR–RFLP test, prolificacy, sheep.

INDIA is endowed with a wide diversity of sheep genetic resources, which forms the backbone of its rural livelihood security systems. Sheep rearing now faces a dilemma to produce more mutton and wool for the growing human population against the realization of shrinking grazing resources and saturation in further growth of the sheep population. In the present scenario, the demand for meat in India has increased rapidly and the emphasis has shifted from wool towards mutton as the main produce from sheep rearing. There is an acute shortage of meat for domestic needs besides huge demand in the international market. The gap between the demand and production of mutton is to be bridged by augmenting the reproductive rate of low producing Indian sheep breeds. The low prolificacy of all indigenous sheep breeds, barring Garole, is a stumbling block for converting the available bio-resources into more remunerative economic wealth on sustainable basis to cope with the rising demand of end users. In order to improve the fecundity of sheep, incorporation of genetic material of prolific sheep is an ideal approach to evolve a large size breed capable of multiple births for economic and remunerative mutton production.

The Booroola fecundity gene (*FecB*) is a dominant autosomal gene in sheep located on the 6th autosomal chromosome and is responsible for increasing the ovulation rate and in turn, prolificacy. The effect of *FecB* is additive for ovulation rate and each copy of the allele increases ovulation rate by about 1.6% and approximately one to two extra lambs in Booroola Merino<sup>1,2</sup>. The high prolificacy in Booroola Merino is the result of a non-conservative mutation (Q249R) in a highly conserved intracellular kinase-signalling domain of the bone morphogenetic protein receptor-1B (BMPRI1B), located in the region containing the *FecB* locus<sup>3–5</sup>. The DNA test has enabled the origin of the evolved Booroola Merino to be traced back to the Garole sheep that were introduced to Australia in the late 18th century from Bengal<sup>6</sup>. Considering the importance of prolific genes, a cross breeding scheme was initiated at the Central Sheep and Wool Research Institute, Avikanagar, Rajasthan to introgress

*FecB* gene possessed by India's most valuable germplasm 'Garole sheep' of hot and humid environment into the non-prolific and best adapted large size mutton sheep breed 'Malpura' of semi-arid tropical environment to produce the Garole × Malpura (GM) crossbred carrying *FecB* gene.

Garole is an exceptionally prolific breed amongst all other non-prolific sheep breeds in India. It is a rare micro-sheep known for its high prolificacy<sup>6–8</sup> and the ability to thrive well under adverse climate. The average adult body weight of this breed ranges between 10 and 14 kg with mean litter size 2.27 in its native habitat<sup>7,9–11</sup>.

On the other hand, the Malpura is a popular mutton breed of Rajasthan and is known for its hardiness and adaptability to the local environment but prolificacy is low and usually produces single lambs. The average adult body weight of Malpura ranges from 30 to 40 kg with mean litter size of 1.05 and 4.71% twinning rate<sup>12–14</sup>. The objective of the present study is to evaluate the effect of crossing on prolificacy and ewe efficiency of newly evolved *FecB* carrier GM crossbred.

In the breeding experiment, Garole rams (*FecB*<sup>BB</sup> or *FecB*<sup>B+</sup>) were mated to wild type homozygous (*FecB*<sup>++</sup>) Malpura ewes to produce F<sub>1</sub> and then *inter se* mating was used to produce F<sub>2</sub> and F<sub>3</sub> generations. Simultaneously, *FecB* status of animals was tested. Blood samples of 90 Garole, 62 Malpura and 301 GM half-breds were randomly collected in anticoagulant-coated tubes. DNA was extracted from white blood cells using standard phenol–chloroform extraction method with minor modifications<sup>15</sup>. The *FecB* genotyping was carried out by forced PCR–RFLP technique using F-12 and R-15 primers<sup>5</sup>. The PCR reaction mixture comprised as follows: 1x PCR buffer (containing (NH<sub>4</sub>)<sub>2</sub>SO<sub>4</sub>), 1.5 mM MgCl<sub>2</sub>, 200 μM dNTP mix, 0.5 U *Taq* DNA polymerase (MBI Fermentas), 0.200 μM of each primer and 100 ng of template DNA in 20 μl reaction volume. The amplification reaction conditions were as follows: initial denaturation at 94°C for 1 min, followed by 94°C for 15 s, 60°C for 30 s, 72°C for 30 s for 35 cycles and final extension at 72°C for 5 min, followed by heat denaturation at 99°C for 15 min. The primer pair amplified a 140 bp region of the BMPRI1B gene. The 5 μl PCR product was digested with *Ava*II restriction enzyme (MBI, Fermentas) at 37°C for 2 h and run in 3.0% agarose gel.

All the sheep were raised under semi-intensive management system and provided with similar grazing/feeding conditions. An extra allowance of concentrate at the rate of 250 g/ewe/d was supplemented under group feeding for three months every year, i.e. from last month of pregnancy to completion of second month of lactation in addition to the 8–10 h grazing in a field interspersed with seasonal shrubs and forbs. The diet was supplemented with minerals and vitamins. The lambs were allowed to suckle their dams from birth until weaning at 3 months of age. At the sheds, lambs were fed tree/pala

leaves, cowpea fodder and concentrate mixture ad-libitum from 20 days of weaning. In addition to grazing throughout the day on natural vegetation, 250 g of a concentrate mixture was provided throughout the post-weaning period (3–12 months of age). All the prophylactic measures against various sheep diseases were carried out as per prescribed health calendar of the Institute in addition to curative treatment of sick animals, as and when required.

In the present study data on reproduction and production traits for *FecB* carrier ( $FecB^{BB}$  or  $FecB^{B+}$ ) and non-carrier ( $FecB^{++}$ ) GM sheep from 2000 to 2006 were utilized (Table 2). The reproductive traits studied were prolificacy (number of live lambs born/ewes lambed), weaning rate (WR = number of lambs weaned/ewes lambed) and litter size at marketing or 6-month age (LS6: number of lambs alive up to 6 months/ewes lambed). The ewe productivity efficiency (EPE) was calculated as the sum total of lambs' weight produced at birth, weaning, 6 and 12 months of age/ewes lambed. The ewes' efficiency (EE) in terms of weight of lambs produced per kg of ewe body weight was also calculated at birth, weaning 6 or 12 months of age. The ewe body weights after lambing were used for calculation of EE. The data on EPE and EE were classified according to *FecB* genotype of ewes and among *FecB* genotypes; 4 groups were made, viz. *FecB* carrier single lamb bearing ewes (*FecB* CSL); *FecB* carrier twin lamb bearing ewes (*FecB* CTL); *FecB* carrier triplet lamb bearing ewes (*FecB* CTRL) and non-carriers. The data were analysed by analysis of variance (ANOVA) using the general linear model procedure of SPSS 14.0 for Windows XP. The data on EPE and EE were classified according to *FecB* genotype, group, generation, parity and year of lambing and taken as fixed effects in model at the same time. Ewe weight was included as covariate in the statistical model to adjust for the wide differences in the ewe body weights. The least squares procedure<sup>16</sup> was used to analyse the data. The *FecB* genotype  $\times$  year interaction and *FecB* genotype  $\times$  parity interaction were non-significant for all the EPE traits studied except significant ( $P < 0.05$ ) effect of *FecB* genotype  $\times$  year interaction on EPE at birth. The ewes lambed only once in a year and the major breeding and lambing season of ewes was autumn (August–September) and spring (January–March) respectively; hence season was not included in the model.

The *FecB* was detected in 96% Garole sheep and 72% GM crosses (Table 1). Presence of the *FecB* mutant allele could not be detected in Malpura animals. In Garole carriers, the *FecB* allele was present in 74 (82%) and 13 (14%) animals in homozygous and heterozygous state respectively. Davis *et al.*<sup>6</sup> also reported that *FecB* is present at a high frequency in Garole sheep. In GM crosses, *FecB* gene was detected in 13% in homozygous (BB) state and 59% animals in heterozygous (B+) state. The results indicated that most of the GM individuals (72%) carried the *FecB* mutation, and genotypic frequency of  $FecB^{B+}$  was

0.59. These results are in close agreement with the earlier report that 68% GM individuals had *FecB* allele<sup>17</sup>. As the *FecB* allele has been fixed in the Garole population and is absent in Malpura, the observed allele frequency in GM sheep is close to the expected frequency of 50%. In GM, *FecB* gene is present in heterozygous state ( $FecB^{BB}$ ) in  $F_1$  and then segregates in  $F_2$ ,  $F_3$  and  $F_4$  after interbreeding among half-breds. The *FecB* mutation was also found in Hu and Han breeds from China and the 12 Hu sheep sampled were all homozygous carriers of *FecB* ( $FecB^{BB}$ ) whereas the sample of 12 Han sheep included all the three genotypes ( $FecB^{BB}$ ,  $FecB^{B+}$ ,  $FecB^{++}$ )<sup>18</sup>.

The *FecB* genotype significantly affects prolificacy, WR and LS6. The GM ewes of all genotypes had 45.7% (1.53 vs 1.05), 35.05% (1.312 vs 0.97) and 29.47% (1.23 vs 0.95) advantages over Malpura for these traits respectively (Table 3). The single and two-copy mutation of the *FecB* allele resulted in 1.71 and 1.83 prolificacy, and 1.46 and 1.42 WR in GM ewes respectively. The single copy effect (0.70 extra lambs) on prolificacy was similar to the Garole  $\times$  Deccani or Garole  $\times$  Bannur crosses<sup>19</sup> and Merino ewes<sup>1</sup>. The second copy mutation increases prolificacy by about 0.12 lambs, whereas in Garole it was reported as 0.18 (ref. 18). The result clearly indicates that a second copy mutation has a partially dominant effect on prolificacy. The prolificacy, WR and LS6 of *FecB* CTL and *FecB* CTRL were higher than non-carriers. The WR and

**Table 1.** *FecB* genotypes (in numbers) and genotypic frequencies in Garole, Malpura and GM sheep

Genetic group	Genotypes/genotypic frequencies			Total
	$FecB^{BB}$	$FecB^{B+}$	$FecB^{++}$	
Garole	74 (0.82)	13 (0.14)	3 (0.03)	90
Malpura	–	–	62 (100)	62
GM				
F <sub>1</sub>	00 (0.00)	59 (0.84)	11 (0.16)	70
F <sub>2</sub>	21 (0.14)	80 (0.52)	52 (0.34)	153
F <sub>3</sub>	17 (0.24)	35 (0.49)	19 (0.27)	71
F <sub>4</sub>	2 (0.29)	3 (0.43)	2 (0.28)	7
Total	40 (0.13)	177 (0.59)	84 (0.28)	301

Figures in parenthesis are genotypic frequencies.

**Table 2.** Number of GM ewes lambed during different years

Year	Genotypes			Total
	$FecB^{B+}$	$FecB^{++}$	$FecB^{BB}$	
2000	06	02	00	08
2001	11	05	00	16
2002	17	07	00	24
2003	37	09	02	48
2004	41	10	03	54
2005	39	21	03	63
2006	36	15	04	55
Total	187	69	12	268

**Table 3.** Prolificacy, weaning rate and litter size at 6 months age among *FecB* carrier and non-carrier ewes

	<i>N</i>	Prolificacy	Weaning rate	Litter size at 6 months
Overall	268	1.53 ± 0.04	1.31 ± 0.04	1.23 ± 0.04
<i>FecB</i> Genotype		**	**	**
<i>FecB</i> <sup>BB</sup>	12	1.83 <sup>a</sup> ± 0.21	1.42 <sup>a</sup> ± 0.23	1.33 <sup>a</sup> ± 0.19
<i>FecB</i> <sup>B+</sup>	187	1.71 <sup>a</sup> ± 0.04	1.46 <sup>a</sup> ± 0.05	1.36 <sup>a</sup> ± 0.05
<i>FecB</i> <sup>++</sup>	69	1.01 <sup>b</sup> ± 0.01	0.88 <sup>b</sup> ± 0.04	0.84 <sup>b</sup> ± 0.05
<i>FecB</i> group		**	**	**
<i>FecB</i> CSL	72	1.01 <sup>a</sup> ± 0.01	0.83 <sup>a</sup> ± 0.04	0.78 <sup>a</sup> ± 0.05
<i>FecB</i> CTL	111	1.98 <sup>b</sup> ± 0.01	1.73 <sup>b</sup> ± 0.05	1.63 <sup>b</sup> ± 0.06
<i>FecB</i> CTRL	16	3.00 <sup>b</sup> ± 0.00	2.38 <sup>c</sup> ± 0.18	2.13 <sup>c</sup> ± 0.18
<i>FecB</i> <sup>++</sup>	69	1.01 <sup>a</sup> ± 0.01	0.88 <sup>a</sup> ± 0.05	0.84 <sup>a</sup> ± 0.05
Generation		**	**	**
F <sub>1</sub>	119	1.61 <sup>a</sup> ± 0.05	1.46 <sup>a</sup> ± 0.06	1.40 <sup>a</sup> ± 0.06
F <sub>2</sub>	70	1.63 <sup>a</sup> ± 0.08	1.39 <sup>b</sup> ± 0.09	1.23 <sup>b</sup> ± 0.09
F <sub>3</sub>	69	1.33 <sup>b</sup> ± 0.06	1.04 <sup>c</sup> ± 0.08	0.99 <sup>c</sup> ± 0.08
F <sub>4</sub>	10	1.40 <sup>b</sup> ± 0.22	0.80 <sup>a</sup> ± 0.04	0.80 <sup>d</sup> ± 0.04
Parity		*	**	*
1	120	1.39 <sup>a</sup> ± 0.05	1.11 <sup>a</sup> ± 0.06	1.06 <sup>a</sup> ± 0.06
2	72	1.65 <sup>b</sup> ± 0.07	1.47 <sup>b</sup> ± 0.08	1.36 <sup>b</sup> ± 0.08
3	38	1.68 <sup>bc</sup> ± 0.11	1.58 <sup>b</sup> ± 0.12	1.42 <sup>b</sup> ± 0.18
4	23	1.52 <sup>c</sup> ± 0.14	1.26 <sup>c</sup> ± 0.13	1.26 <sup>c</sup> ± 0.13
5	10	1.70 <sup>d</sup> ± 0.15	1.50 <sup>b</sup> ± 0.22	1.40 <sup>bd</sup> ± 0.27
6	5	1.80 <sup>d</sup> ± 0.34	1.60 <sup>d</sup> ± 0.51	1.40 <sup>bd</sup> ± 0.51

N, Number of ewes lambbed; NS, Non-significant; \**P* < 0.05; \*\**P* < 0.01; *FecB* CSL, *FecB* carrier single lamb bearing ewes; *FecB* CTL, *FecB* carrier twin lamb bearing ewes; *FecB* CTRL, *FecB* carrier triplet lamb bearing ewes, same superscript does not differ significantly.

LS6 of *FecB*<sup>++</sup> were slightly higher than *FecB* CSL. The *FecB* CTL ewes weaned 78.38% more lambs (1.73 vs 0.97) and *FecB* CTRL ewes weaned 145.36% more lambs (2.38 vs 0.97) than non-carrier Malpura sheep. The Finn-Dorset-Targhee sheep had also weaned more lambs per ewe exposed (1.41 vs 1.18) than Targhee and gain in prolificacy was 48% (ref. 20). The mean prolificacy (1.71) of GM heterozygotes was almost similar to the heterozygotes of Javanese sheep that are segregating *FecB*<sup>B</sup> allele<sup>21</sup>. The high prolificacy was reported in Garole (2.23, ref. 22), Hu (2.1, ref. 23), Han (2.4, ref. 23) and Javanese sheep (2.59, ref. 21). The reason of low prolificacy in GM half-breds was due to presence of heterozygous and non-carrier individuals thereby non-fixation of the *FecB* allele in the flock. The other reasons may be the embryonic loss later leading to low litter size, non-expression of Booroola allele, presence of *FecB* carriers that produce single lambs, nutritional and unknown environmental factors or combination of these factors<sup>6</sup>. The prolificacy and WR decreases with generation indicating the segregation of non-carriers (++) after intermating. The prolificacy, WR and LS6 increases with parity. Booroola Merino sired ewes were also more prolific at 2nd and 3rd parity<sup>24</sup>. The effect of year was non-significant for prolificacy and significant for WR and LS6. Differences due to year of lambing were attributed to the variations in environmental conditions and availability of other inputs.

The *FecB* genotype significantly affected EPE and EE traits and B+ ewes weaned more litter weight (12.77 kg)

as compared to BB (11.55 kg) and ++ (10.39 kg) ewes (Table 4). Another study<sup>19</sup> also reported that the body weight of lambs was higher in ewes bearing twin lambs (carriers) as compared to those bearing single lambs (non-carriers). It is well-established that if the number of lambs born increases, the survivability decreases. The pre-weaning survivability of GM lambs born as twins varied from 83.33 to 92.48% and of triplets from 66.67 to 76.19%. The EPE of Malpura ewes were 13.3 and 20.3 kg at weaning and 6 month age respectively<sup>14</sup>. The results indicated that the production of B+ ewes is more beneficial than the BB or ++ ewes for achieving higher EPE and EE. Comparative evaluation of EPE and EE of *FecB* CTL and *FecB* CTRL ewes of GM with the Malpura ewes reveals the significant potential of prolific GM germplasm for enhancing mutton production. The total lamb production from *FecB* carrying GM ewes bearing twins and triplets was more than non-carriers at birth, weaning, 6 and 12 months of age. The difference in kilogram of lamb produced between GM ewe carriers for *FecB* gene bearing multiple birth and ++ ewes constantly increased with age and the *FecB* CTL and *FecB* CTRL ewes produced lambs equal to their body weight at 6 months age (EPE 1.02 and 1.22 kg). The EPE at weaning, 6 and 12 months of *FecB* CSL ewes was slightly lower than *FecB*<sup>++</sup>. The reasons contributed to these differences are one; the fact that *FecB* CSL ewes were born as twins or triplets which are mostly lighter than *FecB*<sup>++</sup> ewes, and second; the *FecB* carriers are lighter than non-carriers<sup>14,17</sup>. Generation and

**Table 4.** Least squares means of ewes' productivity efficiency (EPE) and kg of lambs produced per kg of ewes' body weight (EE) of *FecB* carrier and non-carrier GM ewes

	N	EPE at				EE at			
		Birth	Weaning	6 Months	12 Months	Birth	Weaning	6 Months	12 Months
Overall	268	2.83 ± 0.06	12.10 ± 1.10	18.28 ± 0.64	21.81 ± 0.91	0.12 ± 0.00	0.51 ± 0.02	0.78 ± 0.03	0.93 ± 0.04
<i>FecB</i> Genotype		**	*	**	**	**	**	**	**
<i>FecB</i> <sup>BB</sup>	12	3.03 <sup>a</sup> ± 0.37	11.55 <sup>a</sup> ± 1.89	17.83 <sup>a</sup> ± 3.42	22.53 <sup>a</sup> ± 4.02	0.13 <sup>a</sup> ± 0.01	0.49 <sup>a</sup> ± 0.08	0.78 <sup>a</sup> ± 0.14	0.99 <sup>a</sup> ± 0.17
<i>FecB</i> <sup>B+</sup>	187	2.97 <sup>a</sup> ± 0.72	12.77 <sup>b</sup> ± 0.45	19.61 <sup>a</sup> ± 0.79	24.19 <sup>a</sup> ± 1.12	0.13 <sup>a</sup> ± 0.00	0.56 <sup>b</sup> ± 0.02	0.86 <sup>a</sup> ± 0.04	1.06 <sup>a</sup> ± 0.05
<i>FecB</i> <sup>++</sup>	69	2.42 <sup>c</sup> ± 0.06	10.39 <sup>c</sup> ± 0.61	14.74 <sup>b</sup> ± 0.96	15.22 <sup>c</sup> ± 1.43	0.09 <sup>c</sup> ± 0.00	0.39 <sup>c</sup> ± 0.02	0.56 <sup>c</sup> ± 0.04	0.57 <sup>c</sup> ± 0.06
<i>FecB</i> group		**	**	**	**	**	**	**	**
<i>FecB</i> CSL	73	2.01 <sup>a</sup> ± 0.05	8.26 <sup>a</sup> ± 0.51	11.90 <sup>a</sup> ± 0.81	14.39 <sup>a</sup> ± 1.16	0.09 <sup>a</sup> ± 0.00	0.35 <sup>a</sup> ± 0.02	0.51 <sup>a</sup> ± 0.03	0.62 <sup>a</sup> ± 0.05
<i>FecB</i> CTL	111	3.37 <sup>b</sup> ± 0.07	14.80 <sup>b</sup> ± 0.54	23.11 <sup>b</sup> ± 0.99	29.21 <sup>b</sup> ± 1.45	0.15 <sup>b</sup> ± 0.00	0.65 <sup>b</sup> ± 0.23	1.02 <sup>b</sup> ± 0.43	1.28 <sup>b</sup> ± 0.06
<i>FecB</i> CTRL	16	4.52 <sup>c</sup> ± 0.14	18.08 <sup>c</sup> ± 1.11	28.81 <sup>c</sup> ± 2.07	32.92 <sup>c</sup> ± 3.23	0.19 <sup>c</sup> ± 0.01	0.78 <sup>c</sup> ± 0.06	1.22 <sup>c</sup> ± 0.09	1.39 <sup>c</sup> ± 0.13
<i>FecB</i> <sup>++</sup>	68	2.43 <sup>a</sup> ± 0.06	10.41 <sup>d</sup> ± 0.63	14.76 <sup>d</sup> ± 0.98	15.09 <sup>a</sup> ± 1.45	0.09 <sup>a</sup> ± 0.00	0.39 <sup>a</sup> ± 0.02	0.56 <sup>a</sup> ± 0.04	0.56 <sup>a</sup> ± 0.06
Generation		*	**	**	**	NS	*	**	**
F <sub>1</sub>	119	2.91 <sup>a</sup> ± 0.07	13.29 <sup>a</sup> ± 0.46	21.09 <sup>a</sup> ± 0.81	27.42 <sup>a</sup> ± 1.13	0.12 ± 0.00	0.56 <sup>a</sup> ± 0.02	0.89 <sup>a</sup> ± 0.04	1.16 <sup>a</sup> ± 0.05
F <sub>2</sub>	70	3.01 <sup>a</sup> ± 0.13	12.52 <sup>a</sup> ± 0.75	17.95 <sup>b</sup> ± 1.38	19.25 <sup>b</sup> ± 1.90	0.13 ± 0.01	0.53 <sup>a</sup> ± 0.03	0.74 <sup>b</sup> ± 0.06	0.81 <sup>b</sup> ± 0.08
F <sub>3</sub>	69	2.52 <sup>b</sup> ± 0.11	10.06 <sup>b</sup> ± 0.79	14.51 <sup>c</sup> ± 1.27	16.12 <sup>c</sup> ± 1.80	0.11 ± 0.05	0.45 <sup>b</sup> ± 0.03	0.64 <sup>c</sup> ± 0.05	0.71 <sup>c</sup> ± 0.08
F <sub>4</sub>	10	2.69 <sup>c</sup> ± 0.32	9.06 <sup>b</sup> ± 2.29	13.15 <sup>c</sup> ± 3.50	12.34 <sup>d</sup> ± 4.34	0.12 ± 0.02	0.38 <sup>c</sup> ± 0.09	0.55 <sup>d</sup> ± 0.14	0.53 <sup>d</sup> ± 0.19
Parity		**	**	**	NS	*	**	NS	NS
1	120	2.46 ± 0.07	9.85 ± 0.50	15.53 ± 0.91	19.20 ± 1.34	0.11 ± 0.00	0.44 ± 0.02	0.69 ± 0.04	0.86 ± 0.06
2	72	3.07 ± 0.10	13.97 ± 0.66	20.29 ± 1.21	23.86 ± 1.85	0.13 ± 0.01	0.58 ± 0.03	0.84 ± 0.05	1.00 ± 0.08
3	38	3.29 ± 0.15	14.84 ± 0.87	21.85 ± 1.63	25.98 ± 2.33	0.13 ± 0.01	0.59 ± 0.04	0.88 ± 0.07	1.03 ± 0.09
4	23	2.98 ± 0.23	12.67 ± 1.29	19.79 ± 1.98	21.94 ± 2.62	0.12 ± 0.01	0.51 ± 0.06	0.80 ± 0.91	0.88 ± 0.12
5	10	3.37 ± 0.31	14.19 ± 2.31	20.50 ± 1.31	22.52 ± 5.50	0.13 ± 0.12	0.54 ± 0.09	0.79 ± 0.17	0.87 ± 0.22
6	5	3.16 ± 0.69	11.80 ± 3.69	16.74 ± 5.89	21.18 ± 7.55	0.14 ± 0.03	0.51 ± 0.02	0.74 ± 0.26	0.92 ± 0.22
Reg. of ewes weight		**	**	*	–	**	NS	NS	–

N, Number of ewes lambed; \* $P < 0.05$ ; \*\* $P < 0.01$ ; NS, Non-significant, same superscript does not differ significantly.

parity significantly affected the EPE from birth to six months. In the present study, it was found that the EPE and EE increased up to 3rd parity and after that an erratic trend was noticed. It is expected that the ewes express the high production efficiency in the initial parities. The year non-significantly affects all the EPE and EE traits studied except EPE at 6 months and EE at 12 months of age.

The viable benefits accrued by gene infusion are reflected by increase in the litter size and ewe's productivity efficiency in terms of kg of lambs produced at marketing age of ewes bearing *FecB* gene (GM) compared to non-carrier (Malpura). This increase is high enough to increase the income of sheep farmers. Hence, from ewes bearing multiple lambs, we can expect to fetch more profit as compared to ewes bearing single lamb.

There was tremendous increase in the prolificacy, WR and in other ewe productive efficiency traits in *FecB* carrying GM half-breds compared to non-carriers. The study showed that B+ ewes are more capable of increasing WR and lambs weight produced per kg ewe weight as compared to BB and ++ ewes; and among carriers, multiple lamb producing ewes were more efficient. Hence, there is a need to increase the number of *FecB* carrying GM ewes that produce twins or triplets by *inter se* mating among carriers and discarding non-carriers for formulating intensive breeding programme through Marker Assisted Selection. The DNA test should be applied in routine in GM

crosses as a marker for identifying *FecB* gene carrier at an early stage. This will accelerate formulation of breeding strategies for improving the prolificacy and genetic improvement of non-prolific sheep breeds. Further study is required to backcross the GM rams with Malpura sheep to propagate the fecundity gene and improving the body weight gain in the *FecB* introgressed progeny.

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## Fuel properties and combustion characteristics of *Lantana camara* and *Eupatorium* spp.

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**In this study, we report fuel properties (basic density, high heating value, proximate and elemental parameters) and ash elemental composition of two important forest weed species, i.e. *Lantana camara* and *Eupatorium* spp. The physical, chemical and elemental properties of *L. camara* and *Eupatorium* spp. were compared with those of a mature tree (20 years of age) of *Eucalyptus* hybrid. The combustion characteristics under oxidizing atmosphere were also studied using thermogravimetric analysis. The burning profiles of the samples were derived by applying the derivative thermogravimetric technique. The two weed species were found to be different in their physical, chemical and elemental properties. The fuel properties and combustion characteristics, which largely depend upon the biochemical composition of biomass, were also different in these two weed species. The results suggested that both *L. camara* and *Eupatorium* spp. can be used as feedstock in thermochemical conversion processes. The emphasis was given to these species because of the huge biomass they produce. These species are widely present in different agroclimatic zones of India and can play a major role in future bio-energy schemes.**

**Keywords:** Biomass, burning profile, fuel properties, thermogravimetric analysis.

Biomass is the most common form of renewable energy sources. The potential of biomass to meet the domestic and industrial energy requirements of India has been well recognized<sup>1</sup>. Biomass fuels are promising, non-toxic and eco-friendly clean fuels<sup>1,2</sup>. Biomasses in various forms are suitable as energy feed stock. They can be either burned directly in a furnace or converted into high energy content fuels using biochemical or thermochemical conversion processes<sup>2</sup>. Among different biomasses, wood has received the most attention because of its long and continuing precedent as a fuel and biomass feed stock<sup>3,4</sup>. However, due to stringent government policies, which are largely aiming towards protection of native forests, there is hardly any supply of fuelwood from the forest<sup>5</sup>. Therefore, it is important to find out ways of utilizing alternative biomass resources for meeting heat and energy requirements. *Lantana camara* and *Eupatorium* spp. are

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