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Haematology of Jaffarabadi buffaloes on an established farm: Effect of various physiological parameters

Ninan Jacob, Arya JS, Padodara RJ and Gajbhiye PU

Abstract
Study was conducted on male and female Jaffarabadi buffaloes from one week of age to 36 months of age and at various lactating and non-lactating stages. The haematological parameters were determined using Auto Hemato Analyser and the data generated was analysed to note the differences between age, sex, lactating stages and non-lactating stages. Age had a significant (P<0.05) effect on TEC levels in both the sexes of Jaffarabadi buffaloes. Significant (P<0.05) difference was noted in the TEC during lactation, whereas non-significant effect was observed between non-lactating animals in Jaffarabadi species. The TEC in males were found to be lower than in females. Age exerted significant (P<0.05) effect on Hb. and PCV levels in Jaffarabadi males but had no significant influence in Jaffarabadi females. Non-significant differences were noted in Hb. and PCV levels at different lactating and non-lactating stages in Jaffarabadi buffaloes. Significant (P<0.05) effect of age was seen in Jaffarabadi males and Jaffarabadi females for MCV, MCH and in Jaffarabadi females for MCHC values. Non-significant difference existed between lactating stages for all three indices in Jaffarabadi buffaloes. Age exerted a significant (P<0.05) effect on TLC values in both the sexes of Jaffarabadi species. Non-significant difference was noted during lactation stages and between non-lactating stages in Jaffarabadi buffaloes.

Keywords: Age, buffaloes, haematology, Jaffarabadi, lactation, sex

1. Introduction
The Jaffarabadi buffalo is a riverine buffalo that originated in Gujarat and is a hybrid of African cape buffalo and Indian water buffalo [1]. The name is derived from their main tract location that is Jaffarabad. It is noted for its higher fat content and milk production [2]. In 2007, they constituted 14.70 lakh of the total 87.73 lakh buffalo population of Gujarat [3]. The Jaffarabadi buffalo has an average lactation length of 319 days with the average 7.86% fat. Milk production of Jaffarabadi buffaloes under field condition ranged from 800 – 2800 lt / lactation with an average of 1681.75 lt / lactation [4]. The lactation length and yield is highest among all Indian Buffalo breeds. Buffaloes have better survival ability under Indian conditions as they are well adapted to a hot and hot humid climate and play a distinct role in the economy of farmers, which is primarily based on agricultural production systems. It is necessary to establish the norms of haematological values for a particular breed and for the particular region, as the clinical experimental investigation and interpretation of results for general health evaluation and subsequent production and reproduction aspects depend upon these normal values. The importance of haemato-biochemical indices in animal husbandry is well established. However, these indices may vary depending on the factors such as age, sex, weather, stress, season and physiological stage of the animal [5]. Hence, the physiological variation in the blood parameters is of great significance for judging the health status of animals and their production and reproduction as blood constituents are indicative of environmental stress in buffaloes of tropical regions [6]. The present study was undertaken on Jaffarabadi buffaloes to study the effect of physiological variations on haematology with an aim to establish base line haematological values for the species.

2. Materials and Methods
The study was carried out on male (n = 6 at each stage) and female (n = 8 at each stage) Jaffarabadi buffaloes of various ages (1 wk to 36 m) and physiological stages (Lactating: 1 – 3 m and Non-lactating: Pregnant and non-pregnant) maintained at the Cattle Breeding Farm, Junagadh Agricultural University, Junagadh, Gujarat. The haematological evaluation was carried out at the Department of Physiology and Biochemistry,

College of Veterinary Science and Animal Husbandry, Junagadh Agricultural University, Junagadh, Gujarat. The project was approved by the Institutional Animal Ethics Committee (IAEC). The animals were maintained under standard feeding and management conditions followed at the Cattle Breeding Farm. The animals were separated from the herd only for the duration of blood collection. Two ml of fresh whole blood collected aseptically, between 0600 hrs to 0800 hrs of the day in K_2 EDTA vacuettes was subjected immediately for haematological analyses viz. TEC, Hb, PCV, MCV, MCH, MCHC and TLC values using Auto Hemato Analyzer (Model: BC – 2800 Vet, Mindray). The data obtained in the present study were subjected to statistical analyses by using the Completely Randomised Design (CRD) [7].

3. Results and Discussion

The Mean ± S.E. haematological values at different ages for male and female Jaffarabadi buffaloes are given in Table -1 and the haematological values at different lactating and non-lactating stages are presented in Table – 2.

3.1 Total erythrocyte count (TEC)

The count of total erythrocytes (10^6/cmm) at all ages and physiological stages ranged from 7.05 ± 0.26 (1 m lactation stage) to 11.03 ± 0.35 (1 wk age) in Jaffarabadi females. The count obtained at 1 wk age showed non-significant decrease at 1 m age. The values further decreased significantly (P<0.05) at 3 m and 6 m age, and then exhibited non-significant decrease upto 24 m age. A non-significant increase was found at 36 m age from that of 24 m age. Total erythrocytes increased non-significantly from 1 m lactation to 2 m lactation, and the levels at 3 m lactation were significantly (P<0.05) higher than that observed at 1 m and 2 m lactation. Non-significantly lower values were observed in non-lactating non-pregnant buffaloes as compared to the pregnant buffaloes. The range of 7.36 ± 0.17 (12 m age) to 10.81 ± 0.43 (1 wk age) was recorded in Jaffarabadi males across all ages and stages studied. A gradual significant decrease was found in the values from 1 wk to 12 m age with the values at 12 m being the lowest among all the ages studied here. No significant differences were observed in total erythrocyte count between Jaffarabadi females and males. The counts at all ages were lower in males than that in females, except at 6 m where the count was found to be higher in Jaffarabadi males than in females. The finding in the present study of decrease in TEC with increase in age in Jaffarabadi females and males was in accordance to the findings of [8] in three different breeds of buffaloes, in Iranian buffaloes [9], and in Murrah buffaloes [10, 11, 12]. The finding of higher TEC in females than in males was in agreement with the reports in African buffaloes [13]. The current findings that TEC in pregnant animals was higher than that in non-pregnant animals agreed with that reported by [14] but not with [15]. The values obtained in present study in all ages and lactating and non-lactating stages in Jaffarabadi buffaloes were higher than those reported in lactating and non-lactating Nili-Ravi buffaloes [16], and in Murrah buffaloes [10, 15]. However, they were in the range obtained by [17] in Murrah buffaloes and in Iranian buffaloes [9]. The TEC values were the highest at 1 wk age with regard to age. Significant haematological changes seen in Jaffarabadi buffaloes as age advanced may be due to increased destruction of erythrocytes, maturational changes of lymphoid organs and environment induced adaptative changes with increasing age [11]. The higher TEC levels observed in our study at 1 wk age is supported by our findings of higher cortisol and thyroid hormones at 1 wk of age. It was opined that glucocorticoids promote the differentiation of embryonic stem cells to hematopoietic cells and prolong the proliferation of erythroid progenitor cells but reduce the rate of spontaneous differentiation and terminal maturation of erythroid cells [18]. Further, thyroid hormone (T_3) promotes differentiation and maturation of erythroid cells toward enucleated Red Blood Cells. Thyroid hormone also promotes synthesis of erythropoietin in kidney [19]. The higher counts in females as compared to the males and also the higher values in pregnant buffaloes as compared to non-pregnant and those in early stages of lactation could be due to the higher plane of nutrition fed to the female calves and to advanced pregnant animals. Nutrition has a significant effect on hematologic values [20]. Increasing counts of erythrocytes as lactation advanced could be attributed to the higher blood volume present in these animals for supply to the mammary gland for milk production.

Table 1: Sex related haematology at different ages in Jaffarabadi buffaloes (Mean ± S.E.).

Age/Animal			Wk	Months					
			1	1	3	6	12	24	36
TEC	10^6/cmm	F	$11.03^{c} \pm 0.35$	$10.77^{c} \pm 0.55$	$9.58^{b} \pm 0.61$	$7.87^{a} \pm 0.24$	$7.66^{a} \pm 0.32$	$7.51^{a} \pm 0.33$	$8.39^{a} \pm 0.28$
		M	$10.81^{c} \pm 0.43$	$10.68^{c} \pm 0.23$	$8.89^{b} \pm 0.16$	$8.15^{ab} \pm 0.39$	$7.36^{a} \pm 0.17$	-	-
Hb	g/dl	F	15.93 ± 0.29	15.73 ± 0.75	14.14 ± 0.73	15.26 ± 0.53	14.60 ± 0.45	14.75 ± 0.64	16.67 ± 0.69
		M	$16.30^{b} \pm 0.32$	$15.52^{b} \pm 0.57$	$13.97^{ab} \pm 0.28$	$16.46^{b} \pm 0.53$	$13.38^{a} \pm 0.56$	-	-
PCV	%	F	42.31 ± 0.82	41.79 ± 2.09	40.40 ± 2.13	40.73 ± 1.27	38.43 ± 1.20	38.80 ± 1.72	43.83 ± 1.73
		M	$43.13^{b} \pm 1.07$	$41.32^{ab} \pm 1.27$	$38.98^{ab} \pm 1.22$	$43.15^{b} \pm 1.42$	$36.88^{a} \pm 0.94$	-	-
MCV	Fl	F	$38.55^{a} \pm 0.79$	$38.94^{a} \pm 0.87$	$42.58^{b} \pm 1.39$	$51.93^{c} \pm 1.12$	$50.48^{c} \pm 1.13$	$51.88^{c} \pm 1.43$	$52.28^{c} \pm 1.17$
		M	$40.22^{abw} \pm 1.74$	$38.80^{a} \pm 1.26$	$43.97^{b} \pm 1.60$	$53.35^{c} \pm 1.92$	$50.32^{c} \pm 1.51$	-	-
MCH	Pg	F	$14.46^{a} \pm 0.29$	$14.61^{a} \pm 0.31$	$14.91^{a} \pm 0.63$	$19.35^{b} \pm 0.31$	$19.13^{b} \pm 0.46$	$19.61^{b} \pm 0.46$	$19.78^{b} \pm 0.44$
		M	$15.12^{a} \pm 0.49$	$14.50^{a} \pm 0.56$	$15.70^{a} \pm 0.47$	$20.28^{c} \pm 0.57$	$18.15^{b} \pm 0.78$	-	-
MCHC	%	F	$37.60^{b} \pm 0.22$	$37.60^{b} \pm 0.12$	$35.10^{a} \pm 1.05$	$37.40^{b} \pm 0.43$	$37.96^{b} \pm 0.21$	$37.98^{b} \pm 0.18$	$37.96^{b} \pm 0.15$
		M	37.45 ± 0.29	37.48 ± 0.41	35.88 ± 0.58	36.47 ± 1.57	36.20 ± 0.89	-	-
TLC	10^3/cmm	F	$7.90^{a} \pm 0.28$	$8.20^{bc} \pm 0.32$	$6.74^{a} \pm 0.33$	$9.26^{cx} \pm 0.39$	$8.70^{bc} \pm 0.34$	$8.09^{b} \pm 0.50$	$8.33^{bc} \pm 0.25$
		M	$7.41^{a} \pm 0.24$	$7.50^{a} \pm 0.29$	$7.65^{a} \pm 0.37$	$7.50^{ay} \pm 0.23$	$8.82^{b} \pm 0.26$	-	-

Note: Means having the same superscript do not differ significantly from each other (P<0.05).
F: n = 8; M: n = 6.
Key: F: Female M: Male
Superscripts: a – e : between females and between males
x – y : between male and female

Table 2: Hematology at different lactation months in Jaffarabadi buffaloes (Mean ± S.E.).

Age/Animal		Lactation Months			Non Lactating	
		1	2	3	P	NP
TEC	10^6/cmm	7.05^{h} ± 0.26	7.36^{h} ± 0.17	8.81^{i} ± 0.67	8.04 ± 0.55	7.68 ± 0.26
Hb	g/dl	14.76 ± 0.39	15.91 ± 0.38	16.13 ± 1.56	16.63 ± 0.96	16.65 ± 0.75
PCV	%	39.08 ± 1.07	42.03 ± 1.08	42.41 ± 3.97	44.83 ± 2.59	43.88 ± 1.98
MCV	fl	55.73 ± 1.49	57.39 ± 1.80	53.50 ± 2.89	56.11 ± 1.00	57.16 ± 1.48
MCH	pg	20.98 ± 0.52	21.64 ± 0.62	20.39 ± 0.95	20.75 ± 0.38	21.60 ± 0.51
MCHC	%	37.76 ± 0.19	37.80 ± 0.12	37.93 ± 0.23	37.09 ± 0.62	37.90 ± 0.13
TLC	10^3/cmm	8.19 ± 0.45	8.66 ± 0.29	8.20 ± 0.35	8.06 ± 0.52	7.34 ± 0.53

Note: Means having the same superscript do not differ significantly from each other (P<0.05).
P – Pregnant, NP – Non Pregnant
Superscripts: h – j: within lactating Jaffarabadi

3.2 Haemoglobin (Hb)

The levels of haemoglobin (g/dl) in Jaffarabadi females at all ages and physiological stages studied ranged from 14.14 ± 0.73 (3 m age) to 16.67 ± 0.69 (36 m age). No significant difference was observed from 1 wk age to 36 m age in haemoglobin levels. The values showed a non-significantly decreasing trend upto 12 m age from which it increased non-significantly upto 36 m age. In lactating Jaffarabadi buffaloes, a progressive non-significant increase was found from 1 m lactation to 3 m lactation stage. The difference in the haemoglobin levels between lactating pregnant and non-pregnant non-lactating Jaffarabadi buffaloes was non-significant. The range of haemoglobin values observed in Jaffarabadi buffaloes was higher than those recorded by [14, 15, 21, 22] in various breeds of buffaloes. The observation of significant (P<0.05) decrease in the values as age increased was in accordance to the reports of [9, 10, 11, 12]. The increase in haemoglobin as lactation progressed noted in our study was in agreement with [23]. Higher values were reported in Murrah heifers as compared to pregnant and non-pregnant buffaloes [14]. This was not in agreement with the findings in the present study. Effect of sex found in Jaffarabadi species was in agreement with observation of [13]. The decreasing haemoglobin levels in all the groups matched with the corresponding decrease in TEC as age advanced in both the species. An increase at 36 m age in the haemoglobin values in Jaffarabadi buffaloes from that noted at 24 m age also corresponded with the increase in TEC at this age in both the species. Blood Hb concentration could be used as an indicator of adaptability to the environment and the animal with higher Hb concentration have been found to be more adaptable than with lower Hb levels [24]. The changes in haematological values could also be dependent on the nutritional status of the animal. The increase in Hb as lactation advanced in Jaffarabadi buffaloes agrees with the erythrocyte count increase at these stages.

3.3 Packed cell volume (PCV)

Packed Cell Volume (%) at all ages and physiological stages in Jaffarabadi females, ranged from 38.43 ± 1.20 (12 m age) to 44.83 ± 2.59 (pregnant non-lactating). No significant difference was found over the ages studied. Non-significant reduction in the levels was observed from 1 wk to 12 m age with slight increase at 24 m age. The levels at 36 m age were non-significantly higher than that observed over the rest of the ages. The packed cell volume levels increased non-significantly from 1 m lactation to 3 m lactation in lactating Jaffarabadi buffaloes. No significant difference was seen in the values between non-lactating pregnant and non-pregnant Jaffarabadi buffaloes. In Jaffarabadi males a range from 36.88 ± 0.94 (12 m age) to 53.11 ± 3.74 (bulls) was observed across all ages and physiological stages studied. The levels from 1 wk age decreased non-significantly upto 3 m age, and then increased non-significantly at 6 m age. The value at 12 m age was significantly (P<0.05) lower than that observed at 1 wk and 6 m age. Comparison between Jaffarabadi females and males revealed no significant difference between them over the different ages studied. Significant effect of age and decreasing value of PCV as age progressed was also reported by [25, 26, 27]. Decrease in PCV levels as age increased in Jaffarabadi animals corroborated with the findings of [8, 9, 11, 12, 17]. Non-significant difference between lactation stages was in tune with the reports of [28]. Higher values noted at pregnancy over that of lactation in Jaffarabadi buffaloes was not in line with the reports of [15]. The finding by [13] of higher PCV in females than in males was not true at all the ages in the current study. The decreasing trend of PCV seen in Jaffarabadi buffaloes' upto 24 m age in females and 12 m age in males corresponded to the declining trend of TEC seen at these ages. The increase in values at 36 m in the females of both the species again was in response to the increase in TEC values at this age. The observations of non-significant increase in Jaffarabadi buffaloes as lactation advanced, higher levels in pregnant Gir and Jaffarabadi animals as compared to non-pregnant animals tallied with the similar observations of erythrocyte count at these stages. Packed cell volume is a representation of the amount of erythrocytes that are present in fixed volume of blood. Hence, the variation in the levels of PCV was in direct consonance to that of total erythrocyte count in most of the ages and stages studied.

3.4 Mean corpuscular volume (MCV), Mean corpuscular haemoglobin (MCH) and Mean corpuscular haemoglobin concentration (MCHC)

Range of erythrocyte indices in Jaffarabadi females was: MCV (fl) – 38.55 ± 0.79 (1 wk age) to 57.16 ± 1.48 (non-lactating non-pregnant); MCH (pg) – 14.80 ± 0.36 (1 m age) to 17.27 ± 0.29 (6 m age) and MCHC (%) – 35.10 ± 1.05 (3 m age) to 37.98 ± 0.18 (24 m age). The levels at 36 m were significantly (P<0.05) higher than that at 1 wk age for MCV and MCH whereas it was at par for MCHC values. No significant differences were seen at different months of lactation and among non-lactating buffaloes for MCV, MCH and MCHC values. In Jaffarabadi males the erythrocyte indices range observed was: MCV – 38.80 ± 1.26 (1 m age) to 59.15 ± 1.36 (bulls); MCH – 14.50 ± 0.56 (1 m age) to 22.28 ± 0.46 (bulls) and MCHC - 35.88 ± 0.58 (3 m age) to 37.72 ± 0.32 (bulls). Significantly (P<0.05) higher values were observed at 12 m over that of 1 wk age for MCV and MCH values, whereas no difference was observed between the two ages for MCHC values. No significant difference was observed between female and male animals of Jaffarabadi

breed for MCV, MCH and MCHC values. The values of MCV, MCH and MCHC in Jaffarabadi buffaloes in the present study were in the range calculated by [22] in Murrah buffaloes and [9] but differed from those reported by [14, 17] in Murrah buffaloes and [15] in pregnant and post-partum buffaloes. The findings of [8, 13] that older animals had significantly higher MCV and MCH values agreed with our findings. However, non-significant effect of sex on MCV, MCH and MCHC in Jaffarabadi buffaloes was contrary to the finding of [15] in free ranging buffaloes. The difference in values found with those reported by different scientists could be due to the breed difference. MCV (fl), MCH (pg) and MCHC (%) as recorded in Jaffarabadi buffaloes are a reflection of the Haemoglobin, PCV and RBC count in these species as it is only a calculated value derived from these three parameters. Hence, the changes in these values are an indication to the extent to which the Haemoglobin, PCV and RBC count varies over the period of study in Jaffarabadi buffaloes.

3.5 Total leucocyte count (TLC)

The count of total leucocytes (10^3/cmm) at all ages and physiological stages ranged from 6.74 ± 0.33 (3 m age) to 9.26 ± 0.39 (6 m age) in Jaffarabadi females. The values increased significantly (P<0.05) at 1 m from that at 1 wk and subsequently decreased significantly (P<0.05) at 3 m age. The values then increased significantly (P<0.05) at 6 m age and then dropped at 12 and 24 m age. A non-significant increase at 36 m was found over the value at 24 m age. The values at 36 m were significantly (P<0.05) higher than the values at 1 wk of age. In lactating Jaffarabadi buffaloes, TLC increased from 1 m to 2 m lactation and then dropped to 1 m levels at 3 m lactation stage. The difference was non-significant. Non-significantly higher values were detected in non-lactating pregnant buffaloes than in non-lactating non-pregnant buffaloes. Comparing the males and females of Jaffarabadi breed, significantly (P<0.05) lower value was noted at 6 m age in Jaffarabadi male than that in the females. Non-significantly lower count was observed at 1 wk and 1 m age, whereas non-significantly higher count was noted at 3 and 12 m age in Jaffarabadi males as compared to the females. In Jaffarabadi buffaloes, the range of TLC observed in our study was lower than that reported by [21, 29] in swamp buffaloes; was higher than that reported by [10] in Murrah buffaloes but was in the range of the results of [30] and [16] in Nili-Ravi buffaloes. The higher values obtained in lactating buffaloes over that of pregnant buffaloes were contrary to the findings of [16]. Non-significant effect of sex (except at 6 m age) in the current study was in agreement to the finding of [9] in Iranian buffaloes. However, [30] reported significant effect of age on TLC. Higher values in pregnant as compared to non-pregnant buffaloes was in tune with the findings of [31]. TLC in free ranging buffalo was found to be higher than that in cattle [13]. The current findings are not in complete agreement with their report as TLC values fluctuated at different ages and stages studied. The leucocyte counts in Jaffarabadi buffaloes were nearly in the same range over different ages and stages studied. The lower values observed at birth and the increase in the values as age progressed (in Jaffarabadi females and males) could be because at birth the neonate attains its immunity from the colostrum that it sucks from the dam. However, as age increases the increase in TLC (10^3/cmm) values indicate the adaptability of the young one to its environment and also its increased immune ability. The counts of leucocyte at different periods of lactation and during pregnancy were comparatively higher than found at most of the ages in Jaffarabadi buffaloes. The presence of higher leucocyte count during these stages could be due to stress. In the present study in Jaffarabadi buffaloes, the cortisol levels at lactation and in pregnancy were much higher than that observed over the rest of the ages. The higher cortisol levels in the lactating buffaloes might be responsible for higher leucocyte counts [32].

4. Conclusion

Significant (P<0.05) effect of age was found in both sexes of Jaffarabadi buffaloes for TEC and TLC. However, with regard to Hb and PCV values significant (P<0.05) effect of age was noticed only in Jaffarabadi males. Significant (P<0.05) difference was noted in the TEC during lactation, whereas non-significant effect was observed between non-lactating animals in Jaffarabadi species. However, Hb, PCV and TLC values differed non-significantly at various stages of lactation and also between the non-lactating stages.

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