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Buffalo umbilical cord blood collection and hematological comparison with peripheral blood of new born calf and its dam*

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Cord blood (CB) transplantation has generated strong interest as an alternative to bone marrow transplantation (BMT) to treat a variety of diseases (Ballen *et al.* 2001). Attempts to collect cord blood from domestic species like equines are also reported (Bartholomew *et al*. 2009). But documented reports are not available for the protocols of cord blood collection and studies on its hematological parameters in buffalo. Therefore, the present study was conducted to explore feasibility for umbilical cord blood collection and compare its hematological parameters with the peripheral blood of new born calf and adult buffalo. This basic information can lead to use umbilical cord blood for applications related to stem cells in this economically important dairy animal in Asian countries.

Buffalo umbilical cord blood (7) was collected immediately after placental expulsion. Blood of new born calf (7) and its dam (7) was collected from the jugular vein in heparinized tubes immediately after birth. The blood for 22 hematological parameters was analyzed using hematology analyzer. The blood collected from umbilical cord, adult buffalo and its new born calf represented 3 groups as group1 (G 1), group 2 (G 2) and group 3 (G 3), respectively and their hematological parameters were compared. Analysis of variance was performed and group means were compared by Duncan's Multiple Range Test (DMRT) using SAS 9.2 software.

We report first time the collection of cord blood in buffalo through umbilical cord from passed placenta. The quantity of cord blood collected from passed placenta varied between 5–6 ml in this study. After fetal expulsion animals take generally 4–6 hours to pass the entire placenta. The collection of cord blood from passed placenta is difficult in buffaloes as the umbilical cord breaks during fetal expulsion and most of the blood gets drained out and clotted. The mean values of hematological parameters with their standard deviations for blood collected from 3 groups i.e. umbilical cord, dam (adult buffalo) and its newborn calf are presented in Table 1.

The total red blood cell (RBC) count was found significantly higher in G3 than G2 with no significant difference between G1 (cord blood) and group 3. Our report of higher values of erythrocytes in G3 than G2 is also in conformation with earlier studies where they showed higher total erythrocyte count in growing heifers than adult animals (Thangraj *et al.* 1979, Sharma *et al.* 1985). No significant difference was found in total leucocyte (WBC) count of G2 and G3. But the WBC count of cord blood (G1) was significantly lower than G2 and G3 groups. NE % was found to be lower in cord blood and new born calves than adult buffaloes. Lowest NE% in calves and highest in adult animals were observed by Simon and Jacob (1961) and Khajuria and Razdan (1966). Significantly lower MCH and MCV were found in cord blood and new born calves than adult animals. Significantly lower MCH and MCV in new born calves than adult animals are similar to the results of Chandra *et al.* (2008). Platelet count (PLT) was significantly higher in new born calves than adult animals and cord blood. Higher platelet count in new born calves provides strong immunity and thus help them surviving against neonatal infections. MPV values of G2 and G3 were significantly different with G1. But no significant difference was observed in G2 and G3 for PCT and RDWc values. Taking in view statistical analysis of all the 22 parameters, this study indicates that 7 parameters, viz. WBC, LYM, MON, NEU, HGB, PLT, PCT % of calf and 15 parameters viz. WBC, LYM, MON, NEU, EOS, BAS, LY %, NE %, HCT %, MCV, MCH, PCT %, RBC, RDWc %, MO % of dam blood were significantly different from

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Table 1. Haematological parameters with their respective means and S D values

Parameters	Umbilical cord blood (G 1)	Adult blood (G 2)	Neonatal blood (G 3)
WBC (× 10^3/µl)	2.21 ^{a} ± 0.73	10.25 ^{b} ± 2.00	11.08 ^{b} ± 1.12
LYM (× 10^3/µl)	1.48 ^{a} ± 0.22	3.82 ^{b} ± 1.89	6.73 ^{c} ± 2.51
MON (× 10^3/µl)	0.04 ^{a} ± 0.02	0.28 ^{b} ± 0.09	0.17 ^{c} ± 0.11
NEU (× 10^3/µl)	0.60 ^{a} ± 0.54	5.79 ^{b} ± 0.90	4.14 ^{c} ± 1.67
EOS (× 10^3/µl)	0.04 ^{a} ± 0.03	0.24 ^{b} ± 0.18	0.09 ^{a} ± 0.04
BAS (× 10^3/µl)	0 ^{a}	0.01 ± 0.01 ^{b}	0 $^{a\,b}$
LY %	73.20 ^{a} ± 12.01	34.27 ^{b} ± 12	58.45 ^{a} ± 18.12
MO %	1.37 ^{b} ± 0.48	1.33 ^{a} ± 2.67	1.50 ^{b} ± 0.98
NE %	28.54 ^{a} ± 13.65	59.86 ^{b} ± 10.52	38.27 ^{a} ± 17.27
EO %	2.17 ^{a} ± 1.47	2.56 ^{a} ± 2.20	0.91 ^{a} ± 0.40
BA %	0 ^{a}	0.06 ± 0.10 ^{a}	0.029 ± 0.05 ^{a}
RBC (× 10^6/µl)	9.28 ^{a} ± 0.78	7.59 ^{b} ± 0.82	9.53 ^{a} ± 0.71
HGB (g/dl)	11.62 ^{a} ± 0.44	12.33 ^{ab} ± 1.24	12.77 ^{b} ± 1.00
HCT %	37.78 ^{a} ± 3.11	43.51 ^{b} ± 4.85	41.49 ^{ab} ± 4.10
MCV (fl)	43.00 ^{a} ± 1.73	57.00 ^{b} ± 4.97	43.43 ^{a} ± 1.90
MCH (pg)	13.10 ^{a} ± 0.98	16.50 ^{b} ± 1.09	13.39 ^{a} ± 0.23
MCHC (g/dl)	28.80 ^{ab} ± 3.33	28.30 ^{b} ± 1.02	30.80 ^{a} ± 1.14
RDWc %	21.70 ^{a} ± 1.17	20.27 ^{b} ± 1.26	21.10 ^{ab} ± 0.97
PLT (× 10^3/µl)	114.57 ^{a} ± 24.15	174.14^{a} ± 62.18	259.86 ^{b} ± 73.67
PCT %	0.13 ^{a} ± 0.02	0.22 ^{b} ± 0.06	0.28 ^{b} ± 0.08
MPV	12.18 ^{ab} ± 1.09	13.43 ^{a} ± 2.50	10.67 ^{b} ± 1.23
PDWc %	34.15 ^{ab} ± 7.62	28.04 ^{b} ± 13.22	41.01 ^{a} ± 1.07

Means with different superscript in a row differ significantly (P <0.05).

Abbreviations: White blood corpuscle (WBC), lymphocytes (LYM), monocytes (MON), neutrophils (NEU), eosionophils (EOS), basophils (BAS), lymphocyte (LY), monocyte (MO), neutrophils (NE), eosionophils (EO), basophils (BA), red blood cells (RBC), haemoglobin (HGB), hematocrit (HCT), mean corpuscular volume (MCV), mean corpuscular haemoglobin (MCH), mean corpuscular haemoglobin concentration (MCHC), red cell distribution width (RDWc), platelet (PLT), thrombocrit (PCT), mean platelet volume (MPV) and platelet distribution width (PDWc)**.**

cord blood. A statistical significant (P<0.05) difference between cord blood and calf blood with respect to 7 parameters provides evidence that cord blood and calf blood are not exactly same regarding their cellular content. There may be some unknown biological mechanism which creates the difference in their constituents.

Small quantity of umbilical cord blood can be collected from buffalo after calving and passing the placenta, information related to buffalo cord blood collection method and hematological parameters is not available for comparison. Cord blood parameters differed significantly from new born calf and adult buffalo blood. This difference in hematological parameters of cord blood may be helpful in immunomodulation so that it can be used in transplantation and representing the neonatal defense system against the earliest encountered pathogens. The results may be helpful in cord blood stem cell research in buffaloes and further therapeutic applications in autologus and xenotransplantation studies in livestock species.

SUMMARY

The present study was carried out to explore the feasibility of umbilical cord blood collection and to compare its hematology with peripheral blood of new born calf and the adult buffalo. Jugular blood samples (7) were collected from new born calf and its dam and umbilical cord blood from passed placenta immediately after placental expulsion. These blood samples were analyzed for 22 hematological parameters. Statistical analysis of 22 hematological parameters from all the 3 sources indicated that 7 parameters, viz. WBC, LYM, MON, NEU, HGB, PLT, PCT % of calf and 12 parameters, viz. WBC, LYM, MON, NEU, EOS, BAS, LY %, NE %, HCT %, MCV, MCH, PCT % of adult blood, respectively, were significantly higher than the cord blood, and 3 parameters, viz. RBC, RDWc %, and MO % of adult blood were significantly lower than that of cord blood. A significant difference between cord blood and calf blood with respect to 7 parameters, viz. WBC, LYM, MON, NEU, HGB, PLT, PCT % proves that cord blood and calf blood are not exactly same regarding their cellular content. This difference in hematological parameters of cord blood may be helpful in immunomodulation so that it can be used in transplantation and represent the neonatal defense system against the earliest encountered pathogens.

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