Volume 7 No.1 | January-June 2017 | ISSN : 0973-1865

Journal of Livestock Biodiversity

Society for Conservation of Domestic Animal Biodiversity

SOCDAB

SOCIETY FOR CONSERVATION OF DOMESTIC ANIMAL BIODIVERSITY

EXECUTIVE COMMITTEE

PRESIDENT

Dr Arjava Sharma, ICAR-NBAGR, Karnal

VICE PRESIDENTS

Dr M.S. Tantia, Principal Scientist, ICAR-NBAGR, Karnal
Dr R. Yinoo, NTR CoVS, Gannavaram
Dr G.C. Gahlot, Prof & Head COVAS RUVAS, Bikaner
Dr R.S. Gandhi, ADG (AP&B), ICAR, New Delhi

GENERAL SECRETARY

Dr R.K .Pundir, Principal Scientist, ICAR-NBAGR, Karnal

JOINT SECRETARIES

Dr A.K. Mishra, Principal Scientist, ICAR-NBAGR, Karnal
Dr Sanjeev Kumar, Principal Scientist, ICAR-CARI, Izatnagar

TREASURER

Dr N.K. Verma, Principal Scientist, ICAR-NBAGR, Karnal

MEMBERS

Dr Monika Sodhi, Principal Scientist, ICAR-NBAGR, Karnal
Dr P.K. Vij, Principal Scientist, ICAR-NBAGR, Karnal
Dr S. Jayakumar, Scientist, ICAR-NBAGR, Karnal
Dr R.S. Kataria, Principal Scientist, ICAR-NBAGR, Karnal
Dr K. P. Ramesha, Principal Scientist, ICAR-NDRI, SRS, Bangalore
Dr A.K. Patel, Principal Scientist, ICAR-CSWRI, Bikaner
Dr Bindya Liz Abraham, Assistant Professor, COVAS, Mannuthy
Dr Simarjeet Kaur, Assistant Professor, GADVASU, Ludhiana
Dr Aruna Pal, Assistant Professor, WBUAFS, Kolkata
Dr D. Cauveri, Assistant Professor, TANUVAS, Chennai

JOURNAL OF LIVESTOCK BIODIVERSITY

An official publication of the Society for Conservation of Domestic Animal Biodiversity

Chief Editor
Dr P.K. Singh,
ICAR-NBAGR, Karnal

Executive Editor
Dr S.K. Niranjan
ICAR-NBAGR, Karnal

Editor
Dr Indrajit Ganguly
ICAR-NBAGR, Karnal

Advisory Board

Chairman

Dr B. Prakash, Meerut

Members

Dr S.N.S. Parmar, Jabalpur

Dr G.K. Gaur, lzatnagar

Dr D. Kumar, Pantnagar

Dr D.N. Das, Bangalore

Dr Umesh Singh, Meerut

Dr S.S. Tomar, Mhou

Dr K.P. Singh, Hisar

Dr Vikas Vohra, Karnal

Editorial Office

Animal Genetics Division, ICAR-National Bureau of Animal Genetic Resources
P.O. Box 129, Karnal-132001 (Haryana), India
www.nbagr.res.in, email : livestockbiodiversity@gmail.com

Subscription (Annual)	**Indian**	**Foreign**
Individual	Rs 300.00	US $ 100.00
Institutional	Rs. 600.00	US $ 150.00
Single copy	Rs. 300.00	US $ 75.00

Published by Society for Conservation of Domestic Animal Biodiversity
Printer : Aaron Media, Karnal

JOURNAL OF LIVESTOCK BIODIVERSITY
VOLUME 7, NUMBER 1, 2017

CONTENTS

Phenotypic and molecular genetic studies on draughtability in Umblachery breed of cattle

M Kousalya Devi [1]*, S M K Karthickeyan[1], S N Sivaselvam[1], R Venkataramanan[2] and K G Tirumurugaan[3]
[1]*Department of Animal Genetics and Breeding, Madras Veterinary College, Chennai – 600 007, India*
[2]*Post Graduate Research Institute in Animal Sciences, Kattupakkam – 603 203, India*
[3]*Translational Research Platform for Veterinary Biologicals, TANUVAS, Chennai – 600 051, India*

ABSTRACT

A study was conducted to assess the draught potential of Umblachery cattle of Tamil Nadu. Phenotypic traits related to draughtability and their associations with 16 SNPs in six candidate genes (ACE, ADRB2, BDKRB2, GPX-1, IGF-1 and VEGFA) and five microsatellites in two candidate genes (IGF-1 and VEGFA) were studied in 112 Umblachery bullocks aged 2.5 to 12 years in the breeding tract. The mean body length, chest girth and height at withers were 120.21±0.89, 151.92 ±1.04 and 123.94 ±0.72 cm respectively. The estimated average body weight was 265.64 ± 5.88 kg. Draughtability parameters (in 43 pairs of bullocks) viz. stride length, speed of ploughing and horse power generated during ploughing averaged 1.20 ± 0.21 m, 0.95 ± 0.03 m/s and 0.39 ± 0.04 hp (for an average draught load of 27.5 ± 2.05 kg obtained in this study) respectively. Among the draughtability parameters, only stride length had highly significant positive correlation with all the three morphometric traits. Optimum draft load at which Umblachery bullocks could give uniform and maximum power output was found to be 75 to 78 kg. The mean serum creatine kinase and lactate were estimated to be 52.35 ± 5.04 U/L and 52.87 ± 8.22 mg/dL before work; and 35.92 ± 4.17 U/L and 35.98 ± 1.68 mg/dL after work respectively. Microsatellite locus, VEGFA-(ACAT)n was found to be significantly associated with serum creatine kinase and lactate, with 373/373 and 365/365 bp as favourable genotypes. Microsatellite locus, VEGFA-(GA)n was found to be highly significantly associated with serum lactate alone, with 175/177 and 177/177 bp as favorable genotypes. SNPs ACE 2620 (A>G) and BDKRB2 41872 (G>A) were found to be significantly associated with body weight and creatine kinase respectively, with GG genotypes in both SNPs as favorable. Thus, ACE, BDKRB2 and VEGFA genes could be used as potential candidate markers for selecting Umblachery cattle breed with high draught power.

Keywords: Draughtability, molecular marker, Umblachery cattle
***Corresponding author:** drkousi31@gmail.com
Manuscript received: 01.3.2017; Manuscript accepted: 25.5.2017

INTRODUCTION

Draught Animal Power (DAP) is listed as one of the 14 renewable resources of energy in UN Conference on New and Renewable Sources of Energy held in Nairobi, 1981. The recent trend for mechanization though accelerates agricultural production, consumes lot of natural gas resources and fossil fuels. Sastry and Thomas (2005) stated that India's petroleum and natural gas resources may last only for 25 to 35 years and coal for 130 to 140 years. India is a rich source of DAP with 25 out of 40 recognized cattle breeds belonging to draught type. The energy for ploughing two-third of cultivable area and two-third of rural transport are coming from animals (Report, 2008). But, there also seems to be a constant decline in the number of male cattle from 101 million in 1992 to around 83 million in 2007 (Report, 2012). Shisode *et al.* (2010) estimated that DAP saved 20 million tonnes of petroleum per year. Though extensive studies had been carried out on physical characteristics, work performance and biochemical parameters of work bullocks, reports on genetic improvement of draught cattle and molecular markers related to draught power are scanty. Hence a candidate gene study was undertaken to find out the association of molecular markers (SNPs and microsatellites) in six candidate genes with the draughtability traits in Umblachery cattle, a prominent draught breed of South India.

MATERIALS AND METHODS

The principal morphometric traits like body length, chest girth and height at withers; and draughtability

traits namely, stride length, speed and horse power during ploughing were measured in 112 and 86 (43 pairs) Umblachery bullocks respectively sampled from the breeding tract comprising of Thanjavur, Thiruvarur and Nagapattinam districts. The age of the bullocks varied from 2.5 to 12 years. In addition, physiological parameters *viz*. pulse rate, respiration rate and rectal temperature were also recorded in animals before and after work. Simultaneously, blood samples were collected from 98 bullocks (both in EDTA and serum vacutainers) before and after work, to isolate the genomic DNA and to estimate the biochemical parameters such as serum creatine kinase and lactic acid using commercially available diagnostic kits (Agappe Diagnostics kit-Catalogue No. 11405007, 11405002 and Bio systems-Catalogue No.735-10 respectively.

The speed of ploughing was calculated by measuring the time taken to cover a particular distance and stride length was measured by dividing the distance covered with the number of strides taken. Horse power generated was estimated with a spring balance by using a modified technique given by Maurya and Devadattum (1982b). Spring balance was used to calculate the pull (in kg) directly. Draught was calculated by multiplying the pull in kg by Cosθ, where Cosθ is the angle the beam of plough makes with the horizontal ground. Thus, horse power is calculated by Draught X Speed (m/s)/75, where 75 is the constant.

Molecular Genetic Marker Identification

Genomic DNA was extracted using standard Phenol-Chloroform extraction procedure (Sambrook *et al*., 1989) with slight modifications by using DNAzol reagent. A total of 16 SNPs already identified and reported in the exons and promoter regions of Angiotensin I-converting enzyme (ACE), Adrenergic beta 2 receptor (ADRB2), Bradykinin beta 2 receptor (BDKRB2), Glutathione peroxidase 1 (GPX-1), Insulin-like growth factor-1 (IGF-1), Vasculo endothelial growth factor alpha (VEGFA) genes and 5 microsatellites in the promoter and intronic regions of IGF-1 and VEGFA genes were screened and genotyped. Out of sixteen SNPs, ten were genotyped using Tetra-Primer ARMS-PCR and six SNPs were genotyped using Restriction Fragment Length Polymorphism (RFLP). PCR was performed for 96 samples for the five microsatellite regions and the amplicons were genotyped (Eurofins Genomics, Bangalore).

Statistical Analysis

Using SPSS version 20, phenotypic correlations between morphometric and draughtability traits were calculated by Pearson's formula. General Linear Model (GLM) was used to find the effect of microsatellites in IGF-1 and VEGFA genes on all draught related traits. Association of effects of each SNP with physiological, biochemical and draughtability parameters was carried out by Restricted Maximum Likelihood Method (REML), fitting mixed model equation using WOMBAT programme of Meyer (2011). Additive genetic effect was fitted as random effect. SNP effect was fitted as fixed effect, in addition to the significant fixed effects obtained from GLM.

RESULTS AND DISCUSSION

Morphometric Traits

The overall least-squares means of morphometric traits (n=112) of body length, chest girth, height at withers and body weight were 120.21 ± 0.89 cm, 151.92 ± 1.04 cm, 123.94 ± 0.72 cm and 273.4 ± 5.08 kg respectively. Body length and chest girth of Umblachery bullocks obtained in this study, concurred with the observations of Rajendran *et al.* (2008) in 383 Umblachery bullocks (118.7 and 150.9 cm respectively). Whereas, height at withers recorded in this study (123.34 ±0.72 cm) was higher compared to that observed by the same author (116.8 cm). When compared with Kangayam cattle (n=102; considered as the ancestral source for Umblachery breed) having the body length, chest girth and height of 154.8, 184.8 and 146.7 cm respectively (Panneerselvam and Kandasamy, 1999), the Umblachery cattle is shorter. In another study made by Ganapathi *et al.* (2013) in Bargur bullocks (n=444), another draught breed of Tamil Nadu, these measurements averaged 126.29, 139.44 and 126.33 cm respectively. The mean of body length, height at withers and chest girth measured in other draught breeds like Hallikar (n-119) and Malvi (n=4),

respectively by Singh *et al.* (2008) and Singh *et al.* (2009), were 138.94, 134.55 and 163.15; and 157.2, 135.2 and 162.7 cm, emphasizing that Umblachery breed is an outcome of selection for short stature.

Physiological Parameters

The least-squares means of physiological parameters *viz*. heart rate, pulse rate, respiration rate and rectal temperature obtained before and after work in this study were 77.51 ± 2.44/min, 74.02 ±2.15/min, 23.56 ± 0.72/min and 38.4 ± 0.1°C; and 90.01 ± 2.44/min, 86.31 ± 2.19/min, 29.18 ± 1.06/min and 39.3±0.1°C, respectively. A highly significant ($P<0.01$) difference was observed in the physiological parameters before and after work. Similar increase in physiological reactions after work was recorded by Maurya and Devadattam (1982a), Bhosrekar and Mangurkar (1989) and Singh (2013) in crossbred bullocks; Kumaravelu *et al.* (1997) in Kangayam; Vinoo *et al.* (2010) in Ongole; and Singh *et al.* (2014) in Malvi bullocks.

Biochemical Parameters

The least-squares means of serum creatine kinase before and after work were 52.35±5.04 and 35.92±4.17 U/L, respectively. Serum levels of creatine kinase before and after work did not differ significantly in Umblachery bullocks. These results are inconsistent with the report of Brancaccio *et al.* (2007) that the significant increase of creatine kinase after exercise was lower in trained subjects. However, the lower level of serum creatine kinase after work and insignificant difference before and after work obtained in the present study indicate the better ability of the bullocks to cope with strenuous exercise without marked muscle damage, which otherwise would result in significant increase in the level of serum creatine kinase after work.

The least-squares means of serum lactate before and after work were 52.87 ± 8.22 and 35.98 ± 1.68 mg/dL respectively. Serum levels of lactate were significantly lower after work than before work. This may be attributed to the reason that, as the samples were collected almost immediately after work (which may not in many case be after exhaustion), excess lactate produced in the muscle as a result of anaerobic glycolysis may be effluxed into plasma from which it is transported to various tissues like RBCs, liver, heart and other skeletal muscle cells where it acts as an important source of energy, resulting in reduced serum lactate levels (Pösö, 2002). It was also stated that monocarboxylate transporter (MCT), a major transporter of lactate, was higher in RBCs of athletic animal species like horse and dog and identified the higher concentration of lactate in RBCs of horses with best individual performance, but not in serum. This might also be a possible reason for lower serum lactate levels after work. However, RBCs, plasma and whole blood all have different values from each other, as there is a marked plasma to RBC lactate gradient during exercise as well as at rest (Goodwin *et al*., 2007). Hence, to perform individual-to-individual comparisons, Pösö (2002) suggested the use of whole blood lactate values.

Draughtability Traits

The overall least-squares means of draughtability parameters of stride length, speed of ploughing and horse power generated during ploughing recorded in 86 bullocks were 1.20 ± 0.21 m, 0.95 ± 0.03 m/s and 0.39 ± 0.04 hp respectively (for an average draft load of 27.5 ± 2.05 kg). The speed of Umblachery cattle recorded in this study is almost similar to reports on indigenous and crossbred bullocks, that include 0.99 m/s in a pair of Hariana bullocks (Devadattam and Maurya, 1978) and 0.93 m/s in Jersey × Red Sindhi crossbreds (n=2) (Maurya and Devadattam, 1982b); but lower than 1.59 m/sec in Ongole bullocks (n=55) (Vinoo *et al.,* 2010). The speed of Umblachery bullocks was also much lesser when compared to Kangayam cattle, exhibiting 1.24 m/s at ploughing (Sree Kumar and Thomas, 1990) and 1.31 m/s at carting (Gogoi, 2012). The stride length recorded was higher on comparison with Kumaravelu (1995) for Kangayam bullocks (n=20) on ploughing (0.88 m) and Gogoi (2012) on carting (0.77 m).

The average draft obtained in this study was 27.5 ± 2.05 kg, for which the mean horse power developed by Umblachery bullocks was 0.39 ± 0.04 hp. This was lesser when compared to HF × Hariana crossbreds (0.75 hp; Gattewar *et al*., 1989), Kangayam bullocks

Table 1. Phenotypic correlations of morphometric traits with draughtability parameters

Traits	Body length	Chest girth	Height at withers	Stride length	Draft	Speed
Chest girth	0.562**					
Height at withers	0.434**	0.815**				
Stride length	0.316**	0.333**	0.399**			
Draft	0.043	0.116	0.103	0.459**		
Speed	0.137	0.123	0.115	0.437**	0.321**	
Horse power	0.086	0.158	0.133	0.491**	0.974**	0.484**

** -Highly Significant (P<0.01)

(0.73 hp; Kumaravelu, 1997) and Ongole bullocks (0.57 hp; Vinoo *et al.,* 2010). However, power output of an animal is a complex trait that depends on various external factors like draft, speed, duration of work, climate, moisture of the soil, type of agricultural operation, as opined by Bhattacharya and Singh (1987). In addition, human factor plays a major role in exhibition of draught power in bullocks. The lesser horse power in Umblachery cattle obtained in this study could be mainly attributed to the heterogenous environment that existed at field level during recording of draught performance; whereas in the other studies cited above, the draught performances were recorded in organised farms under uniform test conditions with lesser number of samples (less than 10).

Phenotypic Correlations

The phenotypic correlations between morphometric and draughtability traits are given in Table 1. The phenotypic correlations of morphometric parameters revealed highly significant (P<0.01) positive correlations among them. Of all the draught parameters, only stride length had highly significant (P<0.01) positive correlation with all the three morphometric traits. Draught parameters were also highly significantly and positively correlated with each other. Highly significant (P<0.01) positive correlations were also detected between draught parameters and draught load applied.

Association of SNPs with Biochemical Parameters and Draughtability Traits

Of the 16 SNPs genotyped, only 13 were polymorphic and three were monomorphic. The effects of 13 SNPs on physiological, biochemical and draughtability parameters were analyzed. It was found that only two SNPs had association with draughtability traits (Table 2), one at ACE 2620 (A>G) for body weight and the other at BDKRB2 41872 (G>A) for serum creatine kinase before and after work. This supports our hypothesis that ACE and BDKRB2 genes are associated with draughtability in *Bos indicus* cattle. However, SNPs previously identified in IGF-1 (Gogoi, 2012) as associated with stride length and serum creatine kinase, did not exhibit any association with draught power in Umblachery bullocks, suggesting requirement of a detailed study with more number of samples. This study also indicates that serum creatine kinase can be used as one of the biomarkers to assess the endurance power.

Draughtability of an animal greatly depends on its body weight as it is evident that the draughtability parameter is being expressed in terms of per cent body weight. For the same draft, heavier bullocks moved faster and consequently produced more power compared to bullocks with lesser weight (Premi and Singh, 1987). They demonstrated a linear

Table 2. Association of SNPs with serum creatine kinase and body weight Umblachery bullocks

SNP locus	Serum creatine kinase (U/L)						Body weight		
	Before work			After work					
	Estimate	d.f.	t-value	Estimate	d.f.	t-value	Estimate	d.f.	t-value
ACE A-2620G	8.31 ± 7.54	72	1.101NS	8.49 ± 8.86	46	0.958NS	-9.79±7.22	79	-1.36*
BDKRB2 G-41872A	14.89 ± 5.83		2.556*	15.65 ± 7.18		2.180*	-15.99±6.15		-2.60NS

d.f. – degrees of freedom;* - Significant (P<0.05); NS – Not significant; 'Estimates' refers to partial regression coefficient.

relationship between work output at maximal load and body weight. Kaushik *et al.* (1987), while discussing structural and functional relationship with draught power stated that draught power generated by the bullocks is largely dependent on their body weight especially the amount distributed over their forelimbs (higher chest girth and larger forelimbs). In the current study, in ACE 2620 (A>G), a non-synonymous mutation, was found to be associated (P<0.05) with the variation in body weight. Similar studies in human athletic populations revealed an insertion/deletion polymorphism in ACE gene associated with endurance power. Myerson *et al.* (1999) found that athletes with DD genotype have a greater percentage of type II fibres which was associated with power performance. Insertion allele was associated with increase in type I (slow-twitch fibres) muscle fibres responsible for endurance. Association of insertion allele with endurance was demonstrated by Montgomery *et al*. (1998) in mountaineers climbing beyond 7000 m; Myerson *et al*. (1999) in long distance runners (>5000 m) and Grenda (2014) in long distant swimmers.

Association of Microsatellites with Biochemical Parameters and Draughtability Traits

A strong association (P<0.01) was detected between VEGFA $(GA)_n$ repeats (found in 13736 to 13755 bp region of VEGFA gene) and serum lactate levels (Table 3). VEGFA $(ACAT)_n$ repeats (found between 13736 and 13755 bp) was identified to have a significant (P<0.05) association with both serum lactate and serum creatine kinase levels before work. A similar conclusion as in this study, (association of VEGFA genes with serum lactate level) was arrived by Ahematov *et al*. (2008) while studying the G-634C polymorphism in human VEGFA gene as they identified the association of 'C' allele with increased maximal oxygen consumption (VO_2max) and decreased lactate level. The possible mechanism of association of serum levels of creatine kinase with VEGFA could be attributed to strenuous exercise damaging the skeletal muscle cell structure at the level of sarcolemma and Z-disks which results in leakage of creatine kinase into the interstitial fluid (Brancaccio *et al*., 2007). During strenuous exercise, VEGFA level increases as a physiological counteracting mechanism during inflammation to maintain an intact endothelial cell layer (Jee and Jin, 2012). This indicates the involvement of VEGFA gene in increased serum creatine kinase levels during intense exercise.

In the light of above discussion, it could be inferred that the SNPs at ACE A-2620G have a potential influence on body weight; BDKRB2 G-41872A on serum creatine kinase; polymorphisms in VEGFA $(GA)_n$ microsatellite locus on serum lactate; and polymorphisms in VEGFA $(ACAT)_n$ locus on serum creatine kinase and lactate levels, thereby exerting a cumulative effect on draught power in *Bos indicus* cattle. These loci influencing various parameters pertaining to draught power in bovines could be used as candidate markers for selecting a better genotype of Umblachery cattle for agricultural operations in marshy fields of Cauveri Delta regions of Tamil Nadu. This would also strengthen the conservation efforts of Umblachery cattle at stakeholders level, through their expanding utility value.

Table 3. Least-squares analysis of variance for the effects of genotypes of VEGFA $(GA)_n$ and VEGFA $(ACAT)_n$ on biochemical parameters (creatine kinase and serum lactate) before and after ploughing

Biochemical parameters		VEGFA $(GA)_n$			VEGFA $(ACAT)_n$		
		MSS	d.f.	F value	MSS	d.f.	F value
Creatine kinase	Before work	3204.89	2	2.58^{NS}	1325.94	5	2.45*
	After work	57.83	2	0.07^{NS}	293.56	4	0.39^{NS}
Serum lactate	Before work	37550.60	2	11.41**	10416.90	5	2.53*
	After work	22.14	2	0.12^{NS}	304.81	5	1.91^{NS}

MSS – Mean sum of squares; d.f.- degrees of freedom; ** - Highly significant (P<0.01); * - Significant (P<0.05); ^{NS} – Not significant

REFERENCES

Ahmetov II, Khakimullina AM, Popov DV, Missina SS, Vinogradova OL and Rogozkin VA. 2008. Polymorphism of the vascular endothelial growth factor gene (VEGF) and aerobic performance in athletes. Human Physiology, 34 (4): 477 – 481.

Bhattacharya NK and Singh K. 1987. Comparison of draftability of crossbred and indigenous cattle and variation in their physiological response during work. In: Utilisation and Economics of Draught Animal Power, (eds. N.S.L. Srivastava and T. P. Ojha), pp. 26-38. CIAE publishing, India.

Bhosrekar MR and Mangurkar BR. 1989. Physiological responses of cross bred and local bullocks in different agro climatic zones. Indian Journal of Animal Science 59 (10): 1324-1328.

Brancaccio P, Maffulli N and Limongelli FM. 2007. Creatine kinase monitoring in sport medicine. British Medical Bulletin 81-82: 209–230.

Devadattam DSK and Maurya NL. 1978. Draftability of Hariana bullocks. Indian Journal of Dairy Science 31: 121-127.

Ganapathi P, Rajendran R, Subramanian A and Meenakshisundaram S. 2013. Bargur cattle: Characterization and management practices. Indian Veterinary Journal 90: 9 – 10.

Gattewar AB, Singh RA and Yadav RS. 1989. Working capacity and behavior of crossbred (F_1) verses Zebu bullocks. Indian Journal of Animal Production and Management 5: 115-118.

Gogoi A. 2012. Characterization of bovine insulin-like growth factor – 1 (IGF-1) gene and its association with draught power. M.V.Sc., Thesis submitted to the Tamilnadu Veterinary and Animal Sciences University, Chennai – 51.

Goodwin ML, Harris JE, Hernandez A and Gladden LB. 2007. Blood lactate measurements and analysis during exercise: Guide for clinicians. Journal of Diabetes Science and Technology 1: 558-569.

Grenda A, Leonska-Duniec A, Kaczmarczyk M, Ficek K, Krol P,

Cieszczyk P. and Zmijewski P. 2014. Interaction between ACE I/D and ACTN3 R557X polymorphisms in Polish competitive swimmers. Journal of Human Kinetics 42: 127-136.

Jee H. and Jin Y. 2012. Effects of prolonged endurance exercise on vascular endothelial and inflammation markers. Journal of Sport Science and Medicine 11: 719-726.

Kaushik SN, Shukla DC and Saxena SK. 1987. Certain aspects of structural functional relationship in animal draught mechanism. In: Utilisation and Economics of Draught Animal Power. (eds. N.S.L. Srivastava and T. P. Ojha), pp.88-91.CIAE publishing, Bhopal.

Kumaravelu N. 1995. Analysis of farming systems involving Kangayam breed of cattle. M.V.Sc. Thesis submitted to Kerala Agricultural University, Thrissur – 680 651

Kumaravelu N, Thomas CK and Sreekumar D. 1997. Farm work performance of Kangayam bullocks in two locations. Indian Veterinary Journal 74: 951-954.

Maurya NL and Devadattam DSK. 1982a. Response of some physiological parameters of crossbred bullocks to different draft and ambient conditions. Indian Journal of Dairy Science 35: 18-25.

Maurya NL and Devadattam DSK. 1982b. Work performance of crossbred bullocks. Indian Journal of Dairy Science 35:26-30.

Meyer K. 2011. WOMBAT-A tool for mixed model analyses in quantitative genetics by restricted maximum likelihood (REML). Journal of Zhejiang University Science 8: 815-821.

Montgomery HE, Marshall R, Hemingway H and Myerson S. 1998. Human gene for physical performance. Nature 393: 221.

Myerson S, Hemingway H, Budget R, Martin J, Humphries S and Montgomery H. 1999. Human angiotensin I-converting enzyme gene and endurance performance. Journal of Applied Physiology 87: 1313–1316.

Panneerselvam S and Kandasamy N. 1999. Physical characters and load hauling capacity of Kangayam bullocks of South India. Draught Animal News 31: 17-20.

Poso AR. 2002. Monocarboxylate transporters and lactate metabolism in equine athletes: A review. Acta Veterinaria Scandinavica 43: 63-74.

Premi SCL and Singh G. 1987. Performance of bullocks under varying conditions of load and climate. In: Utilisation and Economics of Draught Animal Power. (eds. N.S.L. Srivastava and T. P. Ojha), pp.52-63. CIAE publishing, India.

Rajendran R, Raja TV, Thiruvenkadan AK, Nainar AM and Thangaraju P. 2008. Morphobiometrical characteristics and management of Umblachery cattle from coastal region of Tamilnadu, India. Livestock Research and Rural Development 20: 40.

Report, 2008. Eleventh Five Year Plan Report of the Working Group on Animal Husbandry and Dairying (2007-2012). Planning Commision, New Delhi.

Report, 2012. Eleventh Five Year Plan Report of the Working Group on Animal Husbandry and Dairying (2012-2017). Planning Commision, New Delhi.

Sambrook J, Fritsch EF and Maniatis T. 1989. Molecular Cloning: A Laboratory Manual (2nd ed.). Cold Spring Harbor Laboratory, Cold Spring Harbor, New York, ISBN-10:9780879693091.

Sastry NSR and Thomas CK. 2005. Livestock Production Management (4th edn.). Kalyani Publishers, New Delhi. pp.449.

Shisode MG, Bhokare SM, Kulkarni MD and Khanvilkar AV. 2010. Draught animal power - our nation's wealth. SMVS' Dairy Year Book. pp. 31-32.

Singh B, Shivhare M, Khushram M and Shakya S. 2014. Study on the physiological and haematological changes during work performance of Malvi bullocks. Haryana Veterinarian 53 (1): 72-73.

Singh B, Nanavati S and Joshi SK 2013. Study on the draughtability of Malvi bullocks. Indian Journal of Field Veterinarian 8 (4): 61-64.

Singh PK, Pundir RK, Ahlawat SPS, Naveen Kumar S, Govindaiah MG and Asija K. 2008. Phenotypic characterization and performance evaluation of Hallikar cattle in its native tract. Indian Journal of Animal Sciences 78: 211-214.

Singh RC and Singh CD. 2009. Effect of age and body size on the work output of Malvi and crossbred oxen. Indian Journal of Animal Sciences 79: 850-852.

Sreekumar D and Thomas CK.1990. Draught efficiency and thermal strain of Kangayam and Jersey-Red Sindhi crossbred bullocks in hot-humid tropics. Indian Journal of Animal Sciences 60 (5): 582-586.

Vinoo R, Rao GN, Gupta BR and Rao KB. 2010. Estimation of draughtability of Ongole bullocks by different methods. Tamilnadu Journal of Veterinary and Animal Sciences 6: 24-30.

Sire evaluation (single and multi-trait) in terms of relative efficiency, error variance and spearman's coefficient of rank correlation and product moment correlation in Red Sindhi cattle

P K Mallick[1]*, A K Ghosh and Harpal Singh
Govind Ballabh Pant University of Agriculture & Technology, Pantnagar, Uttarakhand 263145 India

ABSTRACT

The sire evaluation method, which estimated the breeding values of sires with the least error variance, was taken as the best and most efficient method. The records of production and reproduction performances of first lactation traits of 717 Red Sindhi cows, progeny of 58 sires, spread over a period of 40 years (1966-2005) from CCBF, Chiplima, Orissa under the control of Department of Animal Husbandry, Dairying and Fisheries, Ministry of Agriculture, Government of India and CBF, Kalsi, Deheradun, Uttarakhand were analyzed. The present investigation to know the effectiveness and efficiency of different sire evaluation methods was evaluated on the basis of efficiency of a particular method within sire variance or error variance, Spearman's coefficient of rank correlations and simple correlation /product moment correlation. Most efficient methods of sire evaluation D, LSM, DFREML-I, DFREML-II, DFREML-III, BLUP-I, BLUP-II and BLUP-III in terms of relative efficiency for (single and multi-trait) were 9.70, 83.33, 100.00, 80.54, 85.17, 96.88, 96.88 and 96.03 and ranked VII, VI, I, V, IV, II, II and III respectively. Spearman's coefficient of rank correlation among breeding values of sires' for all eight methods ranged from 0.80 (and DFREML-II) to1.00 (LSM with DFREML-III, BLUP-I, BLUP-II) which were positive and highly significant (P≤0.01). The rank correlations of DFREML-I (most efficient method) with , LSM, DFREML-II, DFREML-III, BLUP-I, BLUP-II and BLUP-III were 0.87, 0.99, 0.98, 0.99, 0.99, 0.99 and 0.96, respectively and were highly significant (P≤0.01). The high and significant rank correlations between the breeding values of common sires estimated by different methods indicated that all these methods did not differ significantly in ranking of sires. The high relative efficiency of BLUP-I and BLUP-II (both 96.88 percent) in comparison to most efficient method along with high product moment correlation of BLUPF90-I and BLUPF90-II with DFREML-I indicated that either of these two methods could possibly be used for evaluation and ranking of Red Sindhi sires with equal efficiency and accuracy.

Key words- Relative efficiency, error variance, Spearman's coefficient of rank correlation
Corresponding author: p_mallick04@yahoo.co.in
Present Address : [1]Senior Scientist(AG&B) SRRC,CSWRI,Mannvanur,Kodaikanal-3,Tamil Nadu, India
Manuscript received: 11.1.2017; Manuscript accepted: 24.3.2017

INTRODUCTION

The effectiveness of sire evaluation is the backbone of any breed improvement programme. This investigation was planned to evaluate Red Sindhi sires and ranked on the basis of their estimated breeding values considering FLMY of their progeny considering as single trait under univariate models viz. simple daughter's average (D) method, least squares method (LSM), DFREML-I method and BLUP-I method. However, for evaluation of sires by multi traits, the multi-variate models viz. DFREML-II and DFREML- III and BLUP-II and BLUP-III methods were used. The FLMY was taken as principal trait along with FLP in DFREML-II and BLUP-II methods, while both the traits (FLMY and FLP) along with AFC were taken in DFREML-III and BLUP-III methods. These traits were considered based on higher magnitude of heritability and genetic correlation with the principal trait FLMY. The error variance of breeding values of sires was estimated and used in computing the relative efficiency of different sire evaluation methods. The sire evaluation method, which estimated the breeding values of sires with the least error variance, was taken as the best and most efficient method. In the present study, the DFREML-I with univariate model using single trait i.e. FLMY was having the lowest error variance compared to other seven methods used and accordingly, it was adjudged the most efficient sire evaluation method. Relative efficiency of other seven methods of sire evaluation

were calculated with respect to the most efficient method i.e. DFREML-I. The estimated error variance and relative efficiency of different sire evaluation methods used for estimation of breeding value. Spearman's coefficient of rank correlation commonly called as rank correlation is a measure of correspondence between ranks and applies to data in the form of ranks estimated by different methods. In the present study, the ranks of Red Sindhi sire on the basis of breeding values estimated by different sire evaluation methods were taken into account for measuring rank correlation which is of statistical significance to judge the association among ranks of sires estimated by different sire evaluation methods. The coefficients of rank correlations among breeding values of sire computed by eight different methods and were compare the effectiveness of BLUP with other conventional methods of sire evaluation in Red Sindhi cattle.

MATERIALS AND METHODS

The records of production and reproduction performances of first lactation traits of 717 Red Sindhi cows, progeny of 58 sires, spread over a period of 40 years (1966-2005) were analyzed.The effectiveness and efficiency of different sire evaluation methods were evaluated on the basis of within sire variance or error variance, spearman's coefficient of rank correlations (Steel and Torrie, 1960) and product moment correlation. Higher rank correlation amongst the sires from different sire evaluation methods revealed higher degree of similarity of ranking from different methods.

RESULTS AND DISCUSSION

The error variance of breeding values of sires was estimated and used in computing the relative efficiency of different sire evaluation methods. The sire evaluation method, which gives least error variance, was taken as the best.. In the present study, the DFREML-I using single trait i.e. FLMY had the lowest error variance and highest relative efficiencies compared to the other seven methods used. Relative efficiency of other seven methods of sire evaluation were calculated with respect to the most efficient method i.e. DFREML-I. The estimated error variance and relative efficiency of different sire evaluation methods in the present study are presented in Table 1. The BLUP-I with univariate model using single trait as FLMY and BLUP-II with multivariate model using two traits as FLMY and FLP had similar relative efficiency of 96.88% to that of DFREML-1 (100%) and were placed as second best methods followed by BLUP-III with multivariate model using three traits as FLMY, FLP and AFC with a relative efficiency of 96.02 %, which ranked third with respect to the relative efficiency among the methods used. The DFREML-III, DFREML-II, LSM and were found with a relative efficiency of 85.17%, 80.54%, 83.33% and 9.70%, respectively and ranked as IVth, Vth, VIth and VIIth best method for evaluation of Red Sindhi sires in the present study. Further, it was found that among the multivariate sire evaluation methods BLUP-II using two traits (FLMY and FLP) was found to be most efficient sire evaluation methods followed by BLUP-III, DFREML-III and DFREML-II, respectively (Table 1). Arora (1981), Jain (1996) and Jain and Sadana (2000) reported the BLUP method under multi-trait animal model incorporating FLMY with other trait to be more efficient and accurate for sire evaluation in different breeds of cattle and buffalo. In the present investigation, simple daughter average method () was having highest error variance among all methods of sire evaluation used. This could be because non-genetic variations were not estimated and removed from data prior to the estimation of breeding values of sires, which might have resulted into the highest error variance and lowest relative efficiency of this method. This finding was in agreement with the reports of Jain and Malhotra (1971), Gill and Parmar (1988), Sahana (1996), Sahana and Gurnani (2000) and Aswathanarayana et al. (2003) as they have reported highest error variance and least relative efficiency for simple daughter average () than any other method. BLUP-I and BLUP-II were similar and were placed second best as they have shown lower error variance than BLUP-III, DFREML-III DFREMEL-II, LSM and . Henderson (1973), Danell (1982), Harvey (1987), Singh et al. (1992), Raheja (1992), Tailor et al. (2000) and Dahiya et al. (2005) have also reported BLUP as one of the best and most efficient sire evaluation

Table 1. Efficiency of different sire evaluation methods

Sire evaluation method	Traits considered	Error variance	Relative efficiency	Rank
FLMY	3335015.00	9.70	VII	
LSM	FLMY	388293.79	83.33	VI
DFREML-I	FLMY	323576.65	100.00	I
DFREML-II	FLMY and FLP	401780.00	80.54	V
DFREML-III	FLMY, FLP and AFC	379920.00	85.17	IV
BLUP-I	FLMY	334000.00	96.88	II
BLUP-II	FLMY and FLP	334000.00	96.88	II
BLUP-III	FLMY, FLP and AFC	337000.00	96.02	III

method in different breeds of cattle. However, Taneja and Rai (1990), Deb et al. (1998) and Kishore (1993) in different breeds of cattle reported in field condition that LSM was the most efficient method of sire evaluation compared to BLUP method. Banik and Gandhi (2006) in Sahiwal and Mukherjee (2005) in Frieswal also reported that DFREML method was the most efficient method for sire evaluation as it was having lowest error variance compared to LSM and BLUP with univariate model.

The coefficients of rank correlations among breeding values of sire computed by eight different methods were presented in Table 4. The coefficient of rank correlation among breeding values of sires' for all eight methods ranged from 0.80 (D and DFREML-II) to1.00 (LSM with DFREML-III, BLUP-I, BLUP-II) which were positive and significant (P≤0.01). The rank correlations of DFREML-I (most efficient method) with , LSM, DFREML-II, DFREML-III, BLUP-I, BLUP-II and BLUP-III were 0.87, 0.99, 0.98, 0.99, 0.99, 0.99 and 0.96, respectively and were highly significant (P≤0.01). The high and significant rank correlations between the breeding values of sires indicated that all these methods did not differ significantly in ranking of sires. However, the second highest relative efficiencies of BLUP-I and BLUP-II (96.88% of both) in comparison to most efficient method DFREML-I and high rank correlations with DFREML-I indicated that either of these two methods could possibly be used for evaluation of Red Sindhi sires with equal efficiency and reasonable accuracy. These findings agreed with Mukherjee (2005) in Frieswal cattle, Banik and Gandhi (2006) in Sahiwal and Tailor and Kothari (2006) in Surti buffaloes as they have reported significant rank correlations between single trait (DFREML for univariate model) and multi-trait methods (DFREML for multi trait model) of sire evaluation and observed that the breeding values and the ranks of the sires did not differ significantly from both the models. Similarly, for single trait sire evaluation methods various workers have reported highly significant rank correlations among breeding values estimated by different methods of sire evaluation. Vij and Tiwana (1988), Gandhi and Gurnani (1991), Murdia and Tripathi (1992), Raheja (1992), Sahana (1996), Delkur and Kothekar (1999), Tailor et al. (2000), Sahana and Gurnani (2000), Gaur et al. (2001), Vinoo et al. (2005), and Baink and Gandhi (2006) reported high rank correlation between LSM, SRLS and BLUP suggesting that these methods were similar in ranking of dairy sires and any of the methods could be used to obtain unbiased estimate of breeding values of sires. Product moment correlation among sires merit calculated by eight different methods of sire evaluation ranged from 0.66 to 1.00 and all the values were positive and significant. Atil and Khattab (2000) reported that product moment correlations were also higher and almost similar to those of rank correlations. The estimates of product moment correlations confirm the fact that sire evaluation by were highly correlated with LSM; LSM with DFREML-I, DFREML-III, BLUP-I and BLUP-II; DFREML-I with BLUP-I and BLUP-II; DFREML-III with BLUP-I and BLUP-II and BLUP-I with BLUP-II. The product moment correlation of DFREML-I (most efficient method) with , LSM, DFREML-II, DFREML-III, BLUP-I, BLUP-II and BLUP-III were 0.82, 1.00, 0.95, 0.99, 1.00, 1.00 and 0.98, respectively. The high relative efficiency of BLUP-I and BLUP-II (both 96.88%) in comparison to most efficient method along with high product moment correlation of BLUP-I and BLUP-II

Table 2. Spearman's Rank and Product moment correlation among different sire evaluation methods

Methods	D	LSM	DFREML-I	DFREML-II	DFREML-III	BLUP-I	BLUP-II	BLUP-III
D	0.85**	0.87**	0.80**	0.84**	0.84**	0.84**	0.83**	
LSM	1.00**		0.99**	0.99**	1.00**	1.00**	1.00**	0.96**
DFREML-I	0.82**	1.00**		0.98**	0.99**	0.99**	0.99**	0.96**
DFREML-II	0.66**	0.96**	0.95**		0.99**	0.99**	0.99**	0.96**
DFREML-III	0.76**	1.00**	0.99**	0.98**		1.00**	1.00**	0.97**
BLUP-I	0.78**	1.00**	1.00**	0.97**	1.00**		1.00**	0.97**
BLUP-II	0.78**	1.00**	1.00**	0.97**	1.00**	1.00**		0.97**
BLUP-III	0.76**	0.98**	0.98**	0.96**	0.98**	0.98**	0.98**	

with DFREML-I indicated that either of these two methods could possibly be used for evaluation and ranking of Red Sindhi sires with equal efficiency and accuracy. Similar product moment correlation was also reported by Singh and Singh (1999) as they have observed BLUP to be more efficient sire evaluation method than any other method in Murrah sire evaluation.

The high and significant rank correlations between the breeding values of sires estimated by different methods indicated that all these methods did not differ significantly in ranking of sires. The high relative efficiency of BLUP-I and BLUP-II (both 96.88 percent) in comparison to most efficient method along with high product moment correlation of BLUP-I and BLUP-II with DFREML-I indicated that either of these two methods could possibly be used for evaluation and ranking of Red Sindhi sires with equal efficiency and accuracy.

ACKNOWLEDGEMENT

The author is grateful to Joint Commissioner, Govt. of India, Ministry of Agriculture Department of Animal Husbandry, Dairying and Fisheries for allowing permission for collection of breeding data of Red Sindhi cattle from Central Cattle Breeding Farm, Chiplima, Orissa. And author is also thankfully to acknowledge to Director and other staff member of CCBF, Chiplima and CBF, Kalsi, Deheradun for providing a helping hand and necessary facilities during my collection of data.

REFERENCES

Arora KC. 1981. Relative efficiency of different methods of indexing the breeding work of dairy bulls. Thesis Abstract. Haryana Agricultural University 9: 36-37.

Aswathanaryana T, Govindaiah MG and Jayaprakash. 2003. Relative efficiency of various sire evaluation procedures. Indian Veterinary Journal 80: 550-53.

Atil H and Khattab AS. 2000. A comparison of different methods of estimating sires transmitting ability of some milk traits in a herd of Holstein-Friesian cattle. Archive fur Tierzucht 43: 115–22.

Banik S and Gandhi RS. 2006. Animal model versus conventional methods of sire evaluation in Sahiwal cattle. Asian-Australasian Journal of Animal Science 19: 1225-28.

Dahiya DS, Khana AS and Singh RP. 2005. Effectiveness of sire evaluation for milk yield with auxiliary traits in Hariana cattle. Indian Journal of Animal Sciences 75: 518-23.

Deb SM, Gurnani M and Chacko CT 1998. Comparison of sire evaluation methods under field conditions. Indian Journal of Dairy Science 51: 210-15.

Deulkar PB and Kothekar MD. 1999. Sire evaluation considering first lactation yield for improvement of lifetime production in Sahiwal. Indian J. Anim. Sci. 69: 240–42.

Gandhi RS and Gurnani M. 1991. Accuracy of different methods of sire evaluation in Sahiwal cattle. Indian Veterinary Journal 68: 659-62.

Gaur GK, Tripathi VN, Mukherjee S and Choudhary VK. 2001. Efficiency of sire evaluation procedures in Frieswal cattle. Indian Journal of Veterinary Research 10: 1-6.

Harvey WR. 1987. Least squares analysis of data with unequal subclass numbers. ARS H-4, USDA, Washington D.C.

Henderson CR. 1973. Sire evaluation and genetic trends. Proc. Anim. Breed and Genetics Symposium in honor of Dr. J.L. Lush, pp. 10–14. American Soc. Anim. Sci. Assoc. Champaign, Illinois.

Jain A and Sadana DK. 2000. Sire evaluation using animal model and conventional methods in Murrah buffaloes. Asian Australasian Journal of Animal Science 13: 1196-200.

Jain A. 1996. Sire evaluation under multiple trait models in Murrah buffaloes. Ph.D. Thesis, National Dairy Research Institute, Deemed University, Karnal, India.

Kishore G. 1993. Comparison of sire evaluation methods by progeny testing in field conditions of Kerala. M.Sc. Thesis, National Dairy Research Institute, Deemed University, Karnal, India.

Raheja K.L. 1992. Comparison of progeny testing of Sahiwal sires by the different methods of sire evaluation. Indian Journal of Dairy Science 45: 64–69.

Singh BP, Kumar V and Chauhan VPS. 1992. Comparison of different methods of sire evaluation. Indian Journal of Animal Sciences 62: 749–53.

Steel RGD and Torrie JH. 1981. Principle and procedures of Statistics. 2nd edition.

Tailor SP, Banerjee AK and Yadav SBS. 2000. Comparison of different methods of sire evaluation. Indian Journal of Animal Sciences 70: 73-74.

Tajane KR and Rai AV. 1990. Efficiency of sire evaluation methods to improve milk yield of Sahiwal x Holstein-Friesian cattle. Indian Journal of Animal Sciences 60: 183–91.

Vij PK and Tiwana MS. 1988. A note on evaluation of buffalo sires. Indian Journal of Dairy Science 41: 500–01.

Vinoo R, Rao GN, Gupta BR and Rao KB. 2005. Genetic study on productive and reproductive traits of Ongole cattle. Indian Journal of Animal Sciences 74: 438-41.

Phenotypic characterization of Jaunpuri goat of Uttar Pradesh

A K Patel, VK Singh*, Jaswant Singh, N Shyamsana Singh and M K Verma
College of Veterinary Science & A.H., N. D. University of Agri. & Tech.,Kumarganj, Faizabad, India

ABSTRACT

The present study was conducted in two district of eastern Uttar Pradesh viz. Faizabad and Jaunpur. Seven villages from Faizabad and eight villages from Jaunpur have been randomly selected. 17 farmers and 140 goats from Faizabad and 15 farmers and 143 goats from Jaunpur district were selected for present investigation. Data on growth performance and physical characteristics were recorded on 293 Jaunpuri goats of various age groups in its two native districts viz., Jaunpur and Faizabad of Eastern U.P. during 2009-20110. The average body weights of the age groups 0-3, 6-9 and 12-24 months were 4.89±1.02, 15.45±0.30 and 20.44±0.49 kg, respectively. In these respective age groups the mean values (cm) of body length were 32.69±1.85, 43.65±0.52 and 46.34±0.49; chest girth: 38.54±2.27, 55.67±0.77 and 60.91±0.58 and height: 37.69±1.66, 58.38±0.88 and 60.80±0.65 respectively. The findings indicated that Jaunpuri goat is a medium size goat breed and the roman nose is distinct trait of this population.

Keywods: Phenotypic traits, growth trait, Jaunpuri goat
***Corresponding author:** vsvsingh83@gmail.com
Manuscript received: 29.1.2017

INTRODUCTION

Jaunpuri (Jaffarabadi) goat, found in the eastern part of U.P. has been evolved by crossing the local nondescript goat with the Jamunapari breed of goat (Saraswat, 2001). Their home tracts lie in the belt between Ganga and Gomati rivers of eastern U.P. It is one of the neglected goat germplasm, which needs systematic evaluation and characterization of their performance status for its conservation, which otherwise may lead to degeneration of this important indigenous goats of the country. It is not yet considered as a well established goat breed of the country due to lack of scientific literature about this breed. Therefore, the present study was undertaken to evaluate the growth performance and physical characteristics of this goat germplasm in its native home tract..

MATERIAL AND METHODS

The present study was conducted to evaluate the growth performance viz., body weight, body length, chest girth, height and hind girth and physical characteristics of Jaunpuri goat under the field condition during 2009-10. Data on these traits were recorded on 293 animals of either sex belonging to different age groups in its two native districts viz., Jaunpur and Faizabad of Eastern U.P. The data collected were classified into different age groups: 0-3, 3-6, 6-9, 9-12, 12-24 and above 24 months and sex wise also for its further analysis. Data so classified was subjected to statistical analysis using standard formulae (Snedechor and Cochran, 1994).

RESULTS AND DISCUSSION

Body Weight

The average body weight of the Jaunpuri goats at various age groups in its home tracts (Jaunpur and Faizabad districts) are presented in Table 1. The average body weights of this goat of the age groups 0-

Jaunpuri Goat

Table 1. Mean ± SE of Body weight Body weight (kg) and biometry (cm) in Jaunpuri Goat

Age (m)	Sex	Jaunpur	Faizabad	Jaunpur	Faizabad
		Body weight		**Body length**	
0-3	M	5.637±1.475 (8)	-	32.562±2.810 (8)	-
	F	3.700±1.210 (5)	-	32.900±2.147 (5)	-
	P	4.892±1.021 (13)	-	32.692±1.851 (13)	-
3-6	M	12.143±0.822(7)	10.863±1.016 (11)	42.643±0.833 (7)	39.409±0.677 (11)
	F	12.194±0.937 (18)	10.627±0.913 (11)	43.333±0.874 (18)	38.363±0.675 (11)
	P	12.180±0.703 (25)	10.745±0.667	43.140±0.664 (25)	38.886±0.480 (22)
6-9	M	16.958±0.545 (12)	14.556±0.689(9)	46.500±0.931 (12)	41.111±0.739 (9)
	F	15.250±0.285 (12)	15.039±0.587 (18)	45.208±0.852 (12)	41.972±0.798 (18)
	P	16.104±0.349 (24)	14.878±0.448 (27)	45.854±0.632 (24)	41.668±0.583 (27)
9-12	M	17.833±0.881 (3)	19.700±1.044 (5)	47.167±1.691 (3)	47.800±1.594 (5)
	F	16.333±0.601 (3)	15.733±0.925 (6)	47.000±0.289 (3)	43.500±1.057 (6)
	P	17.083±0.583 (6)	17.536±0.906 (11)	47.083±0.768 (6)	45.455±1.106 (11)
12-24	M	22.938±0.803 (8)	19.906±1.222 (17)	50.831±1.003 (8)	46.912±1.276 (17)
	F	21.600±0.643 (20)	18.617±0.789 (17)	48.500±0.489 (20)	46.412±0.800 (17)
	P	21.982±0.525 (28)	19.262±0.725 (34)	49.161±0.496 (28)	46.662±0.743 (34)
>24	M	37.667±1.452 (3)	30.500±3.092 (4)	59.667±0.333 (3)	52.000±3.758 (4)
	F	25.625±0.387 (44)	24.635±1.230 (52)	52.436±0.595 (44)	50.029±0.484 (52)
	P	26.394±0.570 (47)	25.054±1.181 (56)	52.898±0.615 (47)	50.170±0.526 (56)
		Chest Girth		**Body Height**	
0-3	M	39.250±3.222 (8)	-	37.625±2.434 (8)	-
	F	37.400±2.947 (5)	-	37.800±2.189 (5)	-
	P	38.538±2.268 (13)	-	37.692±1.656 (13)	-
3-6	M	50.357±0.585 (7)	50.550 ±0.355 (11)	51.071±0.202 (7)	50.863±1.285 (11)
	F	51.444±1.207 (18)	49.090 ±1.122 (11)	51.694±1.232 (18)	51.955±0.808 (11)
	P	51.140 ±0.882 (25)	49.795 ±0.872 (22)	51.520±0.805 (25)	51.409±0.749 (22)
6-9	M	61.167 ±1.092 (12)	52.833 ±1.648 (9)	65.825 ±1.610 (12)	55.111 ±1.270 (9)
	F	56.667 ±1.029 (12)	52.750 ±1.155 (18)	58.417 ±1.418 (12)	55.028 ±1.023 (18)
	P	58.917 ±0.871 (24)	52.778 ±0.928 (27)	62.120 ±1.303 (24)	55.055 ±0.765 (27)
9-12	M	59.333 ±1.856 (3)	63.300 ±1.881 (5)	64.000 ±3.753 (3)	61.100 ±0.557 (5)
	F	63.500 ±0.764 (3)	58.250 ±1.063 (6)	63.167 ±0.440 (3)	55.917 ±0.898 (6)
	P	61.417 ±1.294 (6)	60.595 ±1.259 (11)	65.583 ±1.700 (6)	58.272 ±0.970 (11)
12-24	M	64.250 ±0.545 (8)	61.088 ±1.342 (17)	68.500 ±1.458 (8)	61.941 ±1.228 (17)
	F	64.800 ±0.766 (20)	61.294 ±1.170 (17)	65.850 ±0.789 (20)	60.941 ±1.224 (17)
	P	64.643 ±0.568 (28)	61.191 ±0.877 (34)	66.607 ±0.738 (28)	61.441 ±0.824 (34)
>24	M	71.833 ±0.441 (3)	67.375 ±2.087 (4)	73.167 ±0.600 (3)	68.125 ±2.695 (4)
	F	66.943 ±0.607 (44)	66.369 ±0.692 (52)	66.191 ±0.425 (44)	62.981 ±0.583 (52)
	P	67.255 ±0.595 (47)	66.441 ±0.660 (56)	66.636 ±0.471 (47)	62.277 ±1.051 (56)
		Hind Girth		**Ear Length**	
0-3	M	39.737±3.548 (8)	-	14.250 ±0.688 (8)	-
	F	38.500±3.421 (5)	-	13.440 ±0.654 (5)	-
	P	39.261±2.459 (13)	-	13.938 ±0.488 (13)	-
3-6	M	52.571±0.297 (7)	54.318±1.817 (11)	16.700 ±0.516 (7)	11.727 ±0.669 (11)
	F	52.278±1.289 (18)	50.045±1.121 (11)	17.183 ±0.407 (18)	14.545 ±0.896 (11)
	P	52.360±0.923 (25)	52.181±1.141 (22)	17.048 ±0.324 (25)	13.136 ±0.626 (22)
6-9	M	63.325±1.187 (12)	55.333±1.233 (9)	17.708±0.234 (12)	12.022±1.142 (9)
	F	56.667±1.450 (12)	54.972±1.179 (18)	18.375±0.365 (12)	13.833±0.879 (18)
	P	59.995±1.149 (24)	55.092±0.873 (27)	18.042±0.223 (24)	13.230±0.706 (27)
9-12	M	61.000±2.516 (3)	67.300±2.528 (5)	16.500±1.258 (3)	15.700±0.583 (5)
	F	66.000±0.764 (3)	61.833±0.749 (6)	19.000±0.287 (3)	14.583±0.987 (6)
	P	63.500±1.622 (6)	64.318±1.433 (11)	17.750±0.804 (6)	15.090±0.594 (11)
12-24	M	66.563±1.021 (8)	64.352±1.827 (17)	18.288±0.319 (8)	14.411±0.711 (17)
	F	66.350±0.577 (20)	64.147±1.104 (17)	17.050±0.340 (20)	14.894±0.733 (17)
	P	66.410±0.506 (28)	64.240±1.051 (34)	17.404±0.280 (28)	14.653±0.504 (34)
>24	M	78.000±1.000 (3)	71.750±3.100 (4)	18.000±0.287 (3)	18.125±0.372 (4)
	F	71.170±0.763 (44)	71.513±1.035 (52)	18.852±0.407 (44)	16.240±0.342 (52)
	P	71.606±0.757 (47)	71.500±0.986 (56)	18.798±0.382 (47)	16.375±0.361 (56)

Within paranetheses are number of observations.

3, 3-6, 6-9, 9-12, 12-24 and above 24 months were 4.89±1.02, 11.47±0.50, 15.45±0.30, 17.38±0.61, 20.44±0.49 and 25.67±0.69 kg, respectively. In general, the males had non-significantly higher average weights than the females at every age groups studied. The findings further indicated that the average body weights of goats of the two districts did not show any significant difference in any of the age groups, which shows the similarities in their genetic background. Similar body weights were also reported in Barbari goats (Singh *et al.* 1979b). However, the body weights of Jamunapari (Mishra and Khan, 1985 and Singh *et al.* 1979b), Beetal, Sirohi and Jhakarana (Mishra and Khan, 1985 and Chopra and Rana, 1995) were more than the present estimates. Whereas, Singh *et al.* (1979a) and Mukherjee *et al.* (1979) reported lower mean values of these traits in Black Bengal goats at the similar ages or age groups.

Body Length

The overall average body length of Jaunpuri goats of age groups 0-3, 3-6, 6-9, 9-12, 12-24 and above 24 months were 32.69±1.85, 41.09±0.53, 43.65±0.52, 46.03±0.77, 46.34±0.49 and 51.41±0.42 cm, respectively. In both districts, no sexual dimorphism was observed in the body lengths of this goat. The goats of the same age groups of the two districts did not show any significant differences in their body length. The present estimates of average body lengths of the Jaunpuri goat were comparable to that of Black Bengal (Mukherjee et al. 1979). However, the body lengths of Jamunapari and Jhakarana (Mishra and Khan, 1985), Beetal (Alam *et al*. 2009) and Barbari (Tiwari *et al*. 2002) goat breeds were more than the present estimates.

Chest Girth

In Jaunpuri goats, the overall average body chest girth of age groups 0-3, 3-6, 6-9, 9-12, 12-24 and above 24 months were 38.54±2.27, 50.50±0.64, 55.67±0.77, 60.85±0.91, 60.91±0.58 and 66.81±0.45 cm, respectively. In general, the males had non-significantly higher average chest girth than the females at the same age groups. The two districts did not show any significant difference in average chest girth of goats in any age groups. Tiwari *et al.* (2002) in Barbari goats and Mukherjee *et al*. (1979) in Black Bengal goats also reported similar mean values of chest girth. However, the present estimates in Jaunpuri goats were lower than that in Jamunapari and Jhakarana (Mishra and Khan, 1985) and Beetal (Alam *et al.* 2009).Barhat (2005) reported lower mean values of the chest girth in Marwari goats.

Body Height

The average height of Jaunpuri goats of the age groups 0-3, 3-6, 6-9, 9-12, 12-24 and above 24 months were 37.69±1.66, 51.47±0.56, 58.38±0.88, 60.15±1.05, 60.80±0.65 and 64.77±0.65 cm, respectively. In both districts, the two sexes did not show any significant differences in their heights except in 9-12 months age group of Faizabad district. Similarly, the average heights of the goats of the two districts did not show any significant differences, except for females of 9-12 months age group. The present estimates were comparable with that of Barbari goat (Mittal, 1979 and Tiwari *et al.* 2002). However, earlier workers (Mishra and Khan, 1985, Chopra and Rana, 1995 and Alam *et al.,* 2009) reported higher mean values in Jamunapri, Beetal and Jhakarana goat breeds. Mukherjee *et al.* (1979) observed lower mean value of these traits in Black Bengal goats.

Hind Girth

In Jaunpuri goats, the average hind girth were 39.26±2.46, 52.27±10.73, 57.40±0.78, 64.03±1.06, 63.75±0.63 and 71.57±0.63 cm, in the age groups 0-3, 3-6, 6-9, 9-12, 12-24 and above 24 months, respectively. The results revealed that the males had non-significantly higher average hind girth than the females at the same age groups. No significant difference was observed between the goats of the two districts in their average hind girth in any of the age groups. Singh *et al.* (1979a) reported similar mean values of these traits in Black Bengal goats also. However, the present estimates were lower than those in Beetal (Alam *et al.* 2009) and Sirohi (Mishra *et al.* 1980), but higher than that in Marwari (Barhat, 2005), breeds of goats.

Ear Length and orientation

The average ear lengths of Jaunpuri goats of the age groups 0-3, 3-6, 6-9, 9-12, 12-24 and above 24

months were 13.94±0.49, 15.17±0.45, 15.50±0.51, 16.03±0.56, 14.87±0.35 and 17.48±0.29 cm, respectively. The two sexes in both districts did not show any significant differences in their ear lengths in any of the age groups. Goats of the two districts did not show significant differences in their average ear length in most ages. However, the present estimates were lower than that in Jamunapari (Acharya, 1982) and Beetal (Alam *et al.* 2009), but higher than that in Black Bengal goats (Acharya, 1982). The present findings indicated that Jaunpuri goats has drooping ear (100 %) with long leafy shape.

Tail Length

The average tail length of Jaunpuri goats of the age groups 0-3, 3-6, 6-9, 9-12, 12-24 and above 24 months were 9.27±0.46, 11.68±0.26, 12.80±0.27, 12.94±0.49, 12.44±0.22 and 12 .67±0.17 cm, respectively. The two sexes in both districts did not show any significant differences in their tail lengths in any of the age groups. The goats of the two districts showed no significant differences in their average tail lengths in most of the age groups.

Horn Characteristics

The findings revealed that in both districts, most of goats (94.47 %) possessed horn, with straight shape (58.66 %) and backward pointed tip (73.85 %) thus, indicating that the Jaunpuri goats are horned type breed of goat.

Roman nose characteristics

The roman nose was present in 96.81 % of the Jaunpuri goats. As Jamunapari breed of goat was one of the foundation breed of the Jaunpuri goats, the genes responsible for this trait might have been inherited from Jamunapari goats, thus possessing this characteristics.

From the present findings, it may be suggested that the goats of the two districts studied might be of the similar strain of Jaunpuri goats. The findings also suggested that Jaunpuri goat is a medium size goat breed and the distinct traits of this goat is roman nose. ..

REFERENCES

Acharya RM. 1982. Sheep and Goat breeds in India, Animal Production and Health Pp:30 FAO Rome.

Alam S, Singh PK, Kaur G, Alam N and Khan BU. 2009. Technical characterization of Black Queen Beetal goat, the pride of Panjab.. Livestock International. Jan-Mar, pp: 15-19.

Barhat NK. 2005. Body weight and measurement in Marwari goats. Indian Journal of Animal Science 75 : 1323-1325.

Chopra SC and Rana ZS. 1995. Bher-bakariutpadan. Directorate of publication. Gandhi Bhawan, CCS Haryana Agri. Univ., Hissar

Mishra RK and Khan BU. 1985. New dimensions in goat production, Souvenir, VIII workshop on goats. CIRG, Makhdoom, Mathura, U.P.

Mishra RK, Arora CL and Acharya RM. 1980. Breed characterization of Sirohi goat. Indian Journal of Animal Science 50: 717-720.

Mittal JP. 1979. A study on birth weight of Barbari and Jamunapari kids. Indian Journal of Animal Sciences 49: 42-47.

Mukherjee DK, Singh CSP and Mishra HR. 1979. A note on phenotypic parameters in gray and Brown Bengal goats Indian Journal of Animal Sciences 49: 671-673.

Singh CSP, Mishra HR, Sharma BD, Mukherjee DK and Singh DK. 1979a. A note on body measurement of Black Bengal goat. Indian Journal of Animal Sciences 49: 669-670.

Singh CSP, Sharma BD, Mukherjee DK and Mishra HR.1979b. A note on some phenotypic parameters of Jamunapari and Barbari goats. Indian Journal of Animal Sciences 49: 762-764.

Saraswat BL. 2001. Evaluation and standardizing breed tract of Jaunpuri goats- An emerging goat breed of rural people. Final Research Report submitted to U.P. Council of Agriculture Research, Lucknow, Uttar Pradesh.

Snedecor GW and Cochran WG. 1994. Statistical Methods, 8th edn. Oxford and I.B.H. Publishing Co., New Delhi.

Tiwari HA, Sinha NK, Atul and Singh GD. 2002. Body growth in barbari male kids from birth to puberty and sexual maturity. Indian veterinary Medical Journal 26: 289-291.

Comparison of three types of Indian donkey populations based on morphometric characteristics

Rahul Behl*, S K Niranjan, Jyotsna Behl and R K Vijh
ICAR-National Bureau of Animal Genetic Resources, Karnal 132001, India

ABSTRACT

The three populations of Indian donkeys; Spiti donkeys distributed in Spiti and Yangthang regions of Himachal Pradesh, Sindhi donkey found in Barmer, Jaisalmer and Jodhpur districts of Rajsthan and Brown type donkeys found mainly in Kurnool and Anathapur districts of Andhra Pradesh were characterized and compared to ascertain whether any significant differences exist in these three populations of Indian donkeys in the studied morphometric parameters. The results of the F-test showed that these three populations to be significantly different ($P<0.01$) from each other, both in male and female animals, at almost all morphometric parameters studied. In pairwise comparison, the Sindhi donkeys showed significant differences from Spiti as well as Brown type donkeys of Andhra Pradesh in most of the studied morphometric parameters both in male and female animals. However, the Brown type donkeys of Andhra Pradesh showed significant differences from Spiti donkeys only in limited number of biometric parameters both in male and female animals.

Keywords: Morphometry, Spiti donkey, Sindhi donkey, Andhra donkey
***Corresponding Author**: behl1969@rediffmail.com
Manuscript received: 28.3.2017; Manuscript accepted: 20.5.2017

INTRODUCTION

Donkey is a hard working animal that is used mainly for transportation as a pack animal as well as in traction. As it is easy to rear and requires very little inputs, it has been traditionally associated with weaker sections of the society (Varshney and Gupta 1994, Behl *et al*. 2009). Although good amount of work has been done to characterize the animal genetic resources in most species of the livestock, only limited information is available on Indian donkey population. Recently National Bureau of Animal Genetic Resources, Karnal has initiated work on characterization of breeds or types of donkeys available in India and characterized three populations of donkeys; Spiti donkeys of Himachal Pradesh, Sindhi donkey of Rajsthan and Brown type donkeys of Andhra Pradesh (Behl *et al*., 2013, Behl *et al*. 2015, Behl *et al*. 2016). Spiti donkeys are found in Spiti and Yangthang regions of Himachal Pradesh at an altitude of 3200-4000 m. They are well adapted to hypoxic conditions and cold arid environment of this region. Sindhi donkeys are distributed in Barmer, Jaisalmer and Jodhpur districts of Rajsthan. These donkeys are well adapted to this desert region. The Brown type donkeys of Andra Pradesh are mainly found in Kurnool and Anathapur districts located in Rayalseema region. The present study was undertaken to compare these donkeys on the basis of morphometric traits to ascertain whether any significant differences exist in these three populations of Indian donkeys in the studied morphometric parameters.

METHODS

The F-test and students t-test on the data were performed as described in Gupta *et* al. (2011) and Norman and Steiner (2000).

RESULTS AND DISCUSSION

All these three populations of Sindhi, Spiti and Brown type donkeys of Andhra Pradesh are small sized donkeys. The heights at withers of these animals varies between 88.59±3.27 cm in spiti donkeys to 98.8±3.9 cm in Sindhi donkeys in males and 88.65±3.3 cm in Spiti donkeys to 97.9±4.9 cm in Sindhi donkeys in female animals. The detailed biometric parameters for these three populations are given in tables 1, 2 and 3, respectively.

To evaluate whether these three populations have

Table 1: Morphometric characteristics of Sindhi donkeys of Rajsthan

Parameter	Male		Female	
	Mean±SD (cm)	Range (cm)	Mean±SD (cm)	Range (cm)
Body length	93.05±5.02	84-103	93.4±6.45	82-105
Height at withers	98.8±3.9	89-109	97.93±4.9	87-105
Heart girth	104.3±5.35	95-118	106.52±5.97	95-116
Face length	46.5±3.22	39-52	45.77±3.1	40-52
Face width	17.5±1.19	15-20	17.34±1.5	15-21
Ear length	22.6±1.63	20-26	22.33±1.88	19.5-26
Neck length	34.2±3.18	29-40	34.06±3.95	24-46
Chest width	20.6±2.41	16-26	20.43±1.67	16-22
Tail length	52.1±4.42	43-65	51.14±4.56	43-62
Fore arm length (FL)	35.24±3.55	23-40	33.93±3.62	27-39
Canon length (FL)	19.7±1.42	16-23	19.0±1.38	16-22
Canon circumference (FL)	12.7±0.78	11-14.5	11.96±0.96	10.5-15
Pastern length (FL)	7.2±0.96	6-10	7.52±0.96	6-9
Pastern Circumference (FL)	12.98±1.20	11-17.5	12.15±0.61	11-13.5
Hoof length (FL)	5.67±0.60	4.5-7	5.96±0.62	5-7
Hoof circumference (FL)	22.45±1.29	19.5-25	21.7±1.34	19-24.5
Gaskin length (HL)	41.52±3.32	32-48	39.9±3.13	34-47
Canon length (HL)	28.39±1.51	26-32	27.33±1.62	24-30
Canon Circumference (HL)	13.3±0.84	11.5-15.5	12.7±0.75	11-14
Pastern length (HL)	7.0±0.69	6-8	7.0±0.71	6-8
Pastern circumference (HL)	13.42±1.07	12-16.5	12.57±0.99	10.5-14
Hoof length (HL)	5.59±0.63	4-7	5.77±0.53	5-7
Hoof circumference (HL)	21.51±1.28	19-25	20.86±1.26	18-24
Estimated body weight (kg)	84.95±10.12	68.66-108.37	89.54±14.57	68.89-118.51

FL-fore limb, HL-hind limb

Table 2: Morphometric characteristics of Spiti donkeys of Himachal Pradesh

Parameter	Male		Female	
	Mean±SD (cm)	Range (cm)	Mean±SD (cm)	Range (cm)
Body length	91.0 ± 2.88	86-97	90.96 ± 2.52	85-95
Height at withers	88.59 ± 3.27	84-95	88.65 ± 3.30	80-94
Heart girth	100.5 ± 5.02	90-115	98.58 ± 4.23	90-107
Neck length	31.10 ± 2.21	27-36	30.15 ± 1.52	28-33
Face length	32.10 ± 1.47	29-35	31.50 ± 1.03	30-34
Ear length	21.39 ± 1.13	19-23	21.50 ± 0.81	20-23
Tail length	54.21 ± 7.63	39-68	55.56 ± 9.26	37-70
Leg length (FL)	80.0 ± 3.46	76-87	81.00 ± 3.02	77-86
Canon circumference (FL)	12.04 ± 0.71	11-13	11.57 ± 0.70	10-13
Canon length (FL)	19.93 ± 0.84	18-21	19.96 ± 0.77	19-22
Pastern Circumference (FL)	11.51 ± 0.85	10-13	11.27 ± 0.78	10-13
Pastern length (FL)	8.66 ± 0.72	7-10	8.62 ± 0.57	8-10
Hoof length (FL)	5.83 ± 0.54	5-7	5.73 ± 0.45	5-6
Hoof circumference (FL)	20.88 ± 1.01	19-23	20.25 ± 1.03	18-22
Leg length (HL)	87.71 ± 2.93	84-91	87.50 ± 1.85	85-90
Canon Circumference (HL)	12.79 ± 0.85	11-14	12.70 ± 0.92	11-14
Canon length (HL)	26.10 ± 1.35	23-28	26.40 ± 1.00	24-28
Pastern circumference (HL)	12.36 ± 1.21	10-15	11.94 ± 1.61	10-15
Pastern length (HL)	8.68 ± 0.56	8-10	8.60 ± 0.65	8-10
Hoof length (HL)	5.53 ± 0.51	5-6	5.45 ± 0.50	5-6
Hoof circumference (HL)	20.67 ± 2.02	18-26	19.18 ± 1.13	17-21
Estimated body weight (Kg)	75.12±9.57	58.35-99.84	75.69±9.85	54.35-91.97

FL-fore limb, HL-hind limb

Table 3: Morphometric characteristics of brown type donkeys of Andhra Pradesh

Parameter	Male		Female	
	Mean±SD (cm)	Range (cm)	Mean±SD (cm)	Range (cm)
Height at wither	94.57±5.24	83-106	89.82±3.36	84-96
Body length	91.67±5.67	80-103	88.36±3.36	82-95
Heart girth	101.6±6.33	87-114	99.46±5.24	91-111
Paunch girth	102.4±7.84	90-126	101.43±7.45	89-116
Face length	41.19±3.72	35-52	38.54±1.32	36-42
Face width	14.45±2.21	12-20	13.07±0.47	12-14
Ear length	22.15±1.48	19-25	21.86±0.97	20-24
Neck length	30.93±2.92	27-39	29.82±1.18	28-32
Chest width	20.49±1.87	17-24	19.52±1.05	17-21
Tail length	57.39±4.89	41-66	56.11±3.52	49-65
Fore arm length	37.57±2.21	32-42	36.85±1.21	34-39
Fore arm circumference	20.64±1.44	18-24	18.25±1.35	16-20
Canon length (FL)	18.95±1.38	16-21	17.75±1.24	16-20
Canon circumference (FL)	12.48±1.09	10-15	11.79±0.74	10-13
Pastern length (FL)	7.38±0.79	6-9	7.07±0.54	6-8
Pastern circumference (FL)	12.33±1.03	11-15	11.32±0.77	10-13
Hoof length (FL)	6.04±0.77	4-8	5.61±0.57	5-7
Hoof circumference (FL)	22.12±1.66	20-26	19.93±1.44	17-23
Gaskin length	39.48±2.88	35-48	38.0±1.05	36-40
Gaskin circumference	23.9±2.02	20-29	22.0±1.66	19-25
Canon length (HL)	22.9±2.23	20-29	21.32±0.82	20-23
Canon circumference (HL)	13.24±1.09	11-16	12.11±0.92	10-14
Pastern length (HL)	7.41±0.87	5-9	7.32±0.61	6-8
Pastern circumference (HL)	12.9±1.01	11-15	11.64±0.83	10-13
Hoof length (HL)	5.8±0.86	5-10	5.19±0.39	5-6
Hoof circumference (HL)	21.58±1.43	19-25	19.5±1.27	17-22
Estimated body weight (kg)	80.14±14.21	53.97-109.01	73.69±9.87	56.96-98.18

FL-fore limb, HL-hind limb

Table 4: Comparison of Sindhi, Spiti and Brown type donkeys for Morphometric characteristics by F-test.

Parameter	Male	Female
Height at wither	**	**
Body length	**	**
Heart girth	**	**
Face length	**	**
Ear length	**	#
Neck length	**	**
Tail length	**	*
Canon length (FL)	**	**
Canon circumference (FL)	*	#
Pastern length (FL)	**	**
Pastern circumference (FL)	**	**
Hoof length (FL)	*	#
Hoof circumference (FL)	**	**
Canon length (HL)	**	**
Canon circumference (HL)	*	*
Pastern length (HL)	**	**
Pastern circumference (HL)	**	**
Hoof length (HL)	#	**
Hoof circumference (HL)	**	**
Estimated body weight (kg)	**	**

FL – fore limb, HL – hind limb, **P<0.01, *P<0.05, # Differences not significant

Table 5: Pairwise comparison of Sindhi, Spiti and Brown type donkeys for Morphometric characteristics by t-test

Parameter	Male			Female		
	Sindhi-Spiti	Sindhi-AP Brown	AP Brown-Spiti	Sindhi-Spiti	Sindhi-AP Brown	AP Brown-Spiti
Height at wither	**	**	**	**	**	#
Body length	#	#	#	#	**	**
Heart girth	**	*	#	**	**	#
Face length	**	**	**	**	**	**
Face width	dna	**	dna	dna	**	dna
Ear length	**	#	**	#	#	#
Neck length	**	**	#	**	**	#
Chest width	dna	#	dna	dna	*	dna
Tail length	#	**	#	*	**	#
Fore arm length	dna	**	dna	dna	**	dna
Canon length (FL)	#	*	**	**	**	**
Canon circumference (FL)	**	#	#	#	#	#
Pastern length (FL)	**	#	**	**	*	**
Pastern circumference (FL)	**	**	#	**	**	#
Hoof length (FL)	#	**	#	#	*	#
Hoof circumference (FL)	**	#	**	**	**	#
Gaskin length	#	**	dna	dna	**	dna
Canon length (HL)	**	**	**	*	**	**
Canon circumference (HL)	*	#	#	#	*	*
Pastern length (HL)	**	**	**	**	#	**
Pastern circumference (HL)	**	**	#	#	**	#
Hoof length (HL)	#	#	#	*	**	*
Hoof circumference (HL)	#	#	#	**	**	#
Estimated body weight (kg)	**	*	#	**	**	#

FL – fore limb, HL – hind limb, **P<0.01, *P<0.05, # Differences not significant, dna- data not available in one or both the breeds

significant differences in body biometric parameters, they were compared by analysis of variance (F-test) at the parameters for which data was available in all the three populations. The results of the F-test showed that these three populations to be significantly different (P<0.01) from each other, both in male and female animals, at almost all morphometric parameters studied (table 4). To have a deeper understanding, these three populations were also evaluated in pairwise comparison by students t-test (table 5). When Spiti donkeys were compared with Sindhi donkeys, both of which can be distinguished by general appearance of the animals as Spiti donkeys are covered with long hairs, also showed significant differences from each other in most body biometric parameters (table 5).

Similarly, Spiti donkeys can be distinguished from Brown type donkeys of Andhra Pradesh by general appearance of the animals. When these two populations were compared for morphometric parameters, they showed significant differences in height at wither, face length, canon length (fore and hind limb), pastern length (fore and hind limb) and ear length (P<0.01), when male animals were compared. However, when female Brown type donkeys of Andhra Pradesh and Spiti donkeys were compaed, they showed significant differences only in body length, face length, canon length (fore and hind limb), pastern length (fore and hind limb) with P<0.01 and canon circumference (hind limb), hoof length (hind limb) with P<0.05. When Sindhi donkeys and Brown type donkeys of Andhra Pradesh, which appear quite similar to each other, were evaluated, they showed significant differences in most body biometric parameters, both when male or female animals were compared (table 5).

These results indicate that the Sindhi donkeys showed significant differences from Spiti as well as Brown type donkeys of Andhra Pradesh in most of the studied morphometric parameters both in male and female animals. However, the Brown type donkeys of Andhra Pradesh showed significant differences from Spiti donkeys only in limited number of biometric parameters both in male and female animals.

ACKNOWLEDGEMENTS

We gratefully acknowledge the kind help received from the Animal Husbandry department of Himachal Pradesh, Rajsthan and Andhra Pradesh and Sri Venkteswera Veterinary University.

REFERENCES

Behl R, Behl J, Attri P N and Nadda S (2015) *Spiti Gadhe : Gun va upyogita*. *Pashudhan Prakash* 6: 19-21.

Behl R, Nirabjan S K, Vijh R K and Dharma Rao M V (2016) *Andhra Pradesh ke Bhuri kism ke gadhe*. *Pashudhan Prakash* 7: 14-16.

Behl R, Sadana D K, and Behl J (2009) Donkey – An underevaluated animal genetic resource of India. Livestock International 12: 5-8.

Behl R, Sadana D K, Behl J, Attri P N, Nadda S and Joshi B K (2011) Donkey genetic resources of India: Spiti donkey. National Bureau of Animal Genetic Resouces, Karnal Publication, monogram 74/2011.

Behl R, Sadana D K, Behl J, Kumar S, Kedar V and Joshi B K(2013) Donkey genetic resouces of India: Sindhi donkeys. National Bureau of Animal Genetic Resouces, Karnal Publication, monogram 80/2013.

Norman G R and Steiner D L (2000) Biostatistics – The Bare Essentials, 2nd edition. B C Decker Inc., Hamilton, London.

Gupta S P (2011) Statistical Methods, 41st edition. Sultan Chand and Sons, New Delhi.

Crossbred progeny production performance in rural areas of Western Maharashtra

S B Gokhale and R L Bhagat*
BAIF, Central Research Station, Uruli Kanchan, Pune-412 202, India

ABSTRACT

The network field progeny testing program sponsored by Indian Council of Agricultural Research (ICAR) is being operated in different parts of country and coordinated through Central Institute for Research on Cattle (CIRC), Meerut. Total 4,092 crossbred progeny (Holstein Friesian x Local) owned by 864 farmers spread over twenty six cattle breeding centres from Ahmednagar, Pune and Satara district of Western Maharashtra were reached to age at first calving (AFC) during the period of 20 years (1994 to 2013) and out of that 3,206 progeny completed their 1st lactation (2 died, 51 discontinued, 820 sold & 13 transferred). The performance of progeny was studied based on region (irrigated & unirrigated), participating units (BAIF, CIRC,GADVASU), sire dam's milk yield (4500, 4501-5500, 5501-6500, 6501-7500 & >7501 kg), season of calving (rainy-June to September, winter-October to January, summer-February to May), age at first calving (<649, 650-938, 939-1208, 1209-1478 & >1479 days) and progeny birth period (1994-97, 1998-01, 2002-05, 2006-09 & 2010-13). The 305 days milk yield was computed from the fortnightly milk yield records of alternate morning and evening milking. The first milk recording was got within fifteen days after calving and incomplete milk recording due to sale, transfer or death of progeny were excluded. The average first lactation 305 days milk yield of crossbred progeny under field conditions was recorded as 2974.95±12.03 kg and availability of irrigation facilities round the year, progeny calving season and birth period had significantly affected milk production levels of crossbred animals at field conditions.

Keywords: progeny, production performance, rural areas, Western Maharashtra
*** Corresponding author:** dr.bhagat@live.com
Manuscript received: 20.3.2017; Manuscript accepted: 02.6.2017

INTRODUCTION

As native cattle of Maharashtra state were very low in milk yield, exotic inheritance was introduced through massive crossbreeding programmes to enhance milk yield. This crossbreeding programme was very much flourished in Western Maharashtra compared to rest part of state due to factors like irrigation facilities, availability of green fodder, market for milk and net-work of cooperatives. The production performance of these crossbreed animals is affected by different factors like seasonal variations and management under field conditions. The performance of crossbred animals maintained at organized herds was evaluated and documented from time to time by various workers (Jadhav and Bhatnagar, 1984, Dalal et.al.,1991, Singh et.al.,2003, Avtar Singh,2005, Kumar et.al.2008), however, the information on performance of crossbreeds under field conditions is very limited especially from Maharashtra state. In this study the production performance of crossbred cattle maintained in farmers' herds participating in progeny testing programme of crossbred bulls taken up at BAIF Development Research Foundation, Urulikanchan is evaluated and the influence of different factors on the performance is investigated.

MATERIALS AND METHODS

The network field progeny testing program sponsored by Indian Council of Agricultural Research (ICAR) is being operated in different parts of country and coordinated through Central Institute for Research on Cattle (CIRC), Meerut. Along with CIRC, BAIF, Pune and Guru Angad Dev Veterinary and Animal Sciences University (GADVASU), Ludhiana contributed the bulls for test inseminations and progeny born out of these sires formed the data for

present investigation. Total 4,092 crossbred progeny (Holstein Friesian x Local) owned by 864 farmers spread over twenty six cattle breeding centres from Ahmednagar, Pune and Satara district of Western Maharashtra were reached to age at first calving (AFC) during the period of 20 years (1994 to 2013) and out of that 3,206 progeny completed their 1st lactation (2 died, 51 discontinued, 820 sold & 13 transferred). These animals were individually maintained and reared by the farmers. The housing of animals ranged from semi-permanent to permanent constructed sheds. All animals were stall fed with dry and green fodder along with concentrates. The performance of progeny was studied based on region (irrigated & unirrigated), participating units (BAIF, CIRC,GADVASU), sire dam's milk yield (4500, 4501-5500, 5501-6500, 6501-7500 & >7501 kg), season of calving (rainy-June to September, winter-October to January, summer-February to May), age at first calving (<649, 650-938, 939-1208, 1209-1478 & >1479 days) and progeny birth period (1994-97, 1998-01, 2002-05, 2006-09 & 2010-13). The 305 days milk yield was computed from the fortnightly milk yield records of alternate morning and evening milking. The milk recoding was done for ten months by employing contract milk recorders and supervised by permanent employees. The first milk recording was got within fifteen days after calving. Incomplete milk recording due to sale, transfer or death of progeny were not included. The data was analyzed using 'R' software and statistical methods suggested by Snedecor and Cochran (1968) taking different factors under study as fixed effects.

RESULTS AND DISCUSSION

The results on the production performance of crossbred progeny based on different factors have been presented in Table 1. The average first lactation 305 days milk yield of crossbred progeny under field conditions was recorded as 2974.95±12.03 kg. The present estimate was more than the results (2955.78±26.76kg) obtained by Gokhale et.al.

Table 1. Production performance of crossbred progeny according to different factors.

#	Source of variation	Particulars	No. of Observations	1st lactation 305days milk yield (kg)	Percent observations
1	Region* *	Irrigated	1542	3120.55±16.45[a]	48.10
		Unirrigated	1664	2840.02±16.79[b]	51.90
2	Participating units	BAIF	2409	2982.06±14.55	75.14
		CIRC	443	2961.21±26.75	13.82
		GADVASU	354	2943.77±30.65	11.04
3	Sire dam's milk yield (kg)	4500	247	2916.64±49.52	7.70
		4501-5500	1349	2985.07±19.10	42.08
		5501-6500	932	2933.77±22.83	29.07
		6501-7500	468	3017.35±26.48	14.60
		>7501	210	3066.76±39.24	6.55
4	Calving Season* *	Rainy	938	3014.30±21.87[a]	29.26
		Summer	1345	2981.20±19.32[ab]	41.95
		Winter	923	2925.83±21.34[b]	28.79
5	Age at first calving (days)	<649	35	2919.62±68.84	1.09
		650-938	1013	2966.74±20.70	31.60
		939-1208	2083	2976.83±15.27	64.97
		1209-1478	63	3057.24±78.59	1.97
		>1479	12	3071.04±202.10	0.37
6	Progeny birth period* *	1994-97	600	2925.15±35.69[a]	18.71
		1998-01	872	2910.91±25.73[a]	27.20
		2002-05	450	2927.36±30.03[a]	14.04
		2006-09	697	3001.42±18.78[b]	21.74
		2010-13	587	3126.04±21.09[c]	18.31
Grand Total		3206		2974.95±12.03	100.00

Averages of similar superscripts in column did not differ from each other

(2007) and quite higher than the average (2677.56±22.12 kg) reported by Gokhale and Mangurkar (1995) for crossbred in Maharashtra. The average first lactation milk yield for crossbred cattle under Indian conditions were 3009.33±31.28 kg for Friesian × Hariana (Dalal et al.,1991), for Zebu × European cattle as 2486.24±80.26 kg (Singh et. al., 2003), for Friesian × Tharparkar as 3505.20±59.86 kg (Jadhav and Bhatnagar, 1984) and 2871.11±32.64 kg in Frieswal (Kumar et.al., 2008). The performance of progeny was evaluated on following aspects;

Region

The status of region (irrigated vs. unirrigated) exhibited significant effect on progeny production performance. Almost similar numbers of progeny were born in both regions and crossbreed animals from irrigated region recorded significantly higher production (3120.55±16.45kg) compared to those from unirrigated region (2840.02±16.79). The better performance in irrigated region was apparently attributed to availability of green fodder, supply of adequate ration and effluent condition of farmers resulting overall better management of animals.

Participating units

Under field progeny testing programme each bull batch comprised of bulls from BAIF Development Research Foundation (BAIF) UruliKanchan, Central Institute for Research on Cattle (CIRC), Meerut and Guru Angad Dev Veterinary and Animal Sciences University (GADVASU), Ludhiana. Out of total progeny under study more than three forth (75.14%) progeny were born by BAIF sires semen and remaining were from CIRC and GADVASU bulls' semen. Although the progeny born out of BAIF sires' semen performed higher compared to progeny born out of sires used from other participating units the results were non-significant, which indicated non influence of bulls from progeny production point views.

Sire dam's milk yield

The technical programme envisaged testing of 30 Frieswal bulls in each batch having dam's mature equivalent (ME) milk yield of minimum 4500 kg or sons of progeny tested sires with index of 9000 kg or more without regard to dam's milk yield. During the study period total 186 bulls were put under test inseminations and they were divided into five groups depending on respective bull's dam milk yield. It was noticed that 71.15 per cent progeny were born from the sires having dam's milk yield ranging from 4501 to 6500 kg. In general it was observed that there was increase in progeny performance as sire dam's milk yield increased but it was not statistically significant.

Season of calving

Out of total 3,206 progeny under study, 41.95 per cent progeny were calved during summer season (February to May), 29.26 per cent in rainy season (June to September), and 28.79 per cent in winter season (October to January). The production performance of progeny calved in rainy season noticed to be significantly higher (3014.30±21.87 kg), followed by summer season calved progeny (2981.20±19.32 kg) and winter season calvers (2925.83±21.31 kg), whereas Sahana and Gurnani (2000) observed that animals calved during rainy season recorded significantly lower milk yield in (Friesian x Zebu) Karan Fries cattle maintained at National Dairy Research Institute, Karnal. The variation in performance of animals calved during different seasons may be attributed to availability of feeds and fodder and also the climatic factors in different seasons. Dubey and Singh (2005) and Kumar et. al. (2008) reported non-significant effect of season of calving on first lactation milk yield.

Age at first calving

Age at first calving is indication of overall management of animals and always efforts was done to achieve the optimum age so that life time production of animals could be achieved satisfactory. Under the present study 96.57 per cent progeny attained their first calving between 650 to1208 days from birth. It was noticed that late attainment of age at first calving helped to increase the production performance but it was not statistically significant and per cent progeny taking more days for reaching the first calving was also negligible. Rao et. al. (2000) also reported non-significant effect of age at first calving on first lactation milk yield in crossbred cattle under field conditions.

Progeny birth period

The progeny under study was born over a period of 20 years from 1994 to 2013 and this birth period was divided into five groups. It was noticed that period of progeny birth had significant effect on production performance and there was improvement over the period, which might be attributed to inclusion of bull batch having higher dam's milk yield, farmers overall awareness and culling of those animals not performing as per the expectation of farmers. Singh et. al. (2005) also noticed progeny birth period had significantly affected performance of Sahiwal x Holstein Friesian crossbred animals at organized farm.

The variability in the performance of crossbred animals under field conditions could be ascribed to differential availability of inputs, agro-ecological conditions, type of farmer and the indigenous and exotic breeds used in crossbreeding (Avtar Singh, 2005). Further the present study on different attributes associated to production performance of crossbred progeny showed that availability of irrigation facilities round the year, progeny calving season and birth period had significant effect on milk production levels of crossbred animals at field conditions.

ACKNOWLEDGEMENT

The financial assistance provided by the Indian Council of Agriculture Research through Central Institute for Research on Cattle (CIRC), Meerut is highly acknowledged.

REFERENCES

Dalal DS, Arora KC and Rathi SS. 1991. Evaluation of performance traits in half bred cattle. Indian Journal of Dairy Science 44:401-405.

Dubey PP and Singh CV. 2005. Estimate of genetic and phenotypic parameters considering first lactation and lifetime performance traits in Sahiwal and crossbred cattle. Indian Journa of Animal Research 75 (11): 1289-94

Gokhale SB and Mangurkar BR. 1995. Field recording and sire evaluation under Indian conditions. Indian J. Anim. Sciences 65: 203-207

Gokhale SB, Bhagat RL, Gokhale RB and Pande AB. 2007. Farmer attributes affecting crossbred cows production performance in rural areas of Western Maharashtra. International Journal of Tropical Agriculture 25: 605-608

Jadhav KL and Bhatnagar DS. 1984. Note on 305 days yield of milk, fat corrected milk and fat in different crossbred cows. Asian Journal of Dairy Research 3:231-234.

Kumar S, Singh YP and Kumar D. 2008. Genetic studies on performance traits in Frieswal cattle. Indian Journal of Animal Sciences 78: 107-110

Rao MK, Reddy Obi A and Bhaskar BV. 2000. Performance of crossbred dairy cattle in farmers' herds. Indian Journal of Animal Sciences 70: 608-612.

Sahana G and Gurnani M. 2000. Performance of crossbred cattle and comparison of sires evaluation methods under organized farm condition. Indian J. Anim. Sciences 70 (4): 409-414

Singh A. 2005. Crossbreeding of cattle for increasing milk production in India: A review. Indian Journal of Animal Sciences 75:383-390.

Singh D, Yadav AS and Dhaka S S. 2003. Genetic and Phenotypic Evaluation of Milk and Fat Production Traits and Their Interrelationship in (Zebu × European) Crossbred Cattle Using Parent Group Mixed Model. Asian-Australasian Journal of Animal Science 16 : 1242-1246.

Singh VK, Singh CV and Kumar D. 2005. Genetic analysis of milk producing efficiency traits in crossbred cattle. Indian Journal of Animal Sciences 75: 37-40

Snedecor GW and Cochran WG. 1968. Statistical Methods. 6th edition Iowa State University press, USA

Husbandry and traditional practices in field flocks of Madras Red sheep

R Venkataramanan*, V Arthy , C Sreekumar, G Manonnmani and H Gopi
Postgraduate Research Institute in Animal Sciences, Kattupakkam – 603 203, India

ABSTRACT

A survey was conducted on the husbandry and traditional practices followed in 107 Madras Red sheep flocks belonging to beneficiaries of ICAR-Network Project on Sheep Improvement-Madras Red field unit (NWPSI). The flocks were maintained under extensive system of management, with flock size ranging from 25 to 180 and majority having between 30-60 breeding females. More than half of the farmers surveyed (52.4%) housed their flocks in thatched sheds, while the remaining left their animals in open areas fenced with chain-link or thorny bushes. Several farmers (68.2%) owned agricultural land. Natural mating was the norm, with an average sex ratio (ram/ewe) of 1 ram per 38 ewes (2.64%). Identification of animals was practised in some flocks using paint or ear notching. Aged ewes were usually disposed by selling to the butcher, with very few farmers maintaining them until death. The main source of income was from sale of ram lambs through middlemen. In addition to sheep, farmers also reared other livestock such as goats, buffaloes, cows and poultry species or a combination of these. The water sources for the animals were lakes, bore wells, ponds and wells. None of the farmers maintained records, insured their animals or cultivated fodder. Traditional treatment practices were followed by the majority of the farmers. Scientific management practices were followed by all the farmers as the inputs were provided by the NWPSI. Superstitious beliefs still exists among the farmers. Shrinking of grazing area, disease, predation by feral dogs and safety for the women grazers were some of the problems encountered.

Keywords: Management; Extensive system; Sheep; Reproduction
***Corresponding author:** venkyvet@gmail.com
Manuscript received: 28.3.2017; Manuscript accepted: 11.5.2017

INTRODUCTION

The total sheep population in India is 65.06 million, of which Tamil Nadu has 4.79 million sheep (19th livestock census, 2012). Tamil Nadu is home of ten recognized sheep breeds (Ganesakale and Rathnasabapathy, 1973; Acharya, 1982), of which Madras Red sheep is a medium-sized hairy sheep well adapted to the agro-climatic conditions of the North-Eastern region of Tamil Nadu (Acharya, 1982). It is a meat type breed mainly distributed in Thiruvallur, Kancheepuram, Chennai, Vellore and Villupuram districts of the state (Raman *et al.,* 2003). They serve as a source of livelihood for several small, marginal and landless farmers. It is important to study the actual husbandry and traditional practices followed by the farmers in field conditions. Such information will help in formulation of breed improvement programmes and suggesting suitable scientific managemental practices.

MATERIALS AND METHODS

Information on 107 beneficiary flocks under the ICAR funded Network Project on Sheep Improvement-Madras Red field unit (NWPSI), functioning at the Postgraduate Research Institute in Animal Sciences (PGRIAS), Kattupakkam was utilised for the study. Information was available for the period from 2015 – 2017.

Data on housing pattern, type of mating, management practices, flock size, sex ratio, time of grazing, type of grazing, additional feeding practices, landholdings, other livestock maintained, traditional practices followed for treatment and identification of animals, scientific management practices followed, disposal of adult females, mode of disposal of ram lambs, insurance, fodder cultivation, record maintenance, management of orphan lambs, source

of water and superstitious beliefs were recorded from the beneficiary flocks. The trend in the flock size over the months was also studied. Statistical analysis was done as proportions for various parameters.

RESULTS AND DISCUSSION

The details of parameters obtained through survey are presented in Table 1. All the farmers followed extensive system of management. Sheep were solely dependent on grazing (in natural range lands during rainy season and harvested fields during summer).Most of the indigenous breeds of sheep are maintained under extensive system of management (Devendran *et al.,* 2010; Balasubramanyam *et al*., 2012).

The flock size in the present investigation ranged from 25 to 180 with an average of 53 breedable females. According to Rao *et al.* (2008), in a study carried out in Chittoor district of Andhra Pradesh, the flock size of Nellore sheep in field conditions ranged from 10 to 183, with majority of the farmers having a flock size ranging between 25 to 50 (50%). In stationary flocks of Coimbatore sheep, the flock size ranged from 40 to 120 with an average of 60 (Devendran *et al.*, 2010).The Dhangar pastoralists of Maharashtra maintained a variety of livestock in herd along with sheep and the flock size ranged between 15 to 400 sheep (Patil *et al.,* 2012). The trend of flock size including adult females and lambs (Figure 1) showed an increase from September, reached maximum during May and decreased to

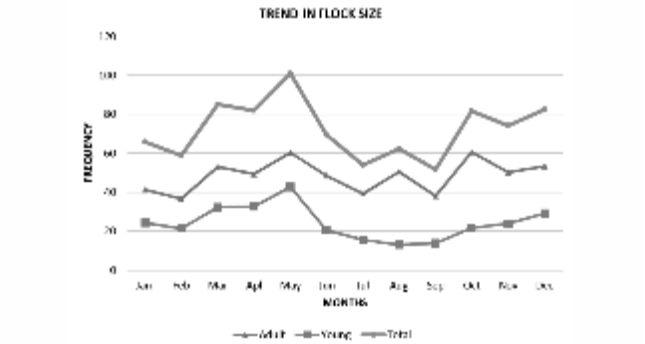


Figure 1. Flock size over the months

Table 1. Management practices in field flocks of Madras Red sheep

Land Holding (%)				
	Land holding	No land holding		
	68.2	31.8		
Grazing Time (%)				
	Eight hours	Six hours		
	66.4	33.6		
Nomadic System (%)				
	Stationary	Nomadic		
	86.9	13.1		
Housing (%)				
	Thatched shed	In open		
	52.4	47.6		
Identification (%)				
	None	Paint	Ear notching	
	86	12.1	1.9	
Disposal of Aged Animals (%)				
	Sold	Maintained until death		
	98.1	1.9		
Mode of Sale of Ram Lambs (%)				
	Middlemen	Butcher	Slaughter	
	95.3	3.8	0.9	
Rearing of Other Livestock (%)				
	Goats	Cow	buffalo	Poultry
	78.4	53.8	27.7	22.4
Water Source (%)				
	Lakes	Bore wells	Ponds	Wells
	68.3	29.9	0.9	0.9

lowest during September. This was synchronous with the lambing season and marketing of ram lambs during festival seasons in the region. The main lambing season was during October to January and Bakrid, one of the important festivals for sale of ram lambs, falls during September.

More than half of the farmers surveyed housed their flocks in thatched sheds (52.4%), while the remaining left their animals in open areas fenced with chain-link or thorny bushes. Nearly 88% farmers in Chittoor district possessed kutcha house, a temporary house with mud walls and roof made up of tree leaves and other waste materials (Rao *et al.,* 2008). For Coimbatore sheep, housing was of open type with the side protection made up of wooden reapers or bamboos (Devendran *et al.*, 2010). No additional feeding was done for the Coimbatore sheep, even during periods of scarcity (Devendran *et al.*, 2010), which was also the case with Madras Red sheep. Nevertheless, 68.2% of farmers owned agricultural land and made use of the harvested field for grazing, while a few farmers fed crop residues like groundnut haulms to their animals during lean months of summer. Accordingly, grazing time in most of the flocks (66.4%) was from 12 noon to 6 PM. Farmers took the sheep for grazing after completion of the agricultural operations. The water source for the animals while grazing included lakes (68.2%), bore wells (29.9%), ponds (0.9%), and wells (0.9%).

Natural mating was the norm, with rams maintained in the flock throughout to exploit the advantages of the two breeding seasons to the maximum. In Chittoor district, only one fifth of the farmers were aware of ram rotation whereas this is not the case with the farmers of NWPSI, as it is mandatory to rotate the rams among the farmers once in two years. Sex ratio (ram/ewe) ranged from 0.59 to 9.09% and the average was found to be 2.64% which is one ram per 38 ewes. NWPSI recommends a sex ratio of one ram per 40 ewes in field flocks of sheep (Report, 2016). According to Vivanco (1985), during the normal ovulatory season and when the rams are allowed to remain permanently with the ewes, a sex ratio of one ram per 40 ewes was sufficient.

Identification of animals was practised in some flocks using paint (12.1%) or ear notching (1.9%). This was practiced by farmers to help in identification of animals which get inadvertently mixed with other flocks while grazing. Disposal of aged ewes was by selling to the butcher (98.1%), while some farmers (1.9%) maintained them until death. The main source of income was through sale of ram lambs, which was done through middlemen (95.3%) or directly to the butcher (3.7 %).Very few farmers (0.9 %) slaughtered the animals for sale of mutton themselves. Rao *et al.* (2008) concluded that farmers, to a greater extent, were exploited by middlemen or butchers. Hence, there is an urgent need to educate the sheep farmers and prevent them from exploitation. They recommended establishment of co-operative and contract farming as ways to come out of the clutches of middlemen.

Integration of other agricultural activities was noticed. In addition to sheep, farmers also reared other livestock such as goats (78.4%), cows (53.8%), buffaloes (27.7%), poultry species (22.4%) and a combination of these (67.2%). Rao and Saxena (1994) reported that a large proportion of livestock was raised under the mixed cropping system in Indian Himalaya. Sheep constituted 10.4% in mixed crop farming systems in the central and eastern Himalayas (Sati and Singh, 2010). Nomadic system of grazing was practised only by few farmers (13.1%), who shifted their flocks during months of scarcity in summer. Most of the Madras Red farmers maintained stationary flocks, as they owned agricultural lands and harvested fields were a good source for grazing during scarcity months. Seasonal migration of sheep from their native tracts to different places, where sufficient water and grazing is available, is an important feature of sheep management which is practiced for the other breeds of sheep from southern parts of Tamil Nadu (Singaravadivelan *et al.,* 2014). None of the farmers maintained records, insured their animals or cultivated fodder.

Traditional treatment practices like use of kumkum for cataract , hot iron for convulsions, camphor and neem oil for maggot wound, lard for oral lesions of foot and mouth disease, application of hot oil for dog bite wound, to name a few, were followed by farmers

in general. Some of these practices were beneficial, while most of them like use of hot iron, hot oil etc., for ailments result in loss due to death of animals. Scientific management practices like provision of salt licks, mineral mixture supplementation, hoof trimming, deworming, vaccination and spraying of drugs for ectoparasites was followed by all the farmers, as the inputs were provided by the NWPSI. These flocks were regularly dewormed and vaccinated by NWPSI. Rao *et al.* (2008) emphasised the importance of modern scientific practices in improving the production of sheep. Superstitious beliefs among the farmers included selling of twinning and early lambing ewes and maintaining one black coloured animal in the flock. Shrinking of grazing area, disease, predation by feral dogs and safety for the women grazers were some of the problems encountered by these sheep farmers. The information on the existing sheep husbandry scenario obtained through this study will help formulate the strategies for adoption of better husbandry practices. Assistance in marketing of sheep could help increase income for the farmers. Practices like record maintenance, insurance cover for animals and cultivation of fodder could further improve income from their sheep enterprises.

ACKNOWLEDGEMENT

The authors thank the ICAR- Network Project on Sheep Improvement for facilities provided to conduct the study.

REFERENCES

Ganesakale D and Rathnasabapathy V. 1973. Sheep breeds of Tamil Nadu. Cheiron 2: 146–55.

Acharya RM. 1982. Sheep and goat breeds of India. FAO Animal Production and Health Paper, No: 30, FAO, Rome, Italy.

Vivanco HW. 1985. Recent developments in reproductive techniques of sheep and goats, FAO.

Animal Production and Health Paper 58 Small ruminant production in the developing countries Proceedings of an Expert Consultation held in Sofia, Bulgaria, 8–12 July.

Rao KS and Saxena KG. 1994. Sustainable development and rehabilitation of degraded village lands in Himalaya. HIMVIKAS, Publication No. 8. G.B. Pant Institute of Himalayan Environment and Development, Almora, India. pp.51-52.

Raman KS, Sundararaman MN, Haribhaskar S and Ganesakale D. 2003. Biometrics and breed characteristics of Madras Red sheep. Indian Journal of Small Ruminants 9: 6–9.

Rao VST, Thammi Raju D and Ravindra Reddy Y. 2008. Adoption of sheep husbandry practices in Andhra Pradesh, India. Livestock Research for Rural Development 20: 114-118.

Devendran P, Kandasamy N, Panneerselvam S and Thiruvenkadan AK. 2010. Rearing environment and husbandry practices of Coimbatore sheep. Indian Journal of Animal Sciences 80: 470-472.

Sati VP and Singh RB. 2010. Prospects of sustainable livestock farming in the Uttarakhand Himalaya, India. Journal of Livestock Science 1: 9-16

Ministry of Agriculture Department of Animal Husbandry, Dairying and Fisheries, Government of India. 19th Livestock Census. 2012.

Patil DS, Meena HR, Tripathi H, Kumar S and Singh DP. 2012. Socio economic profile of sheep reared by Dhangar pastoralists of Maharashtra, India. Journal of Recent Advances in Agriculture 1: 84–91.

Balasubramanyam D, Raja TV, Kumarasamy P and Sivaselvam SN. 2012. Estimation of genetic parameters and trends for body weight traits in Madras Red sheep. Indian Journal of Small Ruminants. 173-179.

Singaravadivelan A, Kumaravelu N, Sivakumar T and Karthickeyan SMK. 2014. Pattern of sheep migration in southern Tamil Nadu. Indian Journal of Veterinary and Animal Sciences Research 43: 405-409.

Report. 2016. Annual Report 2015-16 of ICAR-NWPSI (Madras Red Field Unit).

Production and reproductive performance of crossbred cattle in Coastal Maharashtra

B G Desai[1]*, R S Yadav[2], Shalu Kumar[1], R G Burte[1]and A J Mayekar[1]
[1]*Dr. B.S. Konkan Krishi Vidyapeeth (Agricultural University) Dapoli – 415712, Dist. – Ratnagiri, Maharashtra*
[2]*Department of Livestock Production and Management, Haryana Agricultural University, Hisar - 125004, Haryana*

ABSTRACT

The study was conducted to assess the production and reproductive efficiency of crossbred cattle (Dangi × Red Sindhi × Jersey). A total of 270 crossbred cattle were selected and their information regarding production and reproduction performances were collected from farm records maintained at Cattle Breeding Farm, Nileli, Dist. Sindhudurg, Maharashtra for a period of 15 years (1980 to 1994). Data representing 270 crossbred cattle from 657 total records of productive and reproductive performances for a period of 15 years (1980 to 1996) were analyzed to determine, Persistency Index (PI), Standard Lactation Milk Yield (SLMY), Peak Yield (PY), Days to Attain Peak Yield (DAPY), Wet Average (WA), Lactation Length (LL), Dry Period (DP), Service Period (SP), Gestation Period (GP) and Calving Interval (CI). The overall least squares mean of persistency index (162.65±0.83), standard lactation milk yield (1394.97±10.50), peak yield (8.61±0.05), days to attain peak yield (42.35±0.43), wet average (4.75±0.03), lactation length (302.01±1.58), dry period (81.77±1.65), service period (107.55±2.09), gestation period (279.14±0.30) and calving interval (387.58±2.18), respectively. Therefore, it may be concluded that crossbred cattle (Dangi × Red Sindhi × Jersey) cattle give optimum production and reproductive performance under coastal zone of Maharashtra.

Keyword: Crossbred cattle, productive and reproductive traits.
***Corresponding author:** headahds@gmail.com
Manuscript received: 18.1.2017; Manuscript accepted: 09.5.2017

INTRODUCTION

Maharashtra is believed to have the second largest livestock population state in India. This livestock sector has been contributing considerable portion to the economy of the country and still promising to rally round the economic development of the country. The total cattle population for the state is estimated to be about 16.2 million. Out of this the indigenous cattle constitute about 13.1 million and the remaining 3.1 million are crossbred cattle and 80.86 per cent of the total cattle in the state are local breeds and remaining are crossbred and exotic breeds that accounted for about 19.13 per cent, respectively (Anonymous, 2015).

The native dairy cattle have low genetic potentials for milk production, mature late and have a delayed conception coupled with long calving intervals. But, have excellent qualities such as adaptability to hot climatic conditions, resistance to diseases and general thriftiness under inferior feeding and managemental conditions (Japheth *et al.,* 2015). Whereas, high producing specialised dairy breeds from temperate region when introduced to tropical and subtropical zones generally have a reduced performance in terms of productivity and reproduction which may be due to lack of adaptability to hot climatic conditions and susceptible to tropical diseases. Under such circumstances (Thomas *et al.,* 2012), the productivity of indigenous animals could be increased by crossbreeding the low yielding nondescript cows with high yielding suitable exotic breeds for high milk yield, early sexual maturity and with sufficient capacity to withstand the direct and adverse effects of tropical climatic conditions.

The reproductive performance of breeding female is probably the single most important factor that is a prerequisite for sustainable dairy production system

Table 1. Nutrients composition of concentrate mixture.

Sr. No.	Concentrate mixture	Dry matter (%)	Crude protein (%)	Total digestible nutrients (%)
1.	Milch animals (Milk ration)	90.00	20.00	72.00
2.	Dry stock (Dry stock ration)	90.00	15.00	70.00

and influencing the productivity. Number of services per conception, days opens till conception and calving interval are important reproductive traits which are crucial for determining the profitability of dairy production (Alemayehu *et al.,* 2014).

Age at 1st service (AFS), age at 1st calving (AFC), birth weight (BW), total milk yield (TMY), average milk yield per day (AMYD), calving to 1st service interval (CSI) and calving interval (CI) are the important parameters that determine cattle productive and reproductive efficiency and these are important factors in terms of economics of dairy management (Dematawawa and Beger, 1998). Therefore, the objectives of the present investigation were to evaluate the productive and reproductive performance of crossbred cattle in coastal zone of Maharashtra, India.

MATERIALS AND METHODS

The data of 657 production and reproduction performance observations viz.,Persistency Index (PI), Standard Lactation Milk Yield (SLMY), peak yield (PY), days to attain peak yield (DAPY), wet average (WA), lactation length (LL), dry period (DP), service period (SP), gestation period (GP) and calving interval (CI) of crossbred cattle (Dangi × Red Sindhi × Jersey) generated at Cattle Breeding Farm, Nileli, Dist. Sindhudurga, Maharashtra in jurisdiction of the Dr. B. S. Konkan Krishi Vidyapeeth, Dapoli, Dist. Ratnagiri, Maharashtra State in India for the period of 15 years (1980 to 1996).The standard uniform feeding and management practices were provided throughout the experimental period to all the animals. All the animals were maintained under semi-intensive feeding system. Cows were fed with green and dry fodder by *ad libitum*. Concentrate mixtures were provided as per body requirements at rate 2 kg per cow per day for maintenance ration and 0.5 kg concentrate for per kg of milk at milking time. The feeds along with their nutrient content provided to the herd during experimental period in different seasons are given in Table. 1 and 2. Seasonal proximate analysis for estimating nutrient content was done by methods of AOAC (1980). All 657 observations of productive and reproductive performance were divided into persistency index (PI), standard lactation milk yield (SLMY), peak yield (PY), days to attain peak yield (DAPY), wet average (WA), lactation length (LL), dry period (DP), service period (SP), gestation period (GP) and calving interval (CI).

RESULTS AND DISCUSSION

The overall performance of dairy cattle is judged by the production and reproduction traits and these

Table 2. Composition of different fodders offered during study period.

Sr. No.	Fodders	Dry matter (%)	Crude protein (%)	Total digestible nutrients (%)
A.	*Green fodders*			
1.	Elephant grass	28.20	6.80	54.76
2.	Para grass	26.35	9.40	56.20
3.	NB -21	27.58	9.81	58.30
4.	Maize	25.00	8.17	68.00
5.	Cow pea	19.34	17.32	58.95
6.	Stylo	32.00	16.53	53.75
7.	Subabhul	20.60	25.80	51.55
8.	Rice bean	20.40	18.37	54.90
9.	Local grass	26.93	3.92	42.72
B.	*Dry fodders*			
1.	Local dry grass	87.00	3.05	45.20
2.	Maize stover	85.00	4.10	48.00

Table 3. Overall mean for persistency, production and fertility performance of crossbred cattle.

Sr. No.	Traits	Least-squares Mean±SE	N
Production performance			
1.	Persistency index	162.65±0.83	657
2.	Standard lactation milk yield (kg)	1394.97±10.50	657
3.	Peak milk yield (kg)	8.61±0.05	657
4.	Days to attain peak yield	42.35±0.43	657
5.	Wet average (kg/day)	4.75±0.03	657
6.	Lactation length (days)	302.01±1.58	657
7.	Dry period (days)*	81.77±1.65	471
Reproductive performance			
1.	Service period (days)*	107.55±2.09	471
2.	Gestation period (days)	279.14±0.30	657
3.	Calving interval (days)	387.58±2.18	657

serve as the economic indicator in evaluation of milch animals. The overall persistency, production and fertility performance of crossbred (*Dangi × Red Sindhi × Jersey*) cattle are present in Table 3.

Standard lactation milk yield (SLMY)

The overall mean standard lactation milk yield was 1394.97±10.50 kg. Similar finding were reported by Jalatge (1986) as 1397.00 kg in Sindhi × Jersey and Sharma *et al.* (1994) as 1434.92±62.60 kg in Friesian × Jersey × Ongole cattle breeds, respectively. Lower values for lactation milk yield was observed by Buvanendra and Mahadeven (1975) in Red Sindhi × Jersey cattle (1209.00 kg) and Sharma *et al.* (1994) in Jersey ×Friesian × Ongole crossbred cattle (1206.71±61.22 kg).However, higher values for lactation milk yield was observed by Pyne *et al.* (1988) in Jersey × Haryana cattle (1932.39±39.84 kg), Kumar *et al.* (1991) in Jersey × Ongole cattle (1823.00 kg) and Shelkar *et al.* (1992a) in Jersey × Red Kandhari cattle (1573.93±32.10 kg).

Peak milk yield (PMY)

The overall mean peak milk yield was 8.61±0.05 kg per day. Similar results have been reported in Dhawan *et al.* (2015) in Sahiwal cattle (7.94 ± 0.12 kg). Higher values for peak milk yield has been reported by Gahlot *et al*. (2000) in Tharparker cattle (12.780.10 litres), Kebede (2015) in Holstein-Friesian cattle (11.39±0.58 litre) and Nanavati and Singh (2004) in Gir cattle (10.05±0.10 kg per day), whereas lower values for PMY observed by Bangar and Narayankhedkar (1999[b]) in Gir cattle (7.46 kg).

Days to attain peak yield (DAPY)

The overall mean Days to attain peak yield (DAPY) was 42.35±0.43 days. These findings are in agreement with Tomer *et al.* (1997) in HF × Sahiwal half breed (43.89 ± 3.81 days) and Nanavati and Singh (2004) in Gir cattle (43.37±0.84 days). The higher value for DAPY was reported by Dhawan *et al.* (2015) in Sahiwal cattle (68.62 ± 0.96 days).

Wet average per day (WAPD)

The overall mean wet average was 4.75±0.031 kg per day. These results are in consonant with that of Jadhav (1982) and Bangar and Narayankhedkar (1999[a]) who has reported 4.16±1.24 kg per day and 4.60 kg per day, respectively in Gir cattle. Lower estimates for average WAPD (4.00±0.10 kg/day) have been reported by Sharma *et al.* (1972) and Narain and Garg (1972) who found overall mean for WAPD as 4.17±0.01 kg/day in Tharparkar cattle.

Lactation length (LL)

The overall mean lactation length was 302.01±1.58 days. These findings are in agreement with Sharma *et al.* (1994) who have reported 299.24±12.92 days in crossbred cattle. Lower value for LL was recorded by Dhawan *et al.* (2015) in Sahiwal cattle (295.33 ± 4.36 days), Haque *et al.* (2011) in crossbred cattle (291.49±29.30 days), Kabir and Islam (2009) in Holstein crossbred (295.0±33.96 days), Gahlot *et al*. (2000) in Tharparkaer cattle (285.272.20 days). The higher estimates for lactation length has been reported by Sandhu *et al.* (2011) in Holstein-Friesian cattle (314.19±0.91 days).

Dry period (DP)

The overall means and their standard error for DP was 81.77±1.65 days. Similar finding have been reported by Sandhu *et al.* (2011) in Gir cattle (87.06±1.63 days). Lower average DP (60.26-65.06 days) in Tharparkar exotic crosses have been reported by Thakur and Singh (2000). However, higher estimates for dry period were reported by Gaikwad *et al.* (2011) in Gir cattle (91-120 days) and Bhutkar *et al.* (2014) in Deoni cows (211.93±26.23 days).

Service period (SP): The average SP in crossbred cows was 107.55±2.09 days (Table 3). The present finding is nearer to the estimates reported by Vij *et al.* (1992) in Tharparkar cows (107.98 days). However, higher values for SP were reported by Pandit *et al.* (1999) who reported that 122.45±2.01 days in Gir cows and Sandhu *et al.* (2011) in Holstein-Friesian cattle (29.95±2.14 days). The variation in SP reported by different workers may be due to variation in the managemental efficiency in estrus detection and timely breeding followed in different herds (Savaliya *et al.,* 2016).

Gestation period (GP): The overall least squares mean for GP was 279.86±0.30 days (Table 3). This average value of GP in the present study is near to the estimates of first GP reported by Singh *et al.* (2012) as 279.8±0.69 days in Gir cattle. Similar value of first gestation period were also reported by Raja, (2010) in Sahiwal cattle, whereas the lower value reported by Mondal *et al.* (2005) as 275±4.11, 276±4.26, 274±4.41, 275±3.95 and 277±3.31 days with overall average 275±4.11 days in Jersey cross, Sahiwal cross, Sindhi cross, Holstein cross and Red-Chittagong cattle, respectively and greater value in Gir cattle reported by Gaikwad *et al.* (2011) as 284 to 286 days gestation period. The gestation length is a species characteristic. The variation of gestation length is genetically determined. Variation may be due to maternal influence. A little variation in gestation length within the individual may be contributed mainly by maternal and foetal factors. Age of dam, nutritional body condition of the dam are the maternal factors. On the other hand, foetal factors like the sex of the foetus, formation of twin and hormonal functions of the foetus. Environmental factors such as season, temperature, feeding and managementel (Mostari *et al.,* 2007).

Calving interval (CI): The average CI observed in present study, was 387.58±2.18 days (Table 3) which is near to the estimates of Khirari *et al.* (2014) in non-descript cows (381.23±3.27 days) and Manjusha *et al*. (2016) in crossbred cow (389.46±13.49 days). The estimate value was desirable for profitable milk production. This is because animals are low producers and hence are in the field for grazing along with other cattle and bulls for longer period. The animals in heat are covered naturally by the bulls in the field itself. The mean value was lower than the findings of Kumar *et al.* (2015) in Frieswal cattle (423.05±12.24 days), Kunbhar *et al.* (2015) in Red Sindhi cattle (413.050±10.362 days) and crossbred (372.200±7.486 days), Sandhu *et al.* (2011) in crossbred cattle (408.09±2.10 days) and Dandapat *et al.* (2010) in crossbred cattle (500.13±35.35days) and Sahiwal (522.63±27.99 days).

CONCLUSION

From the investigation, it was revealed that crossbred cattle (*Dangi × Red Sindhi × Jersey*) have large quite satisfactory values in case of lactation milk yield (1394.97±10.50 kg), peak yield (8.61±0.05 kg/day), lactation length (302.01±1.58 days) dry period (81.77±1.65 days), service period (107.55±2.09 days), calving interval (387.58±2.15 days), gestation period (279.14±0.30 days) with persistency index (162.65±0.8%), respectively. Therefore, it may be concluded that crossbred cattle (*Dangi × Red Sindhi × Jersey*) cattle can perform better in coastal zone of Maharashtra.

ACKNOWLEDGEMENT

The authors would like to acknowledge Officer In-charge, Cattle Breeding Farm, Nileli, Dist. Sindhudurga, Maharashtra, India to provide data for research work. Also would like to acknowledge Head of Department, Livestock Production and Management, Haryana Agricultural University, Hisar - 125004, Haryana, India for providing guidance during the work.

REFERENCES

Alemayehu T and Moges N. 2014. Study on reproductive performance of indigenous dairy

cows at small holder farm conditions in and around Maksegnit Town. Global Veterinaria.13 (4): 450-454.

Anonymous. 2015. Dairying in Maharashtra – A statistical profile 2015. National dairy Development Board.

AOAC. 1980. Association of Official Agriculture Chemists, Official Methods of Analysis, 14th Edn., Washington, D. C.

Bangar NP and Narayankhedkar SG. 1999a. Effect of sire and various non-genetic factors on total lactation milk yield in Gir cows. Cheiron 28(5): 168-170.

Bangar NP and Narayankhedkar SG. 199b. Study on lactation curve in Gir and its crosses with HF and Jersey. Dairy Science Abstract 61(9): 659.

Bhutkar SS, Thombre BM and Bainwad DV. 2014. Effect of non-genetic factors on production traits in Deoni cows. IOSR Journal of Agricultural Veterinary Science 7 (12 III): PP 09-14

Buvanendra V and Mahadeven P. 1975. Crossbreeding for milk production in Sri Lanka. World Animal Review 15:7-13.

Dandapat A, Banerjee D and Chakraborty D. 2010. Genetic studies on various production and reproduction traits of Sahiwal and crossbred cattle (HF ×Jersey × Sahiwal) of an organised farm. Veterinary World 3: 167-168.

Dematawewa CMB and Beger PJ. 1998. Genetic and phenotypic parameters for 305- day Yield, Fertility, and Survival in Holsteins. Journal of Dairy Science 81: 2700-2709.

Dhawan S, Yadav AS, Dhakal SS and Chakraborty D. 2015. Genetic studies on production and production efficiency traits in Sahiwal cattle. Indian Veterinary Journal 92 (9): 35–38.

Gahlot GC, Pant KP and Kachawaha RN. 2000. Variation in production performance of Tharparker cows in north-western Rajasthan. Indian Journal of Animal Sciences 70 (7): 718-722.

Gaikwad JS, Jadhav RM, Torawane KB and Todmal SB. 2011. Productive and reproductive performance of Gir cow in Akola district. Asian Journal of Animal Science 6: 94-96.

Haque MN, Haque MR, Parvin A and Hussain MM. 2011. Productive and reproductive performance of different crossbred cattle at Sylhet Govt. Dairy farm. Progressive Agriculture. 22(1 & 2): 47 – 54.

Jadhav PD. 1982. Comparative studies on some of the economic traits in Gir crosses with HF and Jersey. M.V.Sc. Thesis, KKV, Dapoli.

Jalatge EFA. 1986. Crossbreeding of Cattle in Sri Lanka. In 3rd World Congress in genetics Applied to Livestock Production. IX. Pp. 119-124.

Japheth KP, Mehla, RK, Imtiwati and Bhat SA 2015. Effect of non-genetic factors on various economic traits in Karan Fries crossbred cattle. Indian J. Dairy Sci. 68(2): 163-169.

Khirari PB, Bhambure CV, Bharambe VY and Kedari VC. 2014. Physical characteristics, productive and reproductive performance of non-descript cattle in Ratnagiri district of Konkan region, India. Livestock Research International 2: 33-35

Kumar J, Singh YP, Kumar S, Singh R, Kumar R and Kumar P. 2015. Genetic analysis of reproductive performance of Frieswal cattle at military farm, Ambala, Veterinary World. 8: 1032-1037.

Kumar VP, Rao CH, Venkatramaiah A and Naidu KN. 1991. Effect of change in genetic constitution from generation to generation in Ongole × Exotic crossbred cattle. Indian Journal of Dairy Science 44:54-57.

Kunbhar HK, Lasi AB and Memon AA. 2015. Reproductive performance of crossbred cattle under intensive management condition. Advanced Animal and Veterinary Science 3(5s): 7-13.

Mondal SC, Alam MM, Rashid MM, Ali MY and Hossain MM. 2005. Comparative study on the productive and reproductive performance of different dairy genotypes reared in Bangladesh Agricultural University dairy farm. Pakistan Journal of Nutrition 4: 222-225.

Mostari MP, Huque KS, Hasanat MS and Gulshan Z. 2007. Productive and reproductive efficiency of Red Chittagong cattle under farm condition. Prog. Agric. 18(2): 109-114.

Narain P and Garg LK. 1972. Estimation of genetic

change in Indian herds of cattle. Indian Journal of Animal Production 3:143-153.

Pandit RK, Agrawal RG, Shukla SP and Shrivastava OP. 1999. Reproductive and productive efficiency of crossbred Gir cow. Indian Journal of Veterinary Research 8: 33-44.

Pyne AK, Misra SK, Maitra DN, Dattagupta R and Majumdar SC. 1988. Reproductive and productive performance of Jersey Hariana half breds in humid tropical conditions of West Bengal. Indian Journal of Animal Production and Management 4:37-39.

Raja TV 2010. Part lactation records for Sahiwal sire evaluation. Ph.D. Thesis, NDRI, Karnal, India.

Sandhu ZS, Tariq MM, Baloch MH and Qaimkhani MA. 2011. Performance analysis of Holstein-Friesian cattle in intensive management at dairy farm Quetta, Balochistan, Pakistan. Pakistan Journal of Life and Social Science 9: 128-133.

Savaliya BD, Parikh SS, Gamit PM and Gajbhiye PU. 2016. Environmental factors affecting economic traits in Gir cattle. International Journal of Science and Environmental Technology 5:2467-2475.

Sharma GP, Reddy CE, Krishnaiah N, Shivaprasad L and Sri Ramamurthy A. 1994. Studies on productive and reproductive traits of triple cross Ongole breed. Indian Journal of Animal Sciences 15(2):128-140.

Shelkar BS, Sakhare PG and Deshpande KS.1992a. Breeding efficiency and milk producing ability of Red Kandhari and Jersey × Red Kandhari half-bred cows Indian Journal of Animal Sciences 62:79-80.

Singh G, Dutt G, Sharma RB, Singh SK, Fatima A and Chauhan SVS. 2012. An analytical study of reproductive performance in Gir Cows. Indian Research Journal of Extension Education (3), 203-206.

Thakur YP and Singh BP. 2000. Performance evaluation of Jersey × Zebu crossbreds involving different indigenous breeds (C) performance of Jersey × Hariana crossbred. Indian Veterinary Journal 77:127-130.

Thomas CK, Sastry NSR and Ravikiran G 2012. Dairy bovine production. Second revised edition.P-430.

Tomer AKS, Prasad KB, Verma SK and Bhadula SK. 1997. First lactation performance comparison between Holstein Friesian purebred and Sahiwal half-breds in Tarai region of Uttar Pradesh. Indian Journal of Animal Research 31(1):54-56.

Vij PK, Nivsarkar AE, Balaine DS and Raj B. 1992. Influence of inbreeding on performance of Tharparkar cattle. Indian Journal of Animal Sciences 62:689-691.

Analysis of sequence variability and expression pattern of lactoferrin gene in Sahiwal cows (*Bos indicus*) and Murrah buffaloes (*Bubalus bubalis*)

Ankita Sharma[1, 2], S K Swami[1], Manoj Kumar [1], Monika Sodhi[1], R S Kataria[1], Pranay Jain[2], Avnish Kumar Bhatia[1], A K Mohanty[3], S K Niranjan[1], U K Shandilya[1], Parvesh Kumari[1] and Manishi Mukesh[1]*
[1]*ICAR-National Bureau of Animal Genetic Resources, Karnal, Haryana, India*
[2]*University Institute of Engineering and Technology, Kurukshetra University, Kurukshetra, Haryana, India*
[3]*ICAR-National Dairy Research Institute, Karnal, Haryana , India*

ABSTRACT

Lactoferrin (LF), a bioactive glycoprotein is member of transferrin family and plays an important role in immune defense, iron homeostasis, antioxidant and regulation of cell growth. The present investigation was undertaken to analyze the sequence variations and expression pattern of LF gene between cattle and buffaloes. Blood was collected from Sahiwal cows (SAC), Karan Fries cows (KFC) and Murrah Buffaloes (MUB) to isolate peripheral blood mononuclear cells (PBMCs). Total RNA was extracted from PBMCs to synthesize cDNA. Sequence analysis has shown an ORF of 2127 bp with 780 amino acids in all cattle types and buffaloes. The analysis revealed no difference between LF cDNA sequences of SAC and KFC. Whereas, on comparing with exotic cattle, a total of 6 amino acid changes viz., I145V, S538T, T546N, T596S, K627E, and H632R were identified. The comparison between SAC and MUB revealed a total of 22 amino acid changes. The comparison of MUB LF cDNA sequences revealed homology of 96.23%, 96.75%, 91.67%, 91.95%, 73.16%, 70.20%, 64.21%, 73.83% with Indian native cattle, cross bred cattle, taurine cattle, sheep, goat, horse, human, mouse and pig, respectively. Additionally, expression pattern of LF mRNA in somatic cells from different lactation stages (colostrum early, mid and late lactation) of SAC and MUB was successfully evaluated. The analysis revealed highest transcript abundance of LF mRNA in colostrum samples indicating its role in enhancing immune system of new born calves. The study has led to sequence characterization LF cDNA in native, cross bred and riverine buffaloes and identified several variations that could prove important resource for future genotype:phenotype association studies. Further, the expression data has indicated that milk derived somatic cells could be utilized as valuable source to understand mammary gland functioning of Indian cows and buffaloes.

Keywords: Lactoferrin, Sahiwal cattle, Murrah buffaloes, SNP, somatic cells, expression pattern
***Corresponding author:** mmukesh_26@hotmail.com

INTRODUCTION

Lactoferrin (LF) an iron binding glycoprotein from the transferrin family, is synthesized by glandular epithelial cells and neutrophils (Plaffl et al. 2003). It is present in most of the biological fluids including milk, mucosal uterine fluid, saliva, tears, bile and pancreatic juice with most abundance in colostrum. It constitutes an important component of innate immune system and functions as the first line of defense against infectious micro-organisms. The main physiological function of LF is to bind iron that is basic requirement for microorganism's metabolism. The binding action impounds the necessary nutritional requirement for most bacterial pathogens (Gram-negative as well as Gram-positive bacteria), thereby inhibiting their growth. It is also involved in intracellular destruction of bacteria by inducing hydroxyl radical formation, which is catalyzed by iron (Fang and Oliver 1999). It modulates the inflammatory process by early recognition of invading microorganisms and enhance immunity by activating the transcription of important immune-related genes (Plaffl et al. 2003; Yamauchi et al. 2006).

Besides immunomodulatory, anti-inflammatory, antibacterial activity, LF is also known for its anti-viral (both naked and enveloped), anti-fungal, anti-parasitic, osteogenic and anti-cancerous potential (Gibbons et al. 2015, Duarte et al. 2011; Małaczewska and Rotkiewicz 2007, Gonzalez-Chavez et al. 2009). It also inhibits enteric absorption of iron in neonates and serves as a natural antioxidant (Detilleux, 2002). LF is also involved in modulation and regulation of macrophages, lymphocytes and neutrophil function (Smith and Oliver 1981; Sordillo et al. 1987) and hence help in prevention and control of mastitis in

cows (Seyfert and Kuhn 1994; Hirvonen et al. 1999; Teng, 2002). Thus LF, a multifunctional protein plays important role in health of mammary gland and could be a potential candidate gene against disease resistance. In bovines, LF gene spans 34.5 Kb of genomic region with 17 exons and 16 introns. It is single polypeptide chain of 708 amino acids with a molecular weight of 80 KDa (Seyfert and Kuhn, 1994). The polypeptide chain is folded into two symmetrical globular lobes N (amino) and C (carboxyl) terminals, containing one iron and two bicarbonate binding sites each (O'Halloran et al. 2009). The N lobe residue comprises of 1–333 amino acids while C-lobe comprises of 345–708 amino acids. The two lobes are connected by a peptide of residues 334–344 to forms a 3-turn α-helix (Berlutti et al. 2011).

In past, several studies have shown association of certain variants of LF gene with mastitis, somatic cell counts, uterine infections and reproduction trait in dairy cattle (Cao et al. 2011; Li et al. 2004; Hajibemani et al. 2012; Valadan et al. 2011; Wojdak-Makysmiec et al. 2006; Zupin et al. 2015).The association of different LF variants with health status might be attributed to the altered surface properties and iron chelating ability of lactoferrin (Mohammed et al, 2007). Several studies related to characterization of LF gene and its biological role have also been published in mouse, human (Teng et al, 1987; Teng and Gladwell 2006; Fine et al. 2013), pig (Wang 1998), exotic cattle (O'Halloran et al. 2009; Daly et al. 2006; Zhou et al. 2006) and several other species. However, similar systematic efforts are lacking for Indian cattle and buffaloes. Although, few isolated efforts (Raja et al. 2014; Kumari et al. 2014; Kathivaran et al. 2009) have been made in Indian cattle and buffaloes but these studies mainly focused on few exons or genotyping of certain alleles of LF gene. Further, data on comparative sequence variations of LF gene across native, cross-bred and exotic cattle and its expression pattern is also lacking. Hence, this particular study was planned to characterize the complete coding region of LF gene in two cattle types (native and cross-bred cattle) and compare its sequences with riverine buffaloes. Further, efforts were made to evaluate the expression pattern of LF mRNA in somatic cells across lactation stages of native cows and riverine buffaloes.

MATERIALS AND METHODS

Selection of animals, RNA isolation and cDNA synthesis

For sequence characterization of lactoferrin gene, blood samples were collected from 9 adult animals,3 each of Sahiwal cows (SAC), Karan Fries cows (KFC) and Murrah buffaloes (MUB)maintained at cattle farm of National Dairy Research Institute, Karnal. The blood samples were transported to laboratory for isolation of peripheral blood mononuclear cells (PBMCs). The PBMCs were isolated using density gradient centrifugation method by employing HiSep reagent (Himedia). Briefly, blood samples were diluted (1:1) with 1X PBS, and gently over laid on HiSep reagent. The mixture was centrifuged at 400g for 30min at room temperature. After removing the buffy coat, cells were treated with 2 ml chilled RBC lysis buffer and mixed gently with pasture pipette at room temperature for 10 minutes. The reaction was stopped by adding 8.0 ml of 1X PBS to remove the traces of HiSep and RBC lysis buffer followed by centrifugation at 260g for 10 min at room temperature. The supernatant was discarded to obtain a white pellet of PBMCs. The isolated PBMCs were washed twice with 1X PBS. Total RNA was extracted from 9 PBMC samples using Trizol reagent (Invitrogen Corp., CA). To remove the traces of genomic DNA, RNeasy Mini kit columns (Qiagen, Germany) along with on column digestion by RNAse free DNase enzyme (Qiagen, Germany) was used. The RNA quantity and quality was assessed using Nanovue (GE healthcare). RNA integrity was confirmed by denaturing agarose gel electrophoresis. First strand cDNA was synthesized using 1.5µg of purified RNA with Revert Aid First Strand cDNA Synthesis Kit (Fermentas, Thermo Scientific) following manufacturer's instructions. Briefly, the mixture containing RNA, 1 µl Oligo $dT_{(12–18)}$, 1 µl 10mM dNTP mix and 1 µl random primers, was incubated at 65°C for 5min and kept on ice for 3min. Further, a total of 6.0 µl of enzyme mix composed of 5X enzyme buffer, 1.0 µl M-MuLV RT (Fermentas, Thermo Scientific) and 1.0 µl of RNase inhibitor was

Table 1. Checking of contaminations in the RNA isolated from spermatozoa

Primer	Sequence	Purpose
LF1F	GTCCCATGGCCCCGAGGAAAAACGTTCGATGGTGTA	Amplification
LF1R	ACGTGCACCCCTCGTCAGGAAGGCGCAG	
LF2In-1	GGAATCCTTCGCCCGTACTT	Sequencing
LF3In-2	AGGCGCAGGAGAAATTTGGA	Sequencing
LF4In-3	CCTGGCAGAGAACCGGAAAT	Sequencing

added in the reaction. The reaction was performed in an Eppendorf Gradient cycler using the program as 25°C for 5 min, 50°C for 60 min and 70°C for 15 min.

PCR amplification, sequencing and analyzing sequence data

To amplify 2.1 Kb coding region of lactoferrin gene, specific primers were designed using Primer3 NCBI tool. The LF cDNA was amplified using primers designed based on *Bos taurus* sequence (Acc No.L08604.1). The description about the primers used for amplification and sequencing are given in Table 1. The amplification of LF cDNA was performed in 25 µl reaction containing cDNA as template, 10 pmol of forward and reverse primers, 10mM of dNTPs (Invitrogen Corp., CA), 1.0 unit of Taq DNA polymerase (Fermentas, Thermo Scientific) and 5.0µl of 5X reaction buffer. The thermal cycle conditions used were as follows: initial denaturation at 95°C for 2 min 30 sec, 32 cycles at 94°C for 45 sec, 64°C for 45 sec and extension at 72°C for 1.0 min, followed by final extension at 72°C for 10 min. PCR amplified product was visualized on 1.5% agarose gel. After purification, the PCR products were sequenced using BigDye Terminator Cycle Sequencing kit (Applied Biosystems). The chromatogram of each sequence obtained was checked manually. Sequences base calling was performed with Phred available in the suite Codon code Aligner v. 3.5.1. (Codon Code Corp., Dedham, USA). Contigs produced by the overlapping primers were aligned in consensus with *Bos taurus* reference sequence (Acc.No.L08604.1) to generate complete sequence of LF gene using Codon-code aligner and MEGA 6.0 tools. To identify variations/SNPs across coding region of LF gene, the sequences were subjected for multiple sequence alignment using CLC genomic workbench 8.5 software. Protparam tool (http://ca.expasy.org) was utilized to analyze the physiochemical properties such as molecular weight, pI, instability index and grand average of hydropathy (GRAVY) of LF. MEGA 6.0 (Tamura et al. 2011) was used for the phylogenetic analyses Distances were estimated by the p-distance model (Kimura and Crow, 1964) and the standard errors of the estimates were obtained through 1000bootstrap replicates.

Expression analysis of LF gene

To understand the expression pattern of LF mRNA across lactation stages in Sahiwal cows and Murrah buffaloes, a total of 40 milk samples, 20 each from both the dairy species were collected. Five milk samples representing each specific stage of lactation: colostrum (0-2 days), early (10-30 days), mid (90-120 days) and late (>240 days) were utilized to

Table 2. Primer details, annealing temperature, amplification efficiency and slope for target (LF) and reference (GAPDH, ACTB, UBC) genes in qPCR based expression analysis

Gene	Primer sequences	Tm	Efficiency	Slope
LF	GAACATCCCCATGGGCCT	60°C	102.21	-3.19
	CAGCCAGGCACCTGAAAG			
GAPDH	TGGAAAGGCCATCACCATCT	60°C	101.11	-3.28
	CCCACTTGATGTTGGCAG			
ACTB	GCGTGGCTACAGCTTCACC	60°C	104.80	-3.10
	TTGATGTCACGGACGATTTC			
UBC	TCCCTACCTGCATCATGTGC	60°C	102.45	-3.17
	GGAATTTGGGCCAGTGCTC			

isolate somatic cells. Post collection, milk samples were immediately defatted by centrifugation at 4000 rpm at 4°C for 20 min. After removing the fat layer, somatic cells were washed twice with 1X PBS at 3000 rpm for 10 min at 4°C. The cells were trizolated and processed for RNA isolation using Trizol reagent (Invitrogen Corp., CA). The quality and quantity of extracted RNA was assessed using nanovue (GE Healthcare). The purified RNA was used to synthesize first strand cDNA using Revert Aid First Strand cDNA Synthesis Kit (Fermentas, Thermo Scientific) following manufacturer instructions. The reaction was performed in an Eppendorf Gradient cycler using the program: 65°C for 5 min, 42°C for 60 min and 70°C for 5 min. The amplification was performed in 10 µl volume using StepOne Plus instrument (Applied Biosystems) using 96-well optical plate (Applied Biosystem). The reaction mixture consisted of 4 µl diluted cDNA, 6 µl mix of 5µl 2X Syber Green with ROX master mix (Fermentas, Thermo Scientific), 0.4 µl each of 10µM forward and reverse primers, and 0.2µl DNase/RNase-free water. Each sample was run in duplicate along with the reference gene. The amplification reaction conditions followed were: 10 min at 95°C, 40 cycles of 15 sec at 95°C (denaturation) and 1 min at 60°C (annealing + extension). To assess the sensitivity and specificity of the assay, a dissociation protocol with an incremental temperature of 95°C for 15 sec plus 65°C for 15 sec was performed at the end of each run. The qPCR data was normalized using panel of reference genes (*ACTB, GAPDH* and *UBC*) identified in a previous study (Varshney et al. 2012). The description of target gene and reference genes in terms of gene symbols with primer sequences, annealing temperature and their respective reaction efficiency and slope are presented in Table 2.

RESULTS AND DISCUSSION

Sequence analysis of Lactoferrin gene

This study was planned to characterize Lactoferrin, one of the important multifunctional gene in Indian native cattle (*Bos indicus*) and compared with crossbred, exotic cattle (*Bos taurus*) and riverine buffaloes (*Bubalus bubalis*). For characterization of LF coding region of about 2.1 Kb length, 9 cDNA samples of SAC, KFC and MUB, were successfully amplified using specific primer pairs (Figure 1). The manual inspection of chromatograms indicated good quality of nucleotide sequences in each of the sample. The initial sequences analysis of LF cDNA revealed the presence of complete CDS region in all the samples. The gene was found to have an ORF of 2127 bp with 780 amino acids and predicted molecular mass of 78.08 KDa. The comparison of sequence data indicated the existence of highly conserved structural organization of LF gene across the native, crossbred, exotic cattle and buffaloes. The molecular weight, pI and instability index of LF as deduced from Protparam in SAC and KFC cows showed similar

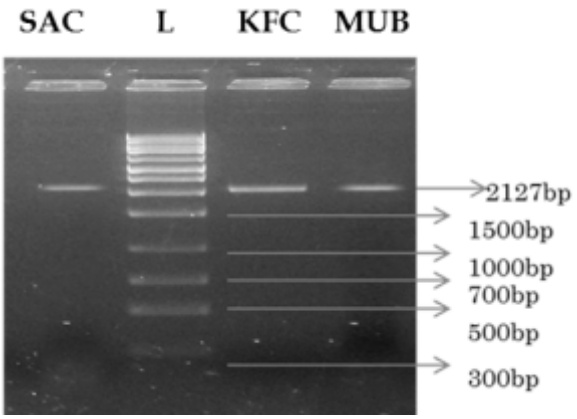


Figure 1. *Full length PCR product of lactoferrin cDNA. Lane 1: Sahiwal cattle (SAC); Lane 2: Molecular weight marker (L); Lane 3 Karan Fries cattle (KFC) and Lane 4: Murrah buffalo (MUB)*

Table 3. Physiochemical properties of LF in SAC, KFC, exotic cattle and MUB

	Physiochemical properties				
Lactoferrin	Length	Mol. Weight	Theoretical PI	Instability index	GRAVY
Exotic cattle	708	78.05	8.69	40.99	-0.289
SAC	708	78.08	8.65	41.79	-0.301
KFC	708	78.06	8.65	41.97	-0.285
MUB	708	77.69	8.36	40.90	-0.253

values as78.08 KDa, 8.65, 41.79, respectively. However, the values were slightly different from deduced *Bos taurus* values as 78.05KDa, 8.69, 40.99; whereas the values in Murrah were 77.69 KDa, 8.36 and 40.90, respectively (Table 3). The GRAVY index, an indicator of the solubility of proteins revealed hydrophilic nature of LF with values of -0.289, -0.301, -0.285, -0.253 for exotic cattle, SAC, KFC and MUB, respectively. The physiochemical parameters observed for LF in present study were somewhat similar to that reported in other cattle breeds (Pierce et al. 1991; Shashidharan et al. 2011).

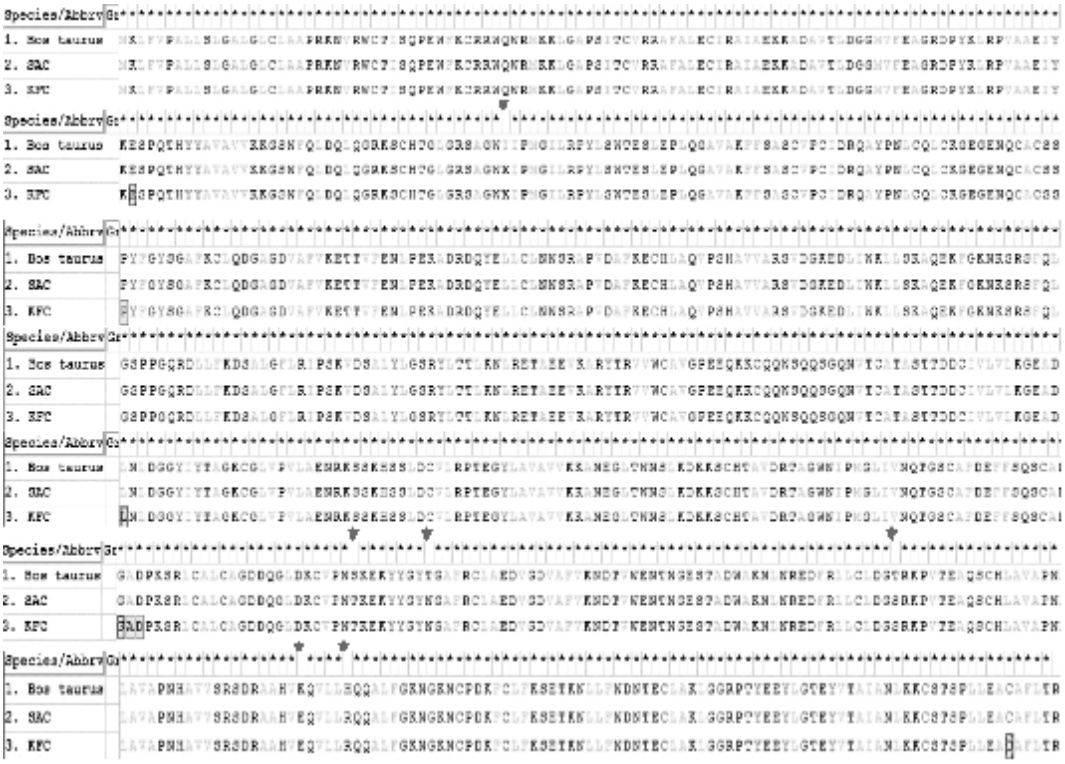
Comparison of Lactoferrin cDNA sequences across native, crossbred and exotic cattle

To identify the sequence variations in LF gene across different cattle types (native, crossbred and exotic), the sequence data for SAC (native) and KFC(cross bred) cows were compared with *Bos taurus* (exotic) reference sequence (acc. no. L08604.1). Interestingly, no variation was observed between SAC and KFC, either at nucleotide or amino acid level. However, when the sequences were aligned with *Bos taurus* sequences, a total of 6variations covering exons 4, 13 and 15 were identified (Figure 2). The 6SNPs found in the present data set were non-synonymous in nature as these could bring changes in amino acid composition in the transformed products. Amongst the 6 amino acid variations, 1 variation was observed in exon 4, 2 in exon 13, and 3 in exon 15. Remaining 14 exons were found to be monomorphic across the three cattle types.

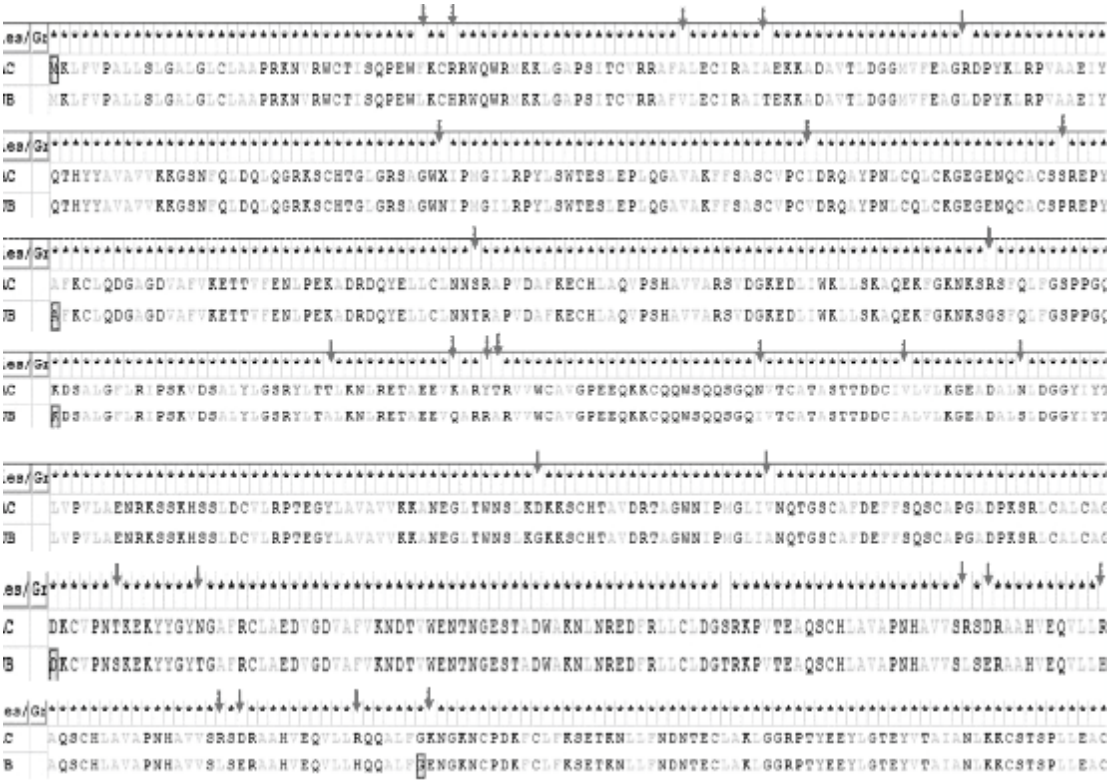
The SNP (A433G) in exon 4 resulted in substitution of isoleucine to valine at amino acid position 145. This particular SNP has also been reported in few other studies in exotic cattle (Li et al. 2004; O'Halloran et al. 2009). The nucleotide changes in exon 13 (T1612A and C1637A) also led to change in amino acid between sequences of SAC/KFC and exotic cattle. The SNP at locus T1612A resulted in substitution of threonine from serine at amino acid position 538. The second locus in exon 13 (C1637A), also led to substitution of a new amino acid. The nucleotide variation of C to A resulted in substitution of asparagine from threonine at 546 amino acid position. Both these SNPs, were found to be located at C lobe of the protein. These changes have also been reported earlier in different exotic cattle types in dbSNP data base (rs379782196 and rs208566369). Similarly, the three nucleotide changes in exon 15 also resulted in amino acid changes. The T1786A resulted in threonine to serine substitution, A1879G resulted in lysine to glutamic acid substitution, and A1895G resulted in histidine to arginine substitution at amino acid positions 596, 627 and 632, respectively.

Sequence analysis of Lactoferrin gene in SAC and MUB

The comparison of LF sequences of SAC and MUB revealed a total of 22 amino acid changes (Fig. 2b and Table 4). Out of 17 exons, 6 were conserved (monomorphic) between SAC and MUB. Exon 9 was most diverse with 5changes followed by 4 changes in exon 2;3 changes in exon 15;2 changes in exon 5,8 & 12; and one change in exon 3,4,7,10 and 16. Other than variations observed at position 145 and 632 amino acids, all other polymorphic sites were unique to buffaloes. At position 145, SAC and KFC had valine while MUB had asparagine. At position 632, both MUB had histidine while it was substituted with arginine in SAC and KFC. These changes depicted the species specific differences between cattle and buffaloes. Although large number of variations were identified in MUB, two variations *viz*., leucine to phenylalanine at amino acid (aa) 172 and threonine to isoleucine at aa 286 in MUB as reported by (Kathiravan et al. 2010) were not observed in the present study. Further, on comparison of MUB LF sequences with that of *Bubalus Arnee* (AJ005203.1), three amino acid changes *viz*., asparagine to lysine at 132, phenylalanine to leucine at 164 and cysteine to serine at 322 amino acid positions were identified. Similar to our findings, Kang et al. (2008) also observed higher variability among species than within species while analyzing 60 LF sequences from 11 species. O'Halloran et al. (2009) identified 47 variations within exonic region in six different Irish cattle populations. Amongst these, two changes were observed to affect the iron chelating ability of protein and hence overall immune function. Other studies have also shown changes in lactoferrin amino acids and altered functional and structural properties (Velliyagounder et al. 2003; Lee et al. 1997). Wojdak-



(a)



(b)

Figure 2. a) Alignment of amino acid sequences of lactoferrin across SAC, KFC and Bos taurus cattle b) Alignment of amino acid sequences of lactoferrin between SAC and MUB

Table 4. Data highlighting changes in LF gene in SAC vs MUB and exotic vs SAC/ KFC

S. No			SAC vs. MUB*		Exotic vs. SAC/ KFC**	
	Exonic Location	Location in Protein	Changes in Nucleotide	Changes in amino acids	Changes in Nucleotide	Changes in amino acids
1.	2	N lobe	T106C	Phe36Leu		
2.	2	N lobe	G116A	Arg39His		
3.	2	N lobe	C182T	Ala61Val		
4.	2	N lobe	G205A	Ala69Thr		
5.	3	N lobe	G263T	Arg88Leu		
6.	4	N lobe	G434A	Val145Asn	A433G	Ile145Val
7.	5	N lobe	A538G	Ile180Val		
8.	5	N lobe	T610C	Ser204Pro		
9.	7	N lobe	G761C	Ser254Thr		
10.	8	N lobe	C907G	Arg303Gly		
11.	8	C lobe	A1036G	Thr346Aal		
12.	9	C lobe	A1072C	Lys358Gln		
13.	9	C lobe	T1081C	Tyr361Arg		
14.	9	C lobe	A1084G		C1086G	Thr362Ala
15.	9	C lobe	A1160T	Asn387Ile		
16.	9	C lobe	T1202C	Val401Ala		
17.	10	C lobe	A1235G	Asn412Ser		
18.	12	C lobe	A1415G	Asp472Gly		
19.	12	C lobe	T1481C	Val494Aal		
20.	13	C lobe			T1612A	Ser538Thr
21.	13	C lobe			C1637A	Thr546Asn
22.	15	C lobe			T1786A	Thr596Ser
23.	15	C lobe	1856	Arg619Leu		
24.	15	C lobe			A1879G	Lys627Glu
25.	15	C lobe	A189G	Arg632His	A1895G	His632Arg
26.	16	C lobe	1915	Lys639Glu		

*The first nucleotide/amino acid is for SAC and second is for MUB. ** The first nucleotide/amino acid is for exotic cattle and second is for SAC/KFC.

Maksymiec et al. (2006) showed significant association between the somatic cell counts and LF variants in Holstein Friesian cows. Huang et al. (2010) successfully tried to associate LF haplotypes with mastitis in Chinese Holstein cattle. Association of polymorphism in LF with occurrence of mastitis has also been observed in crossbred cattle by Rahmani et al. (2012). The present data provides the basic information of genetic structure and polymorphism in lactoferrin, a potent candidate gene for disease resistance in different cattle types and buffalo. The SNPs identified in present study can be explored further to examine their impact on functional properties of this major protein.

Comparative sequence analysis for SAC and KFC revealed hundred percent homology at nucleotide level while homology percent with taurine (exotic) cattle was 96.07% and 96%, respectively. The MUB LF sequences revealed homology of 96.23%, 96.75%, 91.67%, 91.95%, 73.16%, 70.20%, 64.21%, 73.83% with Indian native cattle, cross bred cattle, taurine cattle, sheep, goat, horse, human, mouse and pig, respectively at amino acid level, indicating the high similarity of this gene among the mammalian species. Phylogenetic analysis of SAC, KFC, and MUB LF gene with different species revealed close clustering of cattle types followed by buffaloes and small ruminants (sheep and goat) (Figure 3). The

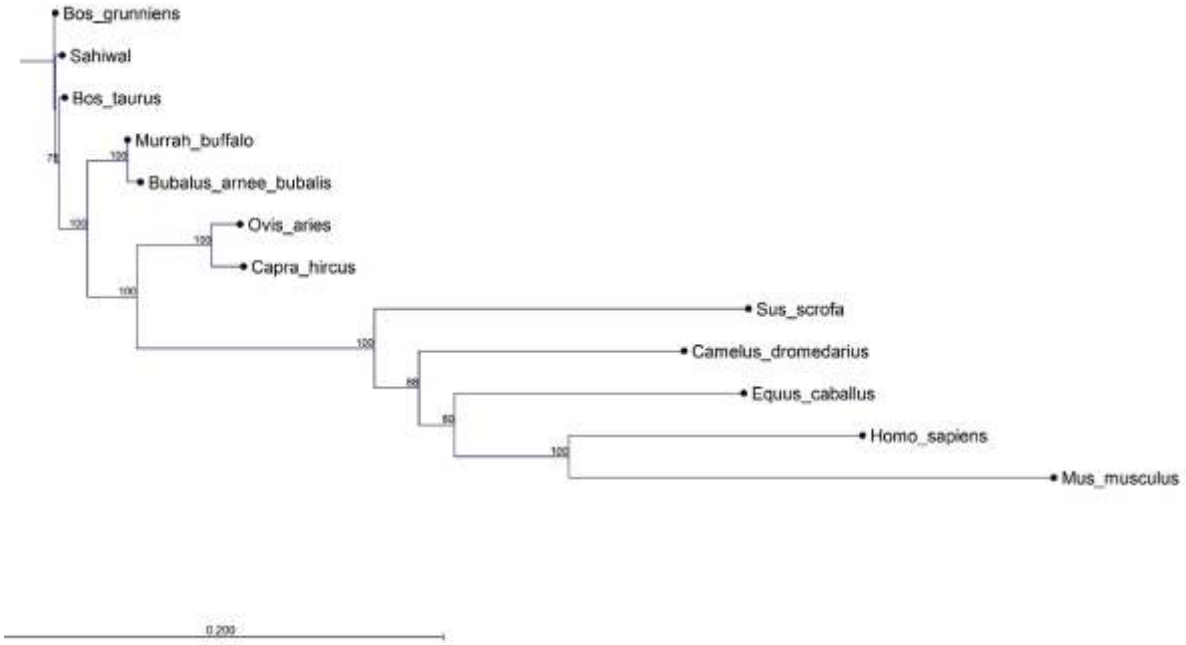


Figure 3. Evolutionary relationship across different species based on LF cDNA sequence data

high sequences similarity of LF gene across livestock species was also reported by Shashidharan et al. (2011) and Teng et al. (2012).

Relative expression pattern of LF mRNA in somatic cells of SAC and MUB

The study has also evaluated the expression pattern of LF gene in milk derived somatic cells harvested across different stages of lactation in MUB and SAC. The somatic cells were used as a source for LF mRNA as this gene is known to be abundantly present in milk secretions. All the 40 somatic cells representing different lactation stages *viz*., colostrum, early, mid and late lactation showed good quality RNA with $A_{260/280}$ ranging from 1.8-2.0. The normalized qPCR data of LF mRNA when compared across different lactation stages, it showed maximum abundance ($p<0.01$) in colostrum samples in both the dairy species (Fig. 4). The presence of LF mRNA at maximal level in colostrum samples could be attributed to its role in providing immunity to the calves. Subsequently as the lactation progressed, its mRNA level decreased continuously from colostrum to early-, and mid- lactation stages before increasing significantly again during late lactation stage. Overall, the pattern of expression of LF transcript was more or less similar in the two species and its expression was higher in colostrum and during late lactation stages. Higher abundance of LF gene in colostrum may be due to its contribution in transferring iron into the colostrum milk, and act as an important immune proteins specialized to combat various infections in new born calves. Whereas, the higher expression in late lactation stage could be due to its role in developmental process and help mammary gland to be prepared for involution stage. The present study also provides a strong clue that milk derived somatic cells could be used as an alternative non-invasive resource to study mammary

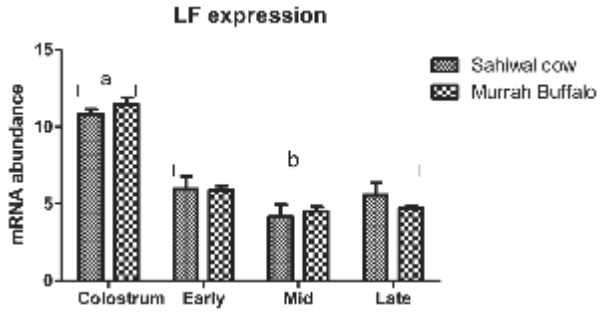


Figure 4. *mRNA abundance of LF across different lactation stages in SAC and MUB. Different letter showed significant changes in expression level ($p<0.01$)*

gland functioning of mammary gland in dairy animals.

In conclusion, the present study has characterized the LF cDNA in native, crossbred and riverine buffaloes. The comparison of coding sequences of LF gene between native cattle (SAC) and exotic cattle resulted in identification of 6 variations. Similarly, the analysis of LF cDNA between native cattle (SAC) and riverine buffaloes (MUB) has identified a total of 22 variations. All the SNPs identified between native and exotic cattle were non-synonymous in nature as these led to substitution of amino acids. Further, the study has successfully delineated the expression pattern of LF mRNA in milk derived somatic cells of SAC and MUB across different lactation stages. The analysis has shown maximum abundance of LF mRNA in colostrum samples of both SAC and MUB attributing to its role in providing immunity to the calves. Such type of studies will provide information on new variations/SNPs in LF gene in native cattle and riverine buffaloes. In future, these could be utilized as genomic resource for *in-silico* as well as genotype : phenotype association studies especially for disease resistance traits in the two major dairy species of our country.

ACKNOWLEDGEMENT

This study was supported by Indian Council of Agriculture Research, New Delhi under NAIP and CABin schemes. The authors duly acknowledge Director, NBAGR, Karnal for providing the research facilities to carry out this study.

REFERENCES

Berlutti F, Pantanella F, Natalizi T, Frioni A, Paesano R, Polimeni A and Valenti P. 2011. Antiviral properties of lactoferrin--a natural immunity molecule. Molecules 16:6992-7018.

Cao L, Zhou Y, Li X and Yi H. 2011. The relationship of haplotype in lactotransferrin and its expression levels in Chinese Han ovarian cancer. Acta Biochimica et Biophysica Sinica 43:884–890.

Detilleux JC. 2002. Genetic factors affecting susceptibility of dairy cows to udder pathogens. Veterinary Immunology and Immunopathology 88: 103-110.

Daly M, Ross P and Giblin L. 2006. Polymorphism with the Lactoferrin gene promotion in various cattle breeds. Animal Biotechnology 17: 33-42.

Duarte DC, Nicolau A, Teixeira JA, Rodrigues LR. 2011. The effect of bovine milk lactoferrin on human breast cancer cell lines. Journal of Dairy Science 94:66-76

El-Loly MM and Mahfouz MB. 2011. Lactoferrin in relation to biological functions and applications-A Review. International Journal of Dairy Science 6:79-111

Fang WH and Oliver SP. 1999 Identification of lactoferrin-binding proteins in bovinE mastitis-causing Streptococcus uberis . FEMS Microbiology Letters 176:91-96.

Fine DH, Toruner GA, Velliyagounder K, Sampathkumar V, Godboley D, Furgang D. 2013. A Lactotransferrin single nucleotide polymorphism demonstrates biological activity that can reduce susceptibility to caries. Infection and Immunity 81:1596-1605.

Gibbons JA, Kanwar JR, Kanwar RK. 2015. Iron-free and iron-saturated bovine lactoferrin inhibit survivin expression and differentially modulate apoptosis in breast cancer. BMC Cancer 15:425.

González-Chávez SA, Arévalo-Gallegos S and Rascón-Cruz Q. 2009. Lactoferrin: Structure, function and applications. International Journal of Antimicrobial Agents 33: 301.e1-8.

Hajibemani A, Sharifiyazdi H, Mirzaei A, Rowshan and Ghasrodashti A. 2012. Characterization of single nucleotide polymorphism in the 5'-untranslated region (5'-UTR) of Lactoferrin gene and its association with reproductive parameters and uterine infection in dairy cattle. Veterinary Research Forum 3:37-43.

Hirvonen J, Eklund K, Teppo AM, Huszenicza G, Kulcsar M, Saloniemi H and Pyörälä S. 1999. Acute phase response in dairy cows with experimentally induced Escherichia coli mastitis. Acta Veterinaria Scandinavica 40:35-46.

Huang JM, Wang ZY, Ju ZH et al. 2010. Two splice

variants of the bovine lactoferringene identified in Staphylococcus aureus isolated from mastitis in dairy cattle. Genetics and Molecular Research 10: 3199-3203.

Kang JF, Li XL, Zhou RY, Li LH, Feng FJ and Guo XL. 2008. Bioinformatic analysis of lactoferrin gene for several species. Biochemical genetics 46:e312-322.

Kathiravan P, Kataria RS, Mishra BP, Dubey PK, Selvakumar M and Tyagi N. 2010. Seven novel single nucleotide polymorphisms identified within river buffalo (Bubalus bubalis) lactoferrin gene. Tropical Animal Health and Production 42: 1021–26.

Kimura M, Crow JF. 1964. The number of alleles that can be maintained in a finite population. Genetics 49:725-38.

Lee TH, Shimazaki K, Yu SL, Nam MS, Kim SJ, Lee KK and Yu DY. 1997. Polymorphic sequence of Korean native goat lactoferrin exhibiting greater antibacterial activity. Animal Genetics.28: 367–369.

Li GH, Zhang Y and Sun DX. 2004. Study on the polymorphism of bovine lactoferrin gene and its relationship with mastitis. Animal Biotechnology 15(1):67-76.

Małaczewska J, Rotkiewicz Z. 2007. Laktoferyna – białko multipotencjalne. Medycyna Weterynaryjna 63: 136–139.

Mohamed JA, DuPont HL, Jiang ZD, Belkind-Gerson J, Figueroa JF. 2007. A novel single-nucleotide polymorphism in the lactoferrin gene is associated with susceptibility to diarrhea in North American travelers to Mexico. Clinical Infectious Diseases 44:945-52.

Kumari N, Goyal S, Dubey PK, Singh S, Niranjan SK, Gupta N, Prasad A and Kataria RS.2014. Cloning and sequence characterization of lactoferrin gene of Indian riverine buffalo. Indian Journal of Animal Sciences 84:47–50.

O'Halloran F, Bahar B, Buckley F, O'Sullivan O, Sweeney T and Giblin L. 2009. Characterization of single nucleotide polymorphisms identified in the bovine lactoferrin gene sequences across a range of dairy cow breeds. Biochimie 91:68-75.

Pierce A, Colavizza D, Benaissa M, Maes P, Tartra A, Montreuil J and Spik G. 1991. Molecular cloning and sequence analysis of bovine lactoferrin. European Journal of Biochemistry 196:177–84.

Plaffl MW, Wittmann SL and Meyerh HD. 2003. Gene expression of immunologically important factors in blood cells, milk cells and mammary tissue of cows. Journal of Dairy Science 86: 538-545.

Raja KN, Gupta ID and Verma A 2014. Sequence characterization of lactoferrin gene promoter region in Bos indicus cattle. Indian Journal of Animal Research 48:6-10

Rahmani SF, Jayashankar MR, Nagaraja CS, Bhaskaran R, Byregowda SM. and Satyanaraya K. 2012. Association of genotypes at promoter region of lactoferrin gene with mastitis in crossbred cattle. Indian Journal of Animal Science 82: 718-720

Seyfert HM and Kuhn C. 1994. Characterization of a first bovine lactoferrin gene variant, based on an EcoRI polymorphism. Animal Genetics 25: 54.

Shashidharan A, Singh R, Bhasker S and Mohankumar C. 2011.Physicochemical characterization and functional site analysis of lactoferrin gene of Vechur cow. Bioinformation 6: 275-78.

Smith KL and Oliver SP. 1981. Lactoferrin: a component of nonspecific defense of the involuting bovine mammary gland. Advances in Experimental Medicine and Biology 137:535-554.

Sordillo LM, Nickerson SC, Akers RM and Oliver SP. 1987. Secretion composition duringbovine mammary involution and the relationship with mastitis. International Journal of Biochemistry 19:1165-1172.

Teng CT, Pentecost BT, Marshall A, Solomon A, et al. 1987. Assignment of the lactotransferrin gene to human chromosome 3 and to mouse chromosome 9. Somatic cells and Molecular Genetics 13: 689-693

Teng CT. 2002. Lactoferrin gene expression and regulation: An overview. Biochemistry and Cell Biology 80: 7-16.

Teng CT and Gladwell W. 2006. Single nucleotide polymorphisms (SNPs) in human lactoferrin gene. Biochemistry and Cell Biology 84:381-4.

Valadan R, Mirzaei A and Sharifiyazdi H. 2011. Detection of single nucleotide polymorphism in TATA box of Lactoferrin gene and its association with uterine infections in cows. Online Journal of Veterinary Research 15: 190-199.

Varshney N, Mohanty AK, Kumar S, Kaushik JK, Dang AK, Mukesh M, Mishra BP, Kataria R, Kimothi SP, Mukhopadhyay TK, Malakar D, Prakash BS, Grover S and Batish VK. 2012. Selection of suitable reference genes for quantitative gene expression studies in milk somatic cells of lactating cows (Bos indicus). Journal of Dairy Science 95:2935-45.

Velliyagounder K, Kaplan JB, Furgang D, Legarda D, Diamond G, Parkin RE. 2003. One of two human lactoferrin variants exhibits increased antibacterial and transcriptional activation activities and is associated with localized juvenile periodontitis. Infection Immunity 71:6141-7.

Wang SR, Jyhshiun L, Cheng IC and Yun TL. 1998. Characterization and functional analysis of porcine Lactoferrin gene. Gene 215:203-12.

Wojdak-Maksymiec K, Kmiec K and Ziemak J. 2006. Association between bovine lactoferrin gene polymorphism and somatic cell count. Veterinary Medicine 51: 14-20.

Yamauchi K, Wakabayashi H, Shin K and Takase M. 2006. Bovine lactoferrin: benefits and mechanism of action against infections. Biochemistry and Cell Biology 84:291-296.

Zhou L, Yang Y, Zhong H, Kong Li, Xin Li, Di GD and Wang GL. 2006. Detection and Characterization of PCR–SSCP Markers of the Bovine Lactoferrin Gene for clinical mastitis. Asian-Australasian Journal of Animal Science 19: 1399-1403.

Zupin L, Polesello V, Coelho AVC, et al. 2015. Lactotransferrin gene functional polymorphisms do not influence susceptibility to human immunodeficiency virus-1 mother-to-child transmission in different ethnic groups. Memórias do Instituto Oswaldo Cruz. 10:222-229.

Preparation of Shrikhand from goat milk using probiotic cultures

V B Shinde, S S Kamble*, D H Kankhare and PT Dhole
Division of Animal Husbandry and Dairy Science
Mahatma Phule Krishi Vidyapeeth, Rahuri.- 413 722. (MS) India

ABSTRACT

Shrikhand is a traditional fermented milk product of Indian origin and is popular in western parts of India. Starter culture is known to play vital role in the development of body, texture, flavor and aroma in the finished products and contributes significantly towards overall acceptance of products by the consumer. In investigation the probiotic cultures used in preparation of shrikhand were *Lb. acidophilus* and *Lb. delbruekii ssp.bulgaricus* alone and in combination and LF-40 culture alone as a control. Probiotic cultures viz., *Lb. acidophilus* and *Lb. bulgaricus* alone and in combination containing viable lactobacilli counts more than 10^7 c.f.u./g in fresh as well as in stored shrikhand. The results revealed that organoleptically acceptable shrikhand from goat milk can be prepared by using probiotic cultures viz., *Lactobacillus acidophilus* and *Lactobacillus delbrueckii ssp.* bulgaricus alone and in combination. There was non significant difference observed for sensory evaluation for all treatments. Flavour score observed highest in (8.00) stored shrikhand prepared by combination of *Lactobacillus acidophilus* and *Lactobacillus delbrueckii.* The probiotic shrikhand can be prepared and stored at 7 + 1 C for a week and possessing good organoleptic quality with therapeutic quality.

Key words: Shrikhand, probiotic, goat milk product.
***Corresponding author:** skamble09@gmail.com
Revised manuscript received: 09.5.2017; Manuscript accepted: 24.6.2017

INTRODUCTION

Goat milk possesses medicinal and therapeutic value. Goat milk is good for human health. The fermented milk products have reputation due to their nutritional and therapeutic properties (Singh, 1996). Probiotics are live microorganisms that confer health benefits to the host when administrated in certain quantities (FAO/WHO, 2001). They are essential for the proper functioning of the digestive tract, commonly referred to as 'friendly', 'beneficial' or 'good' bacteria (Jha *et al* , 2015). The interest in probiotic foods has been increasing in recent years, and has inspired invention and driven the development of new products around the world, in order to maintain the gut health with addition of probiotic bacteria into foods (Rajani *et al* 2016). In India dahi is traditionally prepared using mesophilic starter cultures. The rate of acid production is typically slower in dahi compared to yoghurt fermentation, which is typically completed within 5 hours. Therefore, commercial processors are increasingly using yoghurt cultures (*Strptococcus thermophilus* and *Lactobacillus delbrueckii* supsp *bulgaricus*) to prepare dahi (Korasapati *et al,* 2016). Dahi is used as a base material for preparation of Shrikhand, indigenous butter lassi etc (Mistry, 2001, Aneja *et al*., 2002).

Considering the importance of Shrikhand in human diet, the importance of goat milk and the importance of lactic acid bacteria in the human health, the present study was conducted to determine the suitability of probiotic strains *Lactobacillus delbrueckii* and *Lactobacillus acidophilus* to make an acceptable quality probiotic Shrikhand from goat milk.

MATERIAL AND METHODS

Fresh goat milk was obtained from Osmanabadi goat unit, Post graduate institute, M.P.K.V., Rahuri. The milk was standardized to 6 % fat. The standardized goat milk was heated to 85 ^{0}C for 10 min. and then cooled to 30 ^{0}C . As per the treatment the cultures were inoculated @ of 1 percent. T_0 : LF- 40; T_1 :*Lactobacillus acidophilus*; T_2 :*Lactobacillus delbrueckii ssp. Bulgaricus*; T_3 : Combination of *Lb.*

acidophilus and *Lb. delbrueckii ssp. Bulgaricus* (1:1).

The inoculated cultures wee properly mixed and then incubated at 37 ^{0}C for 8-10 hrs. After incubation period curd were broken and tied in muslin cloths separately and then tied in hanging position to drain whey for 15-16 hrs. Weight of chakka were recorded and ground sugar (@ 45 % by weight of chakka) was added. The chakka and sugar was kneaded to smooth paste and filled in polystyrene cups and then stored at 7 $\pm$ 1^{0}C temperature for one week. The prepared shrikhand samples were taken for organoleptic evaluation, chemical and microbial analysis. Freeze dried pure culture of LF-40, *Lactobacillus acidophilus* ,*Lactobacillusdelbrueckii ssp. Bulgaricus* were obtained from the National Dairy Research Institute , Karnal , Haryana. These cultures were maintained separately in sterilized 10 ml reconstituted skim milk in test tubes. The test tubes were autoclaved at 121^{0}C / 15 lb / 15 min.

Analytical Techniques

Goat milk was analyzed for fat by Gerber method IS: 1224, Part I. Samples of Shrikhand were analyzed for total solids by gravimetric method (IS: 1166 (part II), 1973, fat as per the Gerber method IS: 1224 (Part – II) 1977 and Protein by Kjeldalh method and acidity as per IS-1166 (Part – II) 1973.

Enumeration of viable lactobacilli count

The MRS agar having pH 6.4 with double layer was used for enumeration of viable lactobacilli counts of T_1, T_2, &T_3 samples of shrikhand. Ten grams of shrikhand samples were taken separately in 90 ml sterilized phosphate buffer solution and then subsequently diluted using 9 ml phosphate buffer. The 7th and 8th dilution were taken in duplicate into pertiplates and then MRS agar having pH 6.4 was added and mixed well. The plates were allowed to solidify. The plates were again overlaid with same agar and incubated at 37^{0}C /48 hr and then lactobacilli counts were recorded.

Enumeration of lactic culture (LF-40) counts

The lactose purple agar (LPA) having pH 7.4 $\pm$ 0.1 was used for the enumeration of lactic acid culture (LF-40) counts of control (T_0).

Organoleptic quality of shrikhand

The fresh and refrigerated stored shrikhand (at 7 $\pm$ 1^{0}C for a week) samples were subjected to the organoleptic evaluation. Shrikhand samples were provided to the panel of seven semi-trained judges for sensory evaluation. Samples were evaluated for flavour, body and texture, colour and appearance and overall acceptability by using 9-point Hedonic Scale.

Statistical Analysis

The data were subjected to Completely Randomized Design (CRD) as described by Snedecor and Cochran (1967).

RESULTS AND DISCUSSION

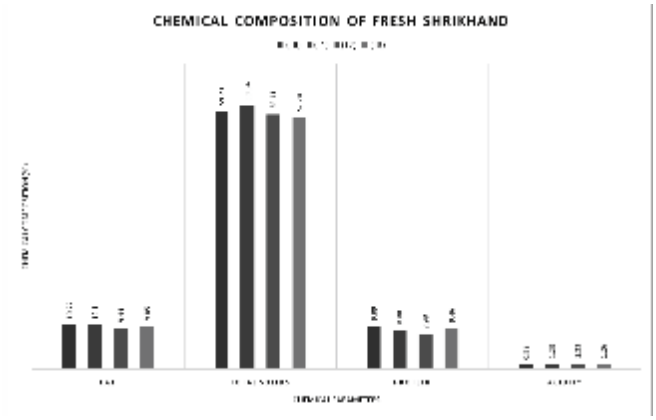
The fresh goat milk used for preparation of Shrikhand was containing on an average 4.05 per cent fat and 3.17 per cent protein. There was non-significant difference in the flavour scores of all fresh shrikhand samples. The samples T_2 prepared by using *Lb.delbrueckii ssp. Bulgaricus*@ 1 per cent secured highest flavour score (8.31) than other types of Shrikhand. The mean flavour score of treatment T_0, T_1, T_3 were 8.00, 7.94, and 8.00 respectively. Kiran et al (2012) reported flavor of *Brevibacillus brevis* MMB 12 fermented product scored 7.0 to 7.1

Table 1. Chemical composition and sensory quality of fresh Shrikhand.

Treatment	Chemical composition (%)				Sensory evaluation			
	Fat	Total solids	Protein	Acidity	Flavour	Body & Texture	Colour & appearance	Overall acceptability
Control (T0)	10.22	59.21	9.89	0.98	8.00	7.63	7.88	7.81
Lb. acidophilus(T1)	10.10	60.34	8.88	1.23	7.94	7.81	7.13	8.06
Lb. delbrueckii ssp (T2)	9.44	58.41	7.98	1.33	8.31	7.81	7.63	8.31
Combination (T3)	9.85	57.74	9.46	1.26	8.00	7.88	7.13	8.80
S.E. +	0.167	0.339	0.056	0.027	--	---	--	--
C.D.	0.514	1.044*	0.174	0.084	NS	NS	NS	NS

Table 2. Acidity and sensory quality of stored shrihkhand

Treatment	Acidity	Sensory evaluation			
		Flavour	Body & texture	Colour & appearance	Overall acceptability
Control (T0)	0.992	7.13	7.56	7.50	7.88
Lb. acidophillus (T1)	1.290	7.71	8.19	8.00	8.06
Lb. delbrueckii ssp. (T2)	1.333	6.25	6.88	7.13	6.81
Combination (T3)	1.308	8.00	8.06	8.30	8.63



between 24 to 192 hours of preservation, and *Lb. delbrueckii Subsp. Bulgaricusc* curd had maximum score of 7.0 which decreased to 6.0 after 96 hrs. Fadela et al (2009) also stated that the rate of acidification depends on amount and type of starter culture, thus affecting the sequence of gelation and determining the characteristic of casein matrix. There was considerably effect on body and texture characteristics of Shrikhand samples prepared by using various starter cultures. Mean score for body and texture of (T_3) Shrikhand was higher (7.88) because body was uniform and smooth than rest of the samples (T_0) ,which recorded lower score (7.63). The score was observed to be minimum as body was weak; texture was gritty and had definite curd particles. The samples T_1 and T_3 scored lower than rest of the treatment for colour and appearance. The treatment T_3 secured highest score (8.8) for overall acceptability than T_0, T_1 and T_3 types, as it had smooth , uniform body and sweetish- sour taste.

Chemical quality of Shrikhand

The result revealed that, fat content in all treatments was in the range of 8.5 to 10.30 per cent (Table -1) which coincided with the results of Zariwala and Sharma (1976). The maximum total solids content was recorded in T1 (60.34 per cent) sample. The treatment T0, T2 and T3 recorded 59.21, 58.41 and 57.74 respectively. The total solids content of samples coincide with the results of Patil (1991) ,Rachakonda (1995) and Bhogra and Mathur (2000). The table revealed that the protein content in control (T_0) was 10 per cent which was slightly higher than (8.81 %) , which was also reported by Karthikeyan (1993). The significant difference was observed in the protein content for all treatments. Acidity in all treatment was observed in the range of 0.98 to 1.26 %. And as the treatment level increases, acidity also increases. Kiran et al (2012) reported that curd

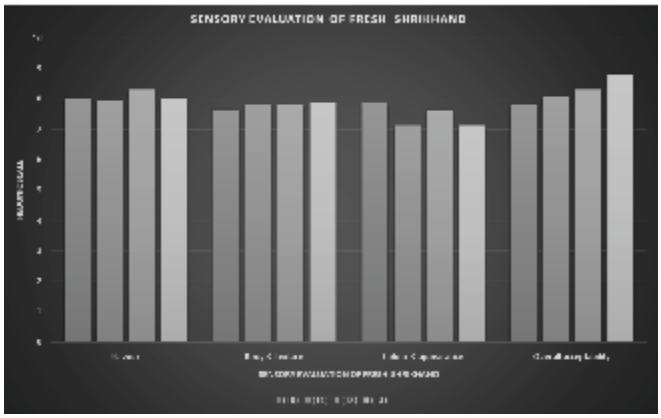


Table 3. Total counts of lactobacilli in fresh and refrigerated stored shrikhand. (Mean of four samples)

Name of product	Counts of lactobacilli (c.f.u./g)		
	T 1	T2	T3
Fresh shrikhand	55 X 107	75 X 107	100 X107
Stored shrikhand	30 X 107	38 X 107	53 X107

Table 4. Lactic culture (LF-40) counts of fresh and refrigerated stored shrikhand. (Mean of four samples)

Name of product	Lactic culture (LF-40) counts (c.f.u./g)
Fresh shrikhand	25 X 107
Stored shrikhand	15 X 107

sample fermented with *Brevibacillus brevis* observed lower percentage of acidity (0.58% to 0.66%) after 12 hrs, than Lb. delbrueckii (0.98 % to 1.68 %).The result of high acid production for *Lb. delbrueckii Subsp. bulgaricus* culture agreed with findings of Kamruzzaman et al. (2002). These were slightly increases in acidity of all stored samples. All stored Shrikhand samples were fulfilled the BIS standard of Shrikhand (maximum 1.4 % lactic acid).

Sensory quality of stored shrikhand

The sensory evaluation of Shrikhand samples stored at 7 ± 1^0C was performed on 8th day. This day was selected on the basis of lactobacilli count, to have their beneficial effect with maximum viability. As day progresses, the viability of lactobacilli drastically lowered down during storage. The results of organoleptic quality of Shrikhand are showed in table -2. Mean score for flavour of stored Shrikhand of T_3(8.0) was significantly higher (8.0) than $T_0$and T_2 and it was at par with the score of T_1 (7.71). The results revealed that the stored Shrikhand of T_2 prepared by using *Lb. delbrueckii spp. bulgaricus* shown significantly lower score (6.88) for body and texture than all other samples as it because slightly grainy. For colour and appearance the score ranges between 7.13 to 8.30. There was significant difference observed for colour and appearance. Overall acceptability score of Shrikhand for T_2 was significantly lower (6.81) while that of shrikhand T_3(8.30). It was clear from the table no.2 that all the cultures gave the Shrikhand of acceptable quality even after one week of refrigerated (at 7 ± 1^0C) storage. It is revealed that fresh Shrikhand was more acceptable than stored Shrikhand and sensory score was declined on storage. Chemical state of milk proteins of products of their breakdown imparts characteristics physical, chemical and sensory properties to most of the dairy products (Sikorski et al 2001).The result for high colour score for *Lb. delbrueckii Subsp bulgaricus* was also observed by Kiran et al (2012). Lower values for acidity was observed for curd prepared by using *Lb. acidophilus* by Patil et al (2015).

Acidity of stored shrikhand

Acidity was significantly lowered in control stored Shrikhand (T_0) prepared by using LF-40 culture than other treatments (table-2). All stored Shrikhand samples were fulfilled the BIS norm for maximum per cent lactic acidity.

Microbial analysis

Lactobacilli counts of fresh and refrigerated stored shrikhand: In order to obtain maximum therapeutic value, the fermented milk product should contain population of viable cells of probiotic cultures more than 10^7c.f.u. at the time of consumption (Speck, 1976;Tamine et al. 1995). It was observed (Table-3) that T_3 showed higher viable lactobacilli count (100 x 10^7c.f.u./g) than fresh Shrikhand samples (T_1 and T_2). On refrigerated storage, viable lactobacilli count of shrikhand (T_3) decreased up to 53 x 10^7c.f.u./g. The viable counts of *Lactobacillus acidophilus* observed in the study were slightly higher than that reported by Patil *et al*. (2015) and Sheikh *et al* (1990). All samples showed the required beneficial viable lactobacilli count in product when fresh and showed decreasing trend in refrigerated storage. Agnihotri and Pal (1997) noted the similar observation.

Lactic culture (LF-40) counts

The lactic culture count of shrikhand prepared by using LF-40 culture was recorded. In fresh shrikhand it was 25 x 10^7 and in stored shrikhand (To) it was 15 x 10^7 c.f.u./g, respectively (Table - 4).

CONCLUSION

Delicious shrikhand with characteristics flavour can be prepared by using probiotic cultures viz., *Lb. acidophilus and Lb. bulgaricus* alone and in combination containing viable lactobacilli counts more than 10^7c.f.u. /g when fresh as well as stored. The probiotic shrikhand can be prepared and stored

at 7 ± 1^{0}C for a week and possessing good organoleptic quality with therapeutic quality.

REFERENCES

Agnihotri UK and Pal UK. 1997. Changes in goat milk shrikhand on ambient and refrigerated storage. Indian Journal of Small Ruminants 3:86-92.

BhograVR and Mathur ON.1992. Chemical quality of some marketedindigenous milk products-major constituents and mineral composition of shrikhand. Journal of Food Science and Technology 29:121-122.

FAO/WHO 2001. Health and nutritional properties of probiotics in food including powder milk with live lactic acid bacteria. Report of a joint FAO/WHO expert.

IS: 1166 (Part II) 1973. Specification for condensed milk. Indian Standard Institute, ManakBhavan, New Delhi.

IS: 1224, 1977. Determination of fat in whole milk, Evaporated (Unsweetened) milk, separated milk, skim milk, buttermilk and cream by Gerber method. Indian Standards Institute, ManakBhavan, New Delhi.

Jha Prajna, Kataria Ankita, Sabikhi Lata , Kumar Satish M.H. and Malik R.K. 2015. Screening of dahi starts for compatibility with Lactobacillus gasseri strains to slect a co-culture for probiotic dahi. Indian Journal of Dairy Science 68: 137 -143.

Kamruzzaman M., Islam M.N. and Rahman M.M.(2002). Shelf life of different types of Dahi at room and refrigeration temperature. Pakistan Journal of Nutrition, 234-237.

Kiran Usha, Appaiah Anu and Appallf Sushma (2012). Extension of shelf life of curd and Indna fermented milk by using a new isolate of Brevibacillus brevis strain as starter culture. Innovative Romanian Food Biotechnology . March (10): 48-55.

Korasapati N.R., Kumar S., Singh A., Kumar Sanjay, Velugoti P.R. and Thippareddi H. (2016). Behavious of Listeria Monocytogenes in Buttermilk, an Indian Femented milk beverage. Indian Journal of Dairy Science 69:447-454.

Karthikeyan S. 1993. Study on the utilization of sweet cream buttermilk for the manufacture of shrikhand .M.Sc. (Agri.) Thesis submitted to Gujarat Agril.Univ., Gujrat (India).

Patil M.S., Dhole P.T. and Chavan K.D.(2015). Studies on quality evaluation of Probiotic custard apple (Annona Reticulata) Dahi. Indian Journal of Nutrition 2: 1-6.

Sheikh N.M., Toarder G.K. , Harron S.N. , Khatoon M (1990). Bacteriology of Dahi . 1.quantitative and qualitative studies of microflora of dahi scientific researcher east regional laboratories. 7:1-8.

Patil VR.1991.Preparation of shrikhand fortified with different fruit pulps. M.Sc. (Agri.) Thesis submitted to KonkanKrishiVidyapeeth, (Maharashtra).

Rachakonda KP. 1995. Manufacture of therapeutic shrikhand using Lactobacillus acidophilus. M.Sc. (Agri.) Thesis submitted to MarathwadaAgril. University, Parbhani (Maharahstra).

Sikorski ZE. 2001. Chemical and Functional properties of food proteins, Techomic Publishing Co.Inc. USA.

Snedecor GW and Cochran WG. 1967. Statistical Methods 7th edn. Oxford and IBH Pub. Co. New Delhi.

Speck ML.1976. Interaction among lactobacilli and man. Journal of Dairy Science 59: 338-343.

Tamine AY, Marshall VME and Robinson RK.1985. Microbiology and technological aspects of milk fermented by bifidobacteria. Journal of Dairy Research 62:151-187.

Zariwala IT and Sharma UP. 1976. Development of starter cultures for chakka and shrikhand in the western region. Annual Report, National Dairy Research Institute, Karnal. Pp.221.

BoLA-DRB3 polymorphism and their association with milk production traits in Indian cattle breeds

Hitesh Lamba[1], Deepak Sharma[2]*, Satyendra Pal Singh[2], Madhu Tiwari[2], Rakesh Goel[2], Vijay Pandey[3] and Sanjeev Kumar Singh[4]

College of Biotechnology, UP Pt. Deen Dayal Upadhyaya Pashu Chikitsa Vigyan Vishwa Vidyalaya Evam Go Anusandhan, Mathura-281001, India

ABSTRACT

The present study was undertaken to investigate the polymorphism in *DRB3* gene in Indian cattle breeds using PCR-RFLP assay. Amplification of 100 DNA samples of Sahiwal and Hariana breeds revealed 304 bp products using specific primer pairs. These amplicons were digested using *Hae* III restriction enzyme. It produced five genotypic patterns viz., AA genotype (170, 82 and 52 bp); BB genotype (222 and 82 bp), EE genotype (170 & 134 bp), heterozygous AB genotype (222, 170, 82 and 52 bp bands) and BD genotype (222, 193, 82 and 29 bp bands). AA genotype was the most frequent among all the screened cattle, followed by the AB, EE, BD and BB genotypes. The frequency of DRB3/*Hae*III A, B, D and E alleles were 0.660, 0.185, 0.035 and 0.120, respectively. Association study revealed no significant difference for milk production traits among all the *DRB3* genotypes in first and second lactation except the dry period (DP), which had significant difference in second lactation. There was significant difference (*P = 0.0106*) for somatic cell count (SCC) among all the genotypes in screened Hariana cattle, while in sahiwal cattle no association was observed. This study needs further investigation on large population size with the incorporation of other breeds of Indian cattle.

Key words: Sahiwal cattle, Hariana cattle, PCR-RFLP, *DRB3*, Polymorphism, milk production traits.

Present Address: [1]College of Biotechnology, DUVASU, Mathura, India [2]Department of Animal Genetics and Breeding, C.V.Sc. & A.H., DUVASU, Mathura, India. [3]Department of Veterinary Biochemistry, C.V.Sc. & A.H., DUVASU, Mathura, India. [4]Department of Veterinary Extension, C.V.Sc. & A.H., DUVASU, Mathura, India.

***Corresponding author:** drdeepakivri@gmail.com

Manuscript received: 12.5.2017; Manuscript accepted: 20.5.2017

INTRODUCTION

BoLA-DRB3 exon 2 alleles in cattle have been found to be associated with resistance or susceptibility to various diseases affecting immunity, somatic cell count (SCC) and mastitis resistance in cattle (Baltian et al. 2012). The *DRB3.2* polymorphism has also been observed to be associated with milk production traits (Starkenburg et al. 1997). Therefore, this MHC gene complex is a potential candidate markers for selection purposes in milch cattle (Parmentier et al. 1999).

Several single nucleotide polymorphisms (SNPs)/ mutations in *BoLA-DRB3* gene and their association with milk production traits have been reported in different exotic cattle (Gelhaus et al. 1995; Miretti et al. 2001; Miltiadou et al. 2003; da Mota et al. 2004; Nassari et al. 2008). This polymorphism has also been studied in various Indian cattle breeds including Ongole (Aravindakshan and Nainar 1999), Gir (Acharya et al. 2002), Rathi (Sharma et al. 2005), Kankrej (Behl et al. 2007) and Sahiwal (Chakravorty et al. 2015); crossbred cattle (Ganguly et al. 2004) and buffalo breed (Arvindakshan et al. 2000; Singh et al. 2004; Stafuzza et al. 2015). Keeping all this in consideration, the present study was undertaken to investigate the status of *BoLA-DRB3* gene polymorphisms in Indian Sahiwal and Hariana cattle breeds using *Hae*III/PCR-RFLP assay and their association with milk production traits and SCC.

MATERIALS AND METHODS

Animal source and DNA Extraction

A total of 100 females of Sahiwal (n = 50) and Hariana (n = 50) cattle maintained at Instructional Livestock Farm Complex (ILFC), DUVASU, Mathuta (Uttar Pradesh), were utilized in the present investigation.

Genomic DNA was isolated from venous blood using the standard protocol of Sambrook and Russel (1991). An amplicon of 304 bp PCR product of exon 2 of *DRB3* gene was amplified by using the primer (Siguardardtottir et al. 1991; 'F': 5 - GAT GGA TCC TCT CTC TGC AGC ACA TTT CCT - 3 & 'R': 5 - CTT GAA TTC GCG CTC ACC TCG CCG CTG -3). The cycle conditions included an initial denaturation at 94°C for 2 min followed by 35 cycles of denaturation at 94°C for 60 sec, annealing at 60°C for 60 sec and extension at 72°C for 60 sec and a final extension at 72°C for 10 min. The PCR product was checked by agarose gel electrophoresis in 1x TAE buffer after staining with ethidium bromide and visualized under UV light.

HaeIII/PCR-RFLP assay and calculation of gene and genotypic frequency

The restriction digestion was carried out at 37°C for 14-16 hr in a total volume of 15µl containing 5.0 µl of PCR product, 1.5 µl of 10X RE buffer and 10 Units (1.0 µl) *Hae*III enzyme (New England Biolabs). The data was generated by estimating the frequency of different RFLP pattern. The allelic frequency and genotypic frequencies of *DRB3* gene was estimated by standard procedure (Falconer and Mackay 1996). The chi square (χ^2) test ($P \leq 0.05$) was performed to test whether the distribution of the genotype frequencies was in the Hardy-Weinberg equilibrium (Snedecor and Cochran, 1989).

The association study of different genotypes with the milk production traits viz., lactation period (LP), total milk yield (TMY), milk yield in 300 days (MY300), dry period (DP) and calving interval (CI) was carried out. Statistical analysis of milk production traits and SCC was carried out using the General Linear Model (GLM) using SPSS software. The following linear model was applied:

$$Yij = \mu + Gi + eij$$

Where: Yij– observed trait value in animal; µ – mean trait value; Gi – effect of genotype; eij– random error. Significant differences among least square means of different genotypes were calculated using Duncan's multiple-range test, and $P \leq 0.05$ were considered to be statistically significant.

RESULTS AND DISCUSSION

*Hae*III/PCR-RFLP assay of the 304 bp of amplified products of *DRB3.2* gene revealed five types of genotypes; one of them was of 170, 82, 52 bp (AA genotype); second of 222 and 82 bp (BB genotype), third of 170 and 134 bp (EE genotype) and heterozygous pattern have 222, 170, 82 and 52 bp bands (AB genotype) and 222, 193, 82 and 29 bp bands (BD genotype) (Fig. 1). This revealed that the screened cattle used in the present study were polymorphic in nature with four types of alleles A, B, D and E. The genotypic and allelic frequencies of DRB3/*Hae*III gene are given Table 1. Chi square analysis revealed the screened cattle population was not in Hardy- Weinberg equilibrium (Table 1).

Arvindakshan and Nainar (1999) observed seven restriction patterns in Jersey crossbred animals and four patterns in Ongole cattle where three restriction

Table 1. Genotypic and allelic frequencies of DRB3/HaeIII gene in cattle.

Breed	Genotypic frequency (%)			Allelic frequency					
	AA	AB	BB	BD	EE	A	B	D	E
Sahiwal	48.0	20.0	6.0	12.0	14.0	0.58	0.22	0.06	0.14
(N = 50)	(n = 24)	(n = 10)	(n = 3)	(n = 6)	(n = 7)				
Hariana	64.0	20.0	4.0	2.0	10.0	0.74	0.15	0.01	0.10
(N = 50)	(n = 32)	(n = 10)	(n = 2)	(n = 1)	(n = 5)				
Total	56.0	20.0	5.0	7.0	12.0	0.66	0.185	0.035	0.12
(N = 100)	(n = 56)	(n = 20)	(n = 5)	(n = 7)	(n = 12)				
Observed	56	20	5	7	12	$\chi2$ cal = 133.65			
						$\chi2$ tab = 16.81 (P<0.01)			
Expected	43.56	24.42	3.42	1.29	1.44	$\chi2$ cal > $\chi2$ tab, df = 6, P<0.01			

Where; N = Sample size, n = Number of animals in particular genotype

Table 2. Association of DRB3/ HaeIII genotypes with milk production traits in Indian cattle

Lactation	Genotype	n	LP	TMY	MY300	DP	CI
First							
(N=73)	AA	45	360.0 ± 14.33	1710.0 ± 104.0	1400.0 ± 71.92	283.0 ± 19.68	525.0 ± 22.31
	AB	14	407.0 ± 32.60	2230.0 ± 231.0	1750.0 ± 136.0	304.0 ± 51.27	506.0 ± 23.54
	BB	4	325.0 ± 20.61	1740.0 ± 71.72	1560.0 ± 105.0	368.0 ± 49.26	470.0 ± 35.58
	BD	3	350.0 ± 49.31	1690.0 ± 41.13	1410.0 ± 122.0	281.0 ± 3.17	413.0 ± 116.0
	EE	7	350.0 ± 33.91	1920.0 ± 273.0	1600.0 ± 219.0	200.0 ± 37.68	539.0 ± 64.61
Second	AA	35	345.0 ± 20.19	1710.0 ± 131.0	1390.0 ± 85.53	291.0 ± 12.19b	454.0 ± 19.96
(N= 57)	AB	11	321.0 ± 34.94	1900 ± 211.0	1610.0 ± 166.0	253.0 ±28.57 b	476.0 ± 34.72
	BB	4	302.0 ± 33.57	1790.0 ± 145.0	1640.0 ± 39.78	206.0 ± 2.98 b	457.0 ± 47.97
	BD	2	377.0 ± 20.5	2300.0 ± 54.00	1840.0 ± 143.0	95.0 ± 30.0a	328.0 ± 129.0
	EE	5	388.0 ± 50.16	2030.0 ± 365.0	1530.0 ± 195.0	282.0 ± 45.91 b	493.0 ± 54.95

Different letters in superscript of a given row indicates significant (P<0.05) difference between genotypes, n — number of individuals in particular genotype, N — total number of individual in particular lactation.

patterns, *viz*., D, E and G were seen in Jersey crossbred cattle but not observed in studied Ongole cattle. Out of the seven *Hae*III patterns observed in their study, six patterns, viz. AA (170, 82, and 52 bp), BB (222 and 82 bp), DD (193, 82 and 29 bp), EE (170, 82, 46 and 6 bp) were also reported by Gelhaus et al. (1995) in cattle. The fragment patterns CC (170, 82 and 49 bp) and FF (170, 82, 48 and 4 bp) reported by Gelhaus et al. (1995). However, a new pattern, tentatively named as 'I' characterized by the absence of any *Hae*III site was observed with frequencies 0.15 and 0.17 in Jersey crossbred and Ongole breeds, respectively. The presence of this apparently zebu-specific pattern is quite understandable in the light of the highly polymorphic nature of the *BoLA-DRB3* gene. In the present investigation, the frequency of BD genotype was very low (2.0%) in the Hariana cattle and this might be due to the random selection of animals for polymorphism study.

The mean values (Mean ± S.E.M.) of each trait related to each genotype in the first and second lactation are presented in Table 2. Association study revealed that no significant difference was observed for any milk production traits among all the *DRB3* genotypes in first lactation. In second lactation, only DP showed significant (*P = 0.01*) difference among genotype. BD genotype had lower DP among all the genotypes in second lactation, which might be due to small sample size. Though, no reports were available regarding association studies of exon 2 of *DRB3* locus with milk

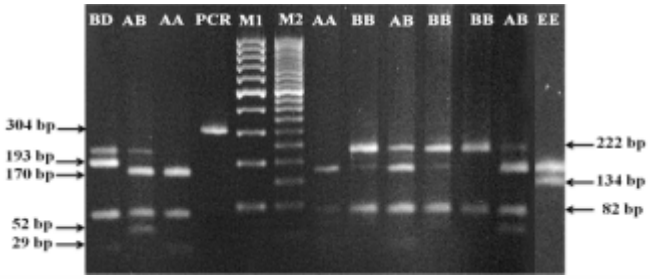


Fig 1. The HaeIII/PCR-RFLP assay in cattle and buffalo revealed five types of banding pattern (genotypes); one of them was of 170, 82, 52 bp (AA genotype); second of 222 & 82 bp (BB genotype), third of 170 & 134 bp (EE genotype) and heterozygous pattern have 222, 170, 82 & 52 bp bands (AB genotype) & 222, 193, 82 & 29 bp bands (BD genotype) M1=100 bp ladder, M2=50 bp ladder

production traits. However, various authors had reported that *DRB3.2* locus with different alleles described by Van Eijk et al. (1992) other than present study had significant association with milk yield and related traits. Starkenberg et al. (1997) found the association of allele *7 with increased milk yield and allele *26 with decreased milk yield in Holsteins cattle and Rupp et al. (2007) observed significant association of increase in milk yield with allele *11 in Canadian Holsteins. *BoLA-DRB3.2**10 has significant association with reduced fat yield in the Jersey population (Ledwidge et al. 2001) and *BoLA-DRB3.2**22 was associated with decreased milk and protein yield in Canadian dairy cattle (Sharif et al. 1998).

The mean values (Mean ± S.E.M.) of SCC for each genotype are presented in Table 3. There was significant difference *(P = 0.0106)* for SCC among AA,

Table 3. Means and S.E.M. of SCC for different DRB3/HaeIII genotypes.

Genotype	Somatic cell count (lakh/ml)	
	Hariana	Sahiwal
AA	3.96 ± 0.26b	3.88 ± 0.32
AB	3.19 ± 0.33ab	3.86 ± 0.54
BB	1.92 ± 0.29a	3.94 ± 1.04
BD	-	5.24 ± 1.17
EE	3.20 ± 0.98ab	3.18 ± 0.73
Overall mean	3.63± 0.21	3.91 ± 0.24

AB, BB, BD and EE genotypes in screened Hariana cattle. The BB genotype had lower SCC than AA, AB and EE genotypes. There was no significant different found for SCC in screened Sahiwal cattle in this investigation, which might be due to small sample size and presence of more number of alleles. Kumar et al. (2008) reported that genotype AA and EE were involved in resistance to mastitis in Nili Ravi breed of buffalo. Significant associations between SCC and various *DRB3* haplotypes and alleles were established by Oddgeirsson et al. (1988) in Canadian Holsteins.

In present study, the genotype AA had higher frequency among all the possible genotypes in screened cattle population. This finding was in concurrence with the reports of Aravindakshan and Nainar (1999) in Ongole, Wojdak-Maksymiec et al. (2010) in Jersey, Sharma et al. (2005) in Rathi. However, in contrast to this finding Aravindakshan and Nainar (1999) observed higher genotypic frequency of BB genotype in Jersey crossbred cattle. In the present study, A allele was the most frequent (0.660). Similar findings were reported by various authors in different cattle breeds including Argentinean Holstein, Caracu, Pantaneiro, Nelore, Gir (Miretti et al. 2001); Rathi (Sharma et al. 2005); Jersey (Wojdak-Maksymiec et al. 2010); Sahiwal (Chakraborty et al. 2015). In contrast, Miretti et al. (2001) and Thiruvaran and Bhushan (2015) observed higher allelic frequency of B in Argentinean creole and Umbalchery cattle, respectively.

In the present study, the *Hae*III/PCR-RFLP assay of 304 bp *DRB3* gene resulted in five genotypes, viz., AA, BB, AB, BD, EE with frequency ranging from 0.07-0.56 and four allele, viz., A, B, D, E with frequency range 0.035-0.660. AA genotype was the most frequent in all the screened cattle, followed by the AB, EE, BD and BB genotypes. Association study revealed no significant difference for milk production traits in first and second lactation except dry period (DP), which had significant difference among genotypes in second lactation. SCC has significant difference among genotypes in Hariana cattle, while in sahiwal cattle association was absent. However, the result showed that this region of *DRB3* gene was highly polymorphic, so this study needs further research in large population size and other different breeds of Indian cattle.

ACKNOWLEDGEMENT

The authors are thankful to Vice Chancellor, DUVASU, Mathura, (U.P.) for providing necessary facilities during entire research work at this esteemed university. The assistance of Instructional Livestock Farm Complex, DUVAU, Mathura, (U.P.) in providing blood samples of Sahiwal and Hariana cattle and data is duly acknowledged.

REFERENCES

Acharya CP, Pipallia DL, Rank DN, Joshi CG, Solanki JV and Shah RR. 2002. Detection of BOLA-DRB3 gene polymorphism in Gir and Kankrej cattle using PCR-RFLP. Indian Journal of Animal Sciences 72: 8.

Arvindakshan TV, Mahalinga NA and Sivaselvam SN. 2000. Polymorphism in exon 2 of the Bubu – MHC - DRB3 gene in indian buffalo (*Bubalus bubalis var. indicus*) detected by PCR – RFLP. Animal Science 70: 221-226.

Arvindakshan TV and Nainar NM. 1999. Genetic polymorphism of BoLA-DRB3 gene in Jersey crossbreed and Ongole cattle revealed by PCR-RFLP. Indian Journal of

Animal Sciences 69 (6): 424-427.

Baltian LR, Ripoli MV, Sanfilippo S, Takeshima SN, Aida Y, Giovambattista G. 2012. Association between BoLA-DRB3 and somatic cell count in Holstein cattle from Argentina. Molecular Biology Reports 39(7): 7215-7220.

Behl JD, Verma NK, Behl R, Mukesh M and Ahlawat SPS. 2007. Characterization of Genetic Polymorphism of the Bovine Lymphocyte Antigen DRB3.2 Locus in Kankrej Cattle (Bos indicus). Journal of Dairy Science 90:2997–3001.

Chakraborty D, Singh A, Tania MS, Verma A and Chakravarty AK. 2015. Genetic polymorphism of BOLA- DRB3.2 locus in sahiwal cattle. Animal Science Reporter 9: 33-40.

da Mota AF, Martinez ML and Coutinho LL. 2004. Genotyping BoLA-DRB3 alleles in Brazilian dairy cattle (Bos indicus) by temperature gradient gel electrophoresis (TGGE) and direct sequencing. European Journal of Immunogenetics 31:31–35.

Falconer DS and Mackay TFC. 1996. Introduction to Quantitative Genetics. 4th edn., Addison Wesley Longman Limited, England, pp.56.

Ganguly I, Sharma A, De S and Mitra A (2004). Genetic polymorphism of BoLA-DRB 3.2 locus in crossbred cattle as revealed by PCR-RFLP analysis. Indian Journal of Animal Sciences 74 (2): 195-199.

Gelhus A, Schnittger L, Mehlitz D, Horstmann R, Meyer C. 1995. Sequence and PCR-RFLP analysis of 14 novel BolA-DRB3 alleles. Animal Genetics 26:147-153.

Wojdak-Maksymiec K, Kmie M, Kowalewska L, Warlinski M. 2010. DRB3 gene polymorphism and somatic cell count in milk of Jersey cows. Journal of Animal and Veterinary Advances 9(9): 1295-1300.

Kumar S, Sangwan ML and Rupender. 2008. Polymorphism in DRB3 exon 2 by PCR-RFLP and its association with mastitis in Nili-Ravi breed. Indian Journal of Biotechnology 398-400.

Ledwidge SA, Mallard BA, Gibson JP, Jansen GB and Jiang ZH. 2001. Multi-primer target PCR for rapid identification of bovine DRB3 alleles. Animal Genetics 32:219- 221.

Mejdell CM, Lie O, Solbu H, Arnet EF and Spooner RL. 1994. Associations of major histocompatibility complexnantigens (BoLA-A) with AI bull progeny test results for mastitis, ketosis, and fertility in Norwgian cattle. Animal Genetics 25: 99–104.

Miretti MM, Ferro JA, Lara MA and Contel EPB. 2001. Restriction fragment length polymorphism (RFLP) in exon 2 of the BoLA-DRB3 gene in South American cattle. Biochemical Genetics 39: 9-10.

Nassiry MR, Shahroodi FE, Mossafer J, Mohammadi A, Manshad E, Ghazanfari S, Mohammad Abadi MR and Sulimova GE. 2005. Analysis and frequency of Bovine Lymphocyte Antigen (BoLA-DRB3) alleles in Iranian Holstein cattle. Russian Journal of Genetics 41:817–822.

Oddgeirsson O, Simpson SP, Morgan AL, Ross DS, Spooner RL. 1988. Relationship between the bovine major histocompatibility complex (BoLA), erythrocyte markers and susceptibility to mastitis in Icelandic cattle. Animal Genetics 19: 11–16.

Ogorevc J, Kunej T, Razpet A and Dovc P. 2009. Database of cattle candidate genes and genetic markers for milk production and mastitis. Animal Genetics 40: 832–851.

Parmentier I, Portetelle D, Gengler N, Prandi A, Bertozzi C, Vleurick L, Gilson R, Renaville R. 1999. Candidate gene markers associated with somatotropic axis and milk selection. Domestic animal endocrinology 17: 139-148.

Rupp R, Hernandez A, Mallard BA. 2007. Association of bovine leukocyte antigen (BoLA) DRB3.2 with immune response, mastitis, and

production and type traits in Canadian Holsteins. Journal of Dairy Science 90(2):1029–1038.

Sambrook J and Russell D. 2001. Molecular Cloning: A Laboratory Manual 3rd edn. Cold Spring Harbor Laboratory Press, Cold Spring Harbor, New York, pp: 6.4-6.11.

Sharif S, Mallard BA, Wilkie BN, Sargeant JM, Scott HM, Dekkers JCM and Leslie KE. 1998 Associations of the bovine major histocompatibility complex DRB3 with production traits in Canadian dairy cattle. Animal Genetics 30:157-160.

Sharma AK, Bhushan B, Kumar P, Sharma D, Saxena VK, Sharma AR and Kumar S. 2005. DNA Polymorphism of DRB 3.2 locus and its association with serum Lysozyme in Rathi cattle (Bos indicus). Journal of Applied Animal Research 28: 61-64.

Siguardardtottir S, Borsch C, Gustafasson K and Andersson L. 1991. Cloning and sequencing analysis of 14 DRB alleles bovine MHC by PCR. Animal Genetics 22: 199-209.

Singh S, Bharat B, Kumar S, Kumar P and Sharma A. 2004. Genetic polymorphism of DRB3.2 gene in Murrah buffalo by PCR-RFLP. Indian Journal of Animal Sciences 74: 432-434.

Stafuzza NB, Olivatto LM, Naressi BCM, Tonhati H and Amaral-Trusty MEJ. 2015. Analysis of DRB3 gene polymorphisms in Jafarabadi, Mediterranean, and Murrah buffaloes from Brazil. Genetics in Molecular Research 15(1): gmr. 15016368.

Starkenburg RJ, Hansen LB, Kehrli ME and Chester H. 1997. Frequencies and effects of alternative DRB3.2 alleles of bovine lymphocyte antigen for Holsteins in milk selection and control lines. Journal of Dairy Science 80: 3411-3419.

Thirumaran SMK and Bhushan B. 2015. Genetic polymorphism of the bovine MHC DRB3 exon 2 of Umblachery cattle distinguished by polymerase chain reaction-restriction fragment length polymorphism (PCR-RFLP). Indian Veterinary Journal 92(3):14-17.

Van Eijk MJT, Stewart-Haynes JA and Lewin HA. 1992. Extensive polymorphism of the BoLA-DRB3 gene distinguished by PCR-RFLP. Animal Genetics 23:483-496.

Growth performance of Chhotanagpuri sheep under field condition

Maroof Ahmad* and D K Singh
Department of Animal Genetics & Breeding, College of Veterinary Science and Animal Husbandry, Birsa Agricultural University, Ranchi 834 006, India

ABSTRACT

Present study was undertaken on 242 Chhotanagpuri lambs in tribal areas of Deoghar District of Jharkhand. The body weights at different ages viz. birth weight (BWT), 3-month (3-WT), 6-month (6-WT), 9-month (9-WT) and 12-month (12-WT) were recorded for the study. The overall body weights of Chhotanagpuri lambs were 1.76± 0.06, 6.52 ± 0.14, 10.43±0.17, 13.44±0.20 and 16.44±0.18 kg at birth, 3, 6, 9 and 12-month of age, respectively. Lambs born during winter season were significantly heavier at birth than those born during summer and monsoon.Male lambs weighted significantly heavier than the female lambs at different stages of growth. Body weight of male lambs at birth, 3, 6, 9 and 12-months of age were 1.86±0.04, 7.02±0.12, 11.02±0.13, 14.16±0.24 and 17.64±0.23 kg respectively. The corresponding values for females were 1.65 ± 0.04, 6.01 ± 0.18, 9.83 ± 0.22, 12.72 ±0.18 and 15.24±0.12 kg. The variations in birth weight and weights at different stages of growth up to 12-month of age due to dam's weight at lambing were also significant. The average daily gain in body weights of Chhotanagpuri lambs were estimated as 52.88 ± 1.44, 43.44±1.10 and 33.38±1.21 g/day during 0-3, 3-6 and 6-12 month of age, respectively.

Key Words: Chhotanagpuri sheep, growth traits, body weight gain
***Corresponding author:** maroofahmad786@yahoo.co.in
Manuscript received: 12.4.2017; Manuscript accepted: 15.6.2017

INTRODUCTION

Chhotanagpuri is the only recognized breed of sheep found in Jharkhand. The breed is distributed in the entire area of Jharkhand including adjacent districts of West Bengal (Purulia, Western part of Bankura and west of Medinapur district). These animals are mostly in the hands of weaker sections of the society. The size of the breed is small having straight nose, short drooping or horizontal ears and short tail. These animals are being maintained solely for mutton production. They produce coarse wool of poor quality, which may be utilized for carpet manufacturing. The information on growth parameters on Chhotanagpuri under field condition is scantyHence, this study was conducted to estimate the pattern of growth in Chhotanagpuri sheep under prevailing natural and ecological conditions of Jharkhand, India.

MATERIALS AND METHODS

Data on 242 Chhotanagpuri lambs belonging to Deoghar district of Jharkhand were used in the present investigation. The data was generated under Mega Sheep Deed Project of ICAR at Birsa agricultural University, Ranchi, Jharkhand. The average elevation of the district is 247 m above msl. Average annual rainfall is 1239 mm, mean summer maximum temperature is 43°C and mean winter minimum temperature is 8 °C. The body weights at different ages viz. birth weight (BWT), 3-month (3-WT), 6-month (6-WT), 9-month (9-WT),12-month (12-WT) and average daily gain were considered for the study. All the observations were taken in the morning before grazing or being allowed feed or water to the animals. The season was divided into three: the main winter (November to February), summer (March-June) and mansoon (July-October). The weight was recorded with the help of 120 kg weighing balance with 100 g accuracy. The animals were maintained only on grazing and allowed for 6-7 hours during day on natural grasses and shrubs. The data collected were analyzed by least-squires method (Harvey, 1990).

RESULTS AND DISCUSSION

The overall body weights of Chhotanagpuri lambs were 1.76± 0.06, 6.52 ± 0.14, 10.43±0.17, 3.44 ±0.20 and 16.44±0.18 kg at birth, 3, 6, 9 and 12-month of age, respectively. Higher values for these traits were reported by Kushwaha (1994), Kushwaha *et al.* (1997) and Kumar (2000) in various breeds of sheep.

Effect of seasons

Lambs born during winter season were significantly heavier at birth than those born during summer and monsoon (Table 1), which did not differ significantly between themselves. This could be probably due to availability of better quality of green fodder during post monsoon and winter season to ewes in gestation as majority of winter lambing occurred in the month of February. Winter born lambs continued to grow heavier than monsoon born lambs but the superiority over summer born lambs disappeared by reaching weaning age.. Winter born lambs weighed significantly heavier than those born during monsoon. Highly significant effect due to season of birth was also reported by Tomar *et al.* (2000).. Non-significant effect due to season of birth have been reported by Singh and Dhillon (1992) and Swain *et al.* (1994) for birth weight and Singh and Dhillon (1992) for 6 and 12 month of body weights in Avikalin sheep

Effect of sex

Male lambs weighted significantly heavier than the female lambs at different stages of growth. Body weight of male lambs at birth, 3, 6, 9 and 12-months of age were 1.86±0.04, 7.02±0.12, 11.02±0.13, 14.16±0.24 and 17.64±0.23 kg respectively. The corresponding values for females were 1.65 ± 0.04, 6.01 ± 0.18, 9.83 ± 0.22, 12.72±0.18 and 15.24±0.12 kg. Non-significant effect due to sex of lamb was observed by Kandasamy *et al.* (1980) on birth weight and Kumar (2000) for weaning weight in various sheep breeds.

Effect of dam's weight at lambing

The variations in birth weight and weights at different stages of growth up to 12-month of age due to dam's weight at lambing were also significant. Ewes weighed 20.00 kg or more at lambing delivered significantly heavier lambs (Table 1) as compared to those weighed 10.00-15.00 and 15.00-20.00 kg at lambing. These groups differed significantly among themselves. Lambs produced by heavier ewes continued to weigh heavier up to 12-month of age than those produced by lighter ones. The average body weights of lambs of ewes weighted ≥20.00 Kg at lambing were 2.03 ± 0.04, 7.72 ± 0.09, 11.08 ± 0.13, 15.22 ± 0.15 and 17.64 ± 0.16 kg at birth, 3, 6, 9 and 12-month of age.

Average Daily gain in body weights

The average daily gain in body weights of Chhotanagpuri lambs were estimated as 52.88 ±

Table 1: Least squares means of body weights (kg) of Chhotanagpuri lambs

Effect	Body weights				
	Birth	**3-M**	**6-M**	**9-M**	**12-M**
Overall(μ)	1.76 ± 0.06 (242)	6.52 ± 0.14(234)	10.43 ± 0.17(217)	13.44 ± 0.20(206)	16.44 ± 0.18(192)
Seasons of birth	*	*	**	**	*
Winter	1.81 ± 0.03^{a}(118)	6.70 ± 0.04^{a}(115)	10.94 ± 0.14^{a}(108)	14.72±0.25^{a}(102)	17.46±0.25^{a}(96)
Summer	1.76 ± 0.04^{b} (87)	6.62 ± 0.07^{a} (85)	10.45 ± 0.15^{ab}(81)	13.38±0.26^{b}(77)	16.64±0.27^{ab}(72)
Monsoon	1.71 ± 0.02^{b}(37)	6.23 ± 0.28^{b}(34)	9.89 ± 0.26^{b}(28)	12.26± 0.13^{c}(27)	15.24±0.11^{b}(24)
Sex	*	**	**	**	**
Male	1.86 ± 0.04^{a} (127)	7.02 ± 0.12^{a}(124)	11.02 ± 0.13^{a}(114)	14.16±0.24^{a}(108)	17.64±0.23^{a}(102)
Female	1.65 ± 0.04^{b} (115)	6.01 ± 0.18^{b}(110)	9.83 ± 0.22 ^{b}(103)	12.72±0.18^{b}(98)	15.24±0.12^{b}(90)
Dam's wt. at lambing	*	**	*	*	*
10.00- 15.00	1.45 ± 0.05^{c} (77)	5.06 ± 0.20^{c} (75)	9.43 ± 0.22^{b}(67)	12.08±0.23^{c}(61)	15.33±0.20^{c}(54)
15.00- 20.00	1.79 ± 0.02^{b} (133)	6.80 ± 0.19^{b}(129)	10.79 ± 0.20^{a}(122)	13.05±0.21^{b}(117)	16.36±0.17^{b}(112)
≥ 20.00	2.03 ± 0.04^{a} (32)	7.72 ± 0.09^{a}(30)	11.08 ± 0.13^{a}(28)	15.22±0.15^{a}(28)	17.64±0.16^{a}(26)

*P≤0.05, ** P≤0.01; Values bearing different superscripts for an effect in a column differed significantly,
Figures in parentheses are number of observations.

Table2: Least-squares means of average daily gain (g/day) in body weight of Chhotanagpuri lambs

Effect	Daily gain in body weights		
	ADG-1 0-3 month	ADG-2 3-6 month	ADG-3 6-12 month
Overall(μ)	52.88 ± 1.44 (234)	43.44±1.10 (217)	33.38±1.21 (192)
Seasons of birth	*		**
Winter	53.74 ± 1.65 [a] (115)	43.68 ± 1.14 (108)	34.24±1.16[a] (96)
Summer	52.84 ± 1.73[ab] (85)	44.33 ± 1.14 (81)	34.06±1.14[a] (72)
Monsoon	50.78 ± 1.02[b] (34)	44.36 ± 0.94 (28)	32.15±1.27[b] (24)
Sex	**	**	**
Male	56.84 ± 1.76[a] (124)	46.29 ± 1.14[a] (114)	35.78 ± 1.14[a] (102)
Female	49.07 ± 1.46[b] (110)	42.89 ± 1.22[b] (103)	31.52 ± 1.22[b] (90)
Dam's wt. (kg) at lambing	**	*	**
10.00- 15.00	42.12 ± 1.62[c] (75)	48.33 ± 1.14[a] (67)	32.72 ± 1.14[a] (54)
15.00- 20.00	54.78 ± 1.70[b] (129)	44.27 ± 1.26[b] (122)	32.05 ± 1.12[a] (112)
≥ 20.00	61.64 ± 1.36[a] (30)	44.21 ± 0.92[b] (28)	36.54 ±1.31[b] (26)

*P≤0.05, ** P≤0.01; Values bearing different superscripts for an effect in a column differed significantly;
Figures in parentheses are number of observations.

1.44, 43.44±1.10 and 33.38±1.21 g/day during 0-3, 3-6 and 6-12 month of age, respectively. Higher values for average daily gain were observed by Sinha and Singh (1997) in Muzaffarnagri sheep. A significant effect of seasons of birth on ADG was observed during 0-3 and 6-12 month of age. However, variations due to sex were significant (P≥0.01) on ADG during different periods of growth under observation. There was significant effect of ewe's weight at lambing on ADG during 0-3, 3-6 and 6-12 month of age. The average daily gain in body weight of winter born lambs during 0-3 (53.74 ± 1.65 g/day) and 6-12 (34.24 ± 1.16 g/day) month of age was significantly more than those born during monsoon (50.78 ± 1.02 and 32.15 ± 1.27 g/day). The differences between lambs born during summer and monsoon seasons for average daily gain in body during 0-3 month of age was not significant but it differed significantly during 6-12 months of age. The effects of sex on daily gain in body weight of lambs during 0-3, 3-6 and 6-12 months of age were significant. Males had significantly higher daily gain in body weight than females.

The variations in average daily gain in body weights during 0-3, 3-6 and 6-12 months of age due to dam's weight at lambing were significant. Lambs of ewes weighed ≥ 20.0 kg at lambing had significantly higher daily gain in body weight (Table 2) during 0-3 months of age than those of ewes weighed 15-20 and 10-15 kg at lambing. During 6-12 months of age, the lambs of ewes weighed ≥ 20.0 kg at lambing had significantly higher daily gain in body weight.

Venkateswarlu and Ramana **(**2013) reported significantly increase in weight gains in concentrate supplemented lambs than sole grazed ram lambs. Higher ADG values were also reported by Chaturvedi *etal.* (2009) in lambs maintained on 8 hours grazing plus supplementation of concentrate at 300g per animal day.

REFERENCES

Chaturvedi OH, Sushil K, Misra AK, Arora AL and Karim SA. 2009. Effect of complete feed or grazing and supplementation of lambs on performance, nutrient utilization and feed cost of production. Indian Journal of Animal Sciences 79: 917-920.

Harvey, W R. 1990. User guide for LSMLMW pc-1 version mixed model least-squires and maximum likelihood computer program. Columbus, Ohio, USA.

Venkateswarlu M and Ramana R Yerradoddi. 2013. Effect of concentrate supplementation on growth and carcass characteristics in grazing ram lambs. International Journal of Food, Agriculture and Veterinary Sciences 3:43-48.

Kandasamy N, Mehta BS, Gupta DC and Arora CL. 1980. A preliminary analysis of preweaning and pot weaning body weights of Nali, Chokla and their crosses with Rambouillet and Soviet Merino. Wool and Woolen of India 17:3.

Kumar R. 2000. Title of Thesis Ph. D. Thesis, G.B. Pant University of Agriculture and Technology, Pantnagar, U.S. Nagar, India.

Singh G and Dhillon JS. 1992. Factors affecting body weights of Avikalin lambs. Indian Journal of Animal Sciences 62: 855-860.

Sinha NK and Singh SK. 1997. Genetic and phenotypic parameters of body weights, average daily gain and first shearing wool yield in Muzaffarnagri sheep. Small Ruminants Research 26: 21-29.

Swain N, Gopikrishna G, Gaur D and Sanyal PK. 1994. Genetic and non-genetic factors affecting birth weight in Bharat Merino sheep in southern sub-temperature region of Tamil Nadu. Indian Journal of Animal Sciences 9:141-142.

Tomar AKS, Mehta BS, Jaishankar J. and Singh G. 2000. Genetic studies on growth and greasy fleece yield in Bharat Merino sheep. Indian Journal of Animal Sciences 70:537-539.

Details and Instructions for Submitting Manuscript to the Journal of Livestock Biodiversity

Name of the Journal	Journal of Livestock Biodiversity
Periodicity of publication	Half yearly

Submission of Manuscript

One original and two copies of research/review manuscripts, not submitted elsewhere for publication, should be submitted to the Executive Editor. The copyright of the paper will be transferred from the author(s) to publisher. It is mandatory to furnish the following certificate along with the manuscript:

This is to certify that the reported work in the paper entitled "---------- " submitted for publication in the "Journal of Livestock Biodiversity" is an original one and has not been submitted for publication elsewhere. I/we further certify that proper citations to the previously reported work have been given and no data/tables/figures have been quoted verbatim from other publications without giving due acknowledgement and without the permission of the author(s). The consent of all the authors of this paper has been obtained for submitting the paper to the "Journal of Livestock Biodiversity".

Signatures and Names of all the Authors

Preparation of Manuscript

Manuscripts should be typed in double space (12 pt, Times New Roman font preferred) on one side of the bond paper of 22x28 cm. All pages should be numbered consecutively. Use SI units, and give equivalent SI units in parenthesis when the use of other units is unavoidable. Symbols should conform to standard guidelines.

Title – Title should be short and informative (15 pt). Only first letter to be typed in capital. Latin names are to be given in italics.

Short Running Title – Not in more than 50 characters, to be all in capitals

Keywords – Five or six keywords (in normal 11 pt) indicating the contents of the manuscript.

Authors – Names of the authors to be typed in first letter capital (12 pt)

Addresses of Authors – Addresses of the institution(s) where the work was carried out including telephone (office only), fax number and e-mail address (10 pt). Author for correspondence should be indicated with an asterisk (*).

Main Headings – Each manuscript should be divided into the following main headings (typed in bold, first letters capital, on the left hand side of the page; 12 pt): Abstract, Introduction, Materials and Methods, Results, Discussion, Acknowledgements, References.

Abstract – Should be brief not exceeding 200 words, typed in normal (11 pt)

Introduction – A brief and precise literature review with essential background and objectives of the research undertaken.

Material and Methods : It should include the relevant details of the source and nature of material, experimental design and the techniques employed. New methods should be described in sufficient details and other can be referred to published work.

Results : Results should contain data, essential for drawing main conclusion from the study. Wherever needed, the data should be statistically analyzed. The data should not be repeated in both table and figure.

Discussion : The discussion should be deal the interpretation of results. Preferably, results and discussion should be combined to avoid repetition.

Conventions : Authors should follow internationally accepted conventions in regard to units, symbols and abbreviations. Only SI units measurement and standard abbreviations should be used. Binomial names of organisms are italicized.

Tables : Tables should be typed in double space on separate sheets, numbered consecutively. All tables should be numbered serially, in order of appearance.

Illustrations : The line drawings, illustrations, photographs etc will be accepted as hard copies. All figures should be numbered, serially in order of appearance. Figure legends should be typed on a separate sheet or sheets, which should be numbered as part of the manuscript (at the end). Photographs should be sharp, high-contrast, glossy prints. Color photographs can be accepted only when absolutely necessary and against actual cost of printing to the author/s.

References : Citations in the text should be by name and year, in chronological order and then alphabetically for the same year, and enclosed in parentheses. The following pattern for research papers, books etc may be followed :

Wilesmith J, Wells G, Ryan J, Gavier-Widen, D and Simmons, M. 1997. A cohort study to examine maternally associated risk factors for bovine spongiform encephalopathy. Veterinary Record 111: 230-243.

Sambrook J, Fritsch EF and Maniatis T. 1989 Molecular cloning a laboratory manual, 2nd edition. Cold Spring Harbor Laboratory Press, Cold Spring Harbor.

Fries R and Propescu P. 1999. Cytogenetics and physical chromosome maps. In: The Genetics of Cattle (eds. R. Fries and A. Ruvinsky), pp. 247-372. CAB International 1999.

Acknowledgements : These should follow immediately after the end of the main text. In references to granting agencies, the names should be written in full.

Copyright : Society for Conservation of Domestic Animal Biodiversity (SOCDAB), publisher of the journal, will acquire copyright over all published material. Authors may reproduce their published material elsewhere subsequently with the usual acknowledgement to 'Journal of Livestock Biodiversity, published by 'SOCDAB' and the volume and page details, but a request to do so will be appreciated and also serve to keep the editorial office informed. Third parties who wish to reproduce published should write to the editorial office for permission.

JOURNAL OF LIVESTOCK BIODIVERSITY
Article Certificate

F. No.
Title of the Article :
Title Changed to :
Author/s name/s :

It is certified that :

1. All the authors have gone through the article and are satisfied with its contents and presentation.
2. The names of authors in the by-line incorporate all the workers who made notable input in the article.
3. The order of names in the by-line is agreeable to all the authors.
4. The adress of the organization where the research was conducted is given in the by-line (change of author's address is given in the footnote).
5. The article is exclusive for this journal, and the results reported in this article have not been/will not be sent (during its consideration) for publication in any other journal.
6. The article has not been rejected for publication in any other journal/rejected in _____ whose comments do not prohibit this article for consideration in this journal and are attached with the article.
7. It is based in part/complete M.Sc/Ph.D thesis work of the first author who submitted the thesis in ----- to the ---------- (University, location).
8. Truthful data and facts are presented in the article.
9. I/we agree to accept the objective comments of the references and do agree to moderate the article into a short note if recommended so for publication in the Journal of Livestock Biodiversity.
10. On publication in the Journal of Livestock Biodiversity (SOCDAB), who will have the absolute right to enter into any agreement with any organization in India or abroad engaged in reprography, photocopying, storage and dissemination of information contained in it and neither we nor any of our legal heirs will have any legal claims on royalty, if any.

Signature of all authors, their names, designations and present correspondence address

Name and designation	Present official address	Signature with date
1.		
2.		
3.		

Optional

The above certificate is correct to the best of my knowledge and I have no objection to the publication of the article cited above in the Journal of Livestock Biodiversity.

Signature & Address of Head of organization

Journal of Livestock Biodiversity

Published by:
Society for Conservation of Domestic Animal Biodiversity