International Journal of Agriculture Innovations and Research
Volume 4, Issue 1, ISSN (Online) 2319-1473
Manuscript Processing Details (dd/mm/yyyy) :
Received : 14/07/2015 | Accepted on : 21/07/2015 | Published : 06/08/2015

A Study Whether Blood Serum Mineral Levels in Ruminants Serve as an Indicator of Their Dietary Status

B.M. Bhanderi, M.R. Garg, P.L. Sherasia

***Abstract* – A study was conducted in 10 male crossbred calves (HF x Jersey) to examine the mineral levels in blood serum during different time interval of the day, maintained under similar dietary and management conditions. Animals were maintained on normal feeding regime for 120 days, following which their blood samples were collected at 8:30, 10:30, 12:30, 14:30, 16:30, 18:30 and 20:30 hours of the day, for estimation of various macro and micro-minerals in blood serum. It was observed that the levels of various macro and micro-minerals in blood serum kept fluctuating throughout the day. The peak levels of calcium (Ca), phosphorus (P), sulphur (S) and potassium (K) were observed between 8:30 and 10:30 hours of the day, whereas, magnesium (Mg) and sodium (Na) attained peak levels after 18:30 hour. Similarly, copper (Cu), zinc (Zn) attained peak levels between 10:30 and 12:30 hours of the day. Peak levels in blood serum for manganese (Mn), iron (Fe), cobalt (Co) and selenium (Se) were found to be highly variable from animal to animal at different hours of the day. By taking blood serum sample at random during a particular time of the day, while the levels of only certain minerals may be seen attaining peak levels, others may be at a much lower concentration than the critical levels. Based on such observations, it is misleading to judge the dietary status of minerals, as most of minerals are tightly regulated in the animal system through homeostatic mechanism. The results of the study indicated that levels of macro and micro-minerals in blood serum of ruminants cannot be considered as an indicator to reflect their dietary status.**

***Keywords* – Blood Serum, Minerals, Dietary Status, Crossbred Calves.**

I. Introduction

In dairy animals, minerals play an important role in growth, milk production and reproduction efficiency [1]. Increased demand for minerals during pregnancy, birth or other physiologically demanding events cannot be met by the animal's ration; the skeletal system serves as an important source for the serum mineral demand, in order to maintain its normal concentration [2-3]. Because of this homeostatic regulatory system, levels of minerals in blood serum are not considered as a good indicator for assessing the dietary minerals status of dairy animals [4]. Several agencies assess dietary mineral status of ruminant animals based on their levels in blood serum, which could be misleading. To demonstrate this, a study was undertaken in crossbred male calves maintained on normal ration under farm conditions. Macro and micro-minerals in blood serum were tested at different intervals and the results are reported in this communication.

II. Materials and Methods

A study was undertaken in ten crossbred (HF x Jersey) male calves (2-2.5 years age) maintained under farm conditions. Calves were fed on green fodder, dry fodder, concentrate mixture and mineral mixture to meet their protein, energy and minerals requirement, as per the NRC [5]. The feed was offered twice daily at 8:45 and 15:45 hrs, after discarding residue, if any. All the calves had free access to clean drinking water throughout a day. Animals were maintained on this feeding regime for 120 days, following which their blood samples were collected at 8:30, 10:30, 12:30, 14:30, 16:30, 18:30 and 20:30 hours interval of the day, in a vacuette tube with serum clot activator from the jugular vein for estimation of macro and micro-minerals. For determination of macro and micro-minerals, serum samples were diluted to 1:5 with 0.1% (w/v) lanthanum chloride solution. Serum calcium (Ca), phosphorus (P), magnesium (Mg), sodium (Na), potassium (K), copper (Cu), zinc (Zn), manganese (Mn), iron (Fe), cobalt (Co) and selenium (Se) were estimated using Inductively Coupled Plasma-Optical Emission Spectroscopy (Model Optima 3300 RL). The concentrations of Ca, P, Mg, Na, K and S were expressed as mg/dl, whereas, concentrations of Cu, Zn, Mn, Fe, Co and Se were expressed in µg/ml.

III. Results and Discussion

3.1 Macro-Minerals Profile in Blood Serum

Levels of macro-minerals (mg/dl) in blood serum of male calves at different time interval of the day are presented in Table I. It was observed that the levels of various macro-minerals in blood serum kept fluctuating throughout the day. The peak levels of Ca, P, S and K were observed between 8:30 and 10:30 hours, whereas, Mg and Na attained peak levels after 18:30 hour.

The serum Ca level varied from 6.30 to 13.16 mg/dl at different time interval of the day (Figure 1). The peak level of Ca was attained at 10:30 and 12:30 hours in 80 and 20% animals, respectively. The fluctuation in mineral level in blood serum of animals may be due to fluctuation in absorption from intestinal lumen and mobilization from bones. In the present study, Ca level in the blood serum below the critical level (<8.0 mg/dl) was observed in many animals, after attaining peak levels. Collection blood samples at random during the day time for assessment of dietary status may not always reflect the true Ca status in ruminant animals. Assessment of Ca status requires analysis of bone since approximately 98 to 99% of the body Ca content is in bone and serum concentrations are maintained by both diet and turnover of bone matrix [6]. Blood concentrations of macro-minerals are not reflective of dietary status when the homeostatic system is functioning properly. Assessment of Ca concentrations around the time of calving is a useful indicator of how well the Ca regulatory system is working and potential for

clinical or subclinical hypo-calcemia problems. Other than the 2 weeks prior to and following calving, blood Ca is not a very diagnostic value as a result of the intact regulatory system.

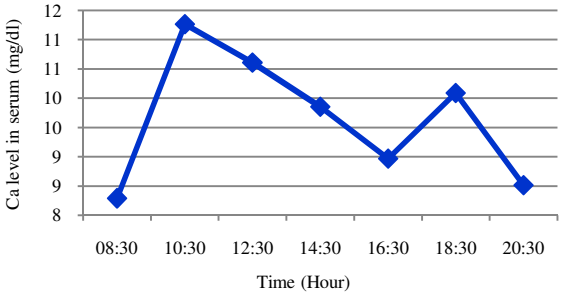

Fig.1. Level of Ca (mg/dl) in blood serum at different time interval of the day

The serum P level varied from 5.83 to 13.28 mg/dl at different time interval of the day (Figure 2). The peak level of P was attained at 10:30 and 16:30 hours in 60 and 30% animals, respectively. Phosphorus status is somewhat difficult to measure in animal tissues. Serum and urine P concentrations can aid in assessing dietary deficiency, but with mobilization of bone P to maintain serum concentration, significant drops in serum and urine may take several weeks to develop. Dietary P and/or response to supplementation are better indicators of deficiency than tissue concentrations unless severe long-term deficiency has occurred.

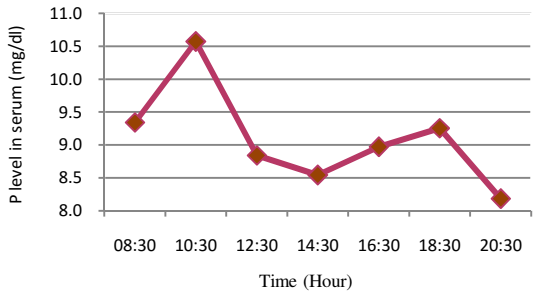

Fig.2. level of P (mg/dl) in blood serum at different time interval of the day

The serum Mg level varied from 1.81 to 3.37 mg/dl at different time interval of the day (Figure 3). The peak level of Mg was attained at 18:30 hour in 80% animals (Table I). Assessment of true Mg deficiency would require analysis of bone since approximately 70% of the body magnesium content is in bone.

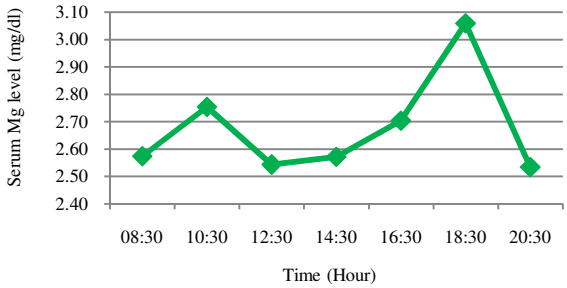

Fig.3. Level of Mg (mg/dl) in blood serum at different time interval of the day

The serum S level varied from 60.28 to 97.95 mg/dl at different time interval of the day (Figure 4). The peak level of S was observed at 8:30 hour in 70% animals. The onward reduction in the blood serum S level may be due to the utilization by rumen microbes for synthesis of S containing amino acids.

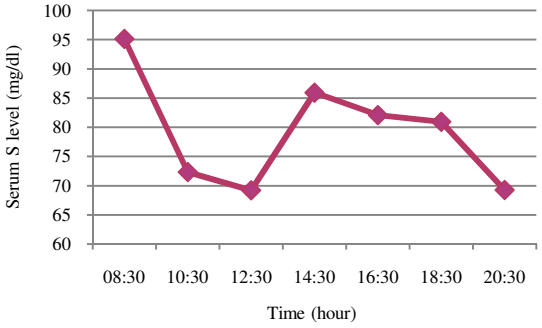

Fig.4. Level of S (mg/dl) in blood serum at different time interval of the day

Table I: Levels of macro-minerals (mg/dl) in blood serum at different time interval

Element	Time (hour)	A1	A2	A3	A4	A5	A6	A7	A8	A9	A10
Ca (*CL: <8.0)	8:30	8.41	8.99	8.77	9.86	6.30	8.24	8.15	8.00	8.94	7.19
	10:30	11.16	9.43	12.02	12.05	11.31	10.79	11.85	11.27	10.95	11.84
	12:30	9.03	13.16	11.82	11.90	10.51	10.86	10.93	9.93	8.79	9.18
	14:30	7.58	10.39	11.01	10.52	9.08	10.21	10.06	10.62	9.26	9.81
	16:30	8.05	7.60	7.98	8.24	8.28	10.49	10.92	8.64	8.98	10.50
	18:30	10.11	8.96	9.62	10.38	9.25	8.84	11.25	10.69	10.72	11.07
	20:30	9.17	8.51	8.46	8.24	6.76	8.80	8.36	10.04	7.46	9.32
P (*CL: <4.5)	8:30	8.56	9.48	7.35	10.05	9.65	9.62	8.85	10.21	10.14	9.50
	10:30	10.24	13.28	8.86	9.66	11.07	10.73	10.60	11.56	9.77	9.94
	12:30	9.90	10.72	7.27	8.80	9.80	8.76	9.89	9.20	8.23	5.83
	14:30	6.52	10.64	7.26	8.75	8.93	7.93	8.98	9.00	8.55	8.85
	16:30	8.75	7.57	6.67	7.54	8.17	9.61	11.42	8.55	10.87	10.54

	18:30	9.41	10.22	7.67	9.15	8.43	8.83	9.08	10.87	8.81	10.06
	20:30	8.70	8.67	6.50	7.90	7.76	8.09	8.18	9.59	7.86	8.56
Mg (*CL: <1.20)	8:30	2.17	3.09	2.22	2.87	2.94	2.30	2.22	2.88	2.53	2.52
	10:30	2.54	3.14	2.59	2.81	3.37	2.69	2.54	2.98	2.27	2.61
	12:30	2.60	3.08	2.12	2.74	3.28	2.45	2.44	2.79	2.02	1.91
	14:30	1.81	3.26	2.51	2.72	2.94	2.35	2.39	3.02	2.19	2.53
	16:30	2.31	2.74	2.30	2.52	3.06	2.82	2.96	2.74	2.59	3.00
	18:30	2.95	3.34	2.75	3.13	3.29	2.49	3.10	3.63	2.87	3.04
	20:30	2.73	3.02	2.26	2.51	2.54	2.47	2.65	2.61	2.09	2.46
S (**CL: <75)	8:30	90.84	95.94	92.46	96.23	94.08	91.79	84.66	92.82	99.41	113.14
	10:30	71.49	82.21	74.75	65.91	73.45	73.13	70.23	70.74	61.40	79.80
	12:30	73.23	69.07	60.28	69.52	71.10	68.43	73.74	68.04	73.90	64.73
	14:30	66.86	97.95	89.08	84.47	83.40	83.22	86.72	96.04	76.70	94.63
	16:30	90.59	77.84	85.26	82.16	92.37	78.50	85.33	67.98	72.97	87.59
	18:30	83.60	78.58	75.62	73.81	77.58	72.88	90.74	90.52	77.37	88.68
	20:30	77.08	69.98	64.38	61.48	66.70	69.38	67.08	78.81	60.48	77.07
K (**CL: <25)	8:30	25.98	22.53	26.48	31.11	28.56	30.59	32.75	28.67	38.54	33.49
	10:30	27.89	30.04	27.66	28.65	25.66	29.50	32.67	27.70	28.22	26.16
	12:30	26.84	24.74	23.66	25.81	27.40	23.96	26.81	24.30	23.67	15.15
	14:30	17.65	25.67	26.60	25.25	23.63	22.28	24.67	21.35	24.65	24.47
	16:30	22.37	18.84	21.26	20.32	22.90	25.79	31.19	19.11	31.68	28.99
	18:30	26.76	23.30	26.80	24.92	21.17	22.70	32.28	25.68	31.80	26.60
	20:30	23.33	20.13	21.84	21.47	20.96	21.53	19.09	26.21	24.57	22.29
Na (**CL:<300)	8:30	309.63	297.50	299.75	333.38	295.58	330.59	339.12	322.53	392.61	381.25
	10:30	329.56	340.32	327.11	319.77	314.48	325.89	330.41	315.25	310.77	305.15
	12:30	310.09	282.13	256.37	320.01	310.22	302.75	312.70	293.51	270.41	208.11
	14:30	227.03	315.21	320.40	310.30	294.71	281.65	310.15	334.85	285.62	303.48
	16:30	307.57	266.09	289.30	289.29	306.51	372.46	391.19	321.28	352.46	363.08
	18:30	352.04	339.40	343.61	350.68	350.79	318.96	372.92	427.51	366.40	362.47
	20:30	333.26	299.79	292.64	279.86	266.98	288.65	283.69	349.96	295.55	302.74
CL=Critical levels,*[7],** [8]											

The serum K level varied from 15.15 to 38.54 mg/dl at different time interval of the day (Figure 5). The peak level of K was attained at 10:30 and 12:30 hours in 60 and 30% animals, respectively. Tissue concentrations of Na and K poorly correlate with dietary deficiency.

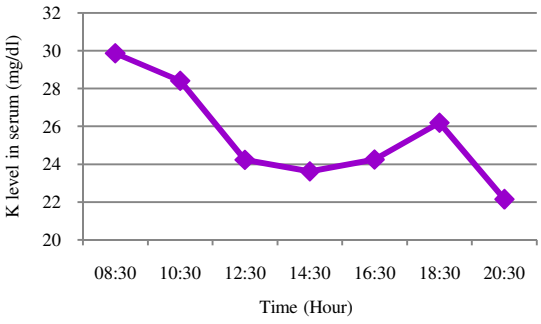

Fig.5. Level of K (mg/dl) in blood serum at different time interval of the day

The serum Na level varied from 207.11 to 427.51 mg/dl at different time interval of the day (Figure 6). Of the animal samples available, serum and urine are the best for measuring Na and K deficiency, but disease states can cause electrolyte shifts that result in lowered serum or urinary Na and K even when dietary concentrations are adequate. In addition, serum that is haemolyzed or left on the clot too long may have falsely increased K concentration due to loss from the red blood cells. Thus, dietary Na and K concentrations are a better guide to assess mineral status of dairy animals.

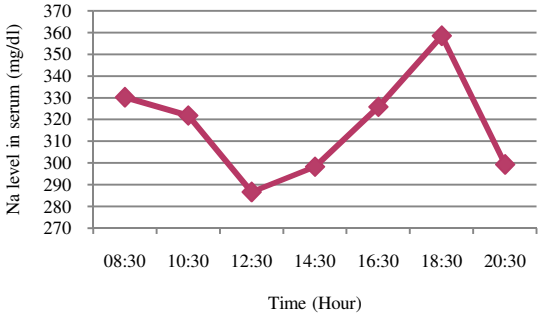

Fig.6. Level of Na (mg/dl) in blood serum at different time interval of the day

3.2 Micro-Minerals Profile in Blood Serum

Levels of micro-minerals (µg/ml) in blood serum of male calves at different time interval are presented in Table II. The serum Cu level in animal to animal varied from 0.42 to 0.97µg/ml at different time interval of the day (Figure 7). Peak level of Cu was attained between 10:30 and 16:30 hours of the day. The Cu level in blood serum kept fluctuating throughout the day. In view of this, the best method for assessing Cu status is via analysis of liver tissue, although much testing is performed on serum. Serum concentration does not fall until liver Cu is significantly depleted.

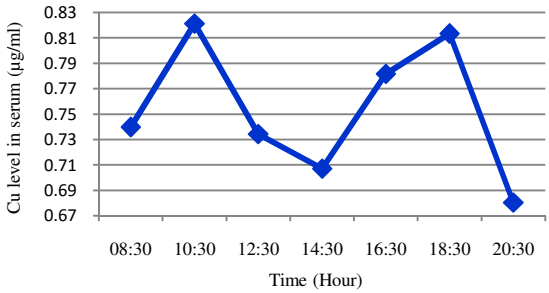

Fig.7. Level of Cu (µg/ml) in blood serum at different time interval of the day

The serum Zn level varied from 0.49 to2.16 µg/ml at different time interval of the day. The peak level of Zn was attained at 10:30 and 12:30 hours in 50 and 30% animals, respectively (Figure 8). Tissue and serum Zn concentrations do not reflect body status well [9]. It has been suggested that pancreas Zn concentration is the best means of truly assessing Zn status.

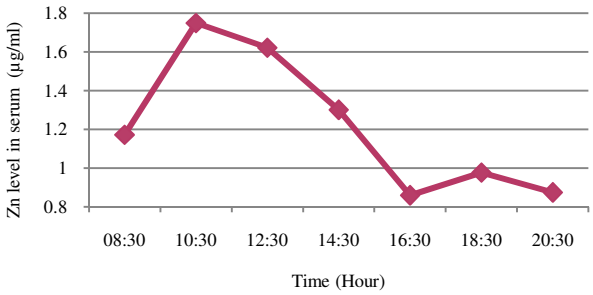

Fig.8. Level of Zn (µg/ml) in blood serum at different time interval of the day

Peak levels in blood serum for Mn, Fe, Co and Se were found to be highly variable from animal to animal at different time interval (Figures 9-12). Of the samples available, liver is the most indicative of whole body status of Mn, followed by whole blood, and then serum. As red blood cells have higher Mn concentration than serum, haemolysis can result in increased serum concentration.

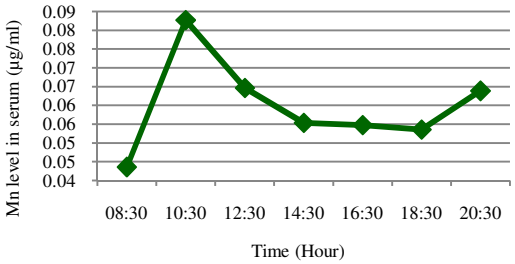

Fig.9. Level of Mn (µg/ml) in blood serum at different time interval of the day

Table II: Levels of micro-minerals (µg/ml) in blood serum at different time interval

Element	Time (hour)	A1	A2	A3	A4	A5	A6	A7	A8	A9	A10
Cu (*CL: <0.65)	8:30	0.73	0.59	0.75	0.78	0.77	0.77	0.68	0.67	0.93	0.75
	10:30	0.86	0.72	0.93	0.80	0.81	0.96	0.83	0.74	0.84	0.73
	12:30	0.91	0.68	0.73	0.81	0.73	0.80	0.83	0.73	0.70	0.42
	14:30	0.60	0.65	0.82	0.75	0.65	0.74	0.74	0.74	0.71	0.66
	16:30	0.86	0.50	0.77	0.74	0.71	0.88	0.89	0.71	0.97	0.79
	18:30	0.92	0.67	0.75	0.87	0.73	0.72	0.84	0.96	0.97	0.70
	20:30	0.78	0.57	0.62	0.63	0.68	0.69	0.66	0.71	0.88	0.59
Zn (*CL:<0.80)	8:30	0.95	1.10	1.50	1.95	1.12	1.06	1.02	0.85	0.94	1.23
	10:30	1.54	1.46	1.92	2.01	1.25	1.58	1.73	1.70	2.16	2.13
	12:30	1.32	2.14	2.14	1.80	1.03	1.91	1.75	1.11	1.37	1.64
	14:30	0.95	1.05	1.42	1.23	1.27	1.16	1.38	0.85	1.73	1.97
	16:30	0.83	0.75	0.84	0.74	0.87	0.78	1.27	0.53	0.99	0.99
	18:30	0.95	0.96	0.89	1.10	0.75	0.80	1.31	0.59	1.30	1.12
	20:30	0.84	0.75	0.76	1.37	0.66	0.94	0.49	1.23	0.76	0.93
Mn (*CL: <0.02)	8:30	0.031	0.113	0.025	0.013	0.066	0.053	0.028	0.047	0.020	0.039
	10:30	0.066	0.089	0.100	0.075	0.131	0.088	0.053	0.058	0.095	0.072
	12:30	0.065	0.095	0.083	0.054	0.069	0.061	0.068	0.055	0.037	0.059
	14:30	0.055	0.050	0.059	0.101	0.039	0.041	0.062	0.049	0.049	0.048
	16:30	0.053	0.026	0.043	0.057	0.041	0.049	0.055	0.039	0.129	0.055

	18:30	0.041	0.045	0.037	0.061	0.026	0.053	0.077	0.040	0.082	0.073
	20:30	0.071	0.036	0.035	0.046	0.199	0.038	0.046	0.039	0.072	0.056
Fe (*CL: <1.10)	8:30	2.30	1.46	1.58	1.88	3.91	2.40	3.33	2.77	2.62	2.60
	10:30	1.57	2.75	2.86	2.64	2.45	2.91	3.36	3.11	1.98	2.99
	12:30	2.12	2.76	3.20	2.23	2.62	2.38	3.00	2.33	1.60	1.80
	14:30	3.10	3.06	2.62	3.98	2.14	2.48	2.88	2.80	1.67	2.20
	16:30	1.16	1.40	1.45	1.30	1.79	1.94	2.48	1.65	1.56	1.98
	18:30	1.26	1.61	1.86	1.51	1.45	1.67	1.66	2.01	1.54	1.98
	20:30	1.08	1.15	1.27	1.51	1.42	2.08	1.46	1.71	1.26	1.96
Co (*CL: <0.007)	8:30	0.007	0.004	0.002	0.002	0.010	0.004	0.004	0.003	0.007	0.007
	10:30	0.008	0.012	0.016	0.011	0.046	0.007	0.009	0.008	0.009	0.007
	12:30	0.015	0.026	0.010	0.006	0.004	0.006	0.006	0.004	0.003	0.004
	14:30	0.005	0.002	0.005	0.014	0.011	0.006	0.032	0.006	0.012	0.012
	16:30	0.008	0.011	0.008	0.006	0.007	0.006	0.010	0.004	0.006	0.005
	18:30	0.005	0.005	0.005	0.010	0.002	0.003	0.003	0.006	0.008	0.011
	20:30	0.005	0.006	0.006	0.005	0.004	0.002	0.003	0.002	0.004	0.005
Se (*CL: <0.03)	8:30	0.088	0.091	0.081	0.067	0.074	0.055	0.091	0.062	0.063	0.039
	10:30	0.108	0.113	0.103	0.077	0.107	0.070	0.048	0.034	0.042	0.049
	12:30	0.095	0.045	0.094	0.086	0.104	0.092	0.087	0.109	0.079	0.072
	14:30	0.081	0.110	0.094	0.102	0.086	0.103	0.100	0.094	0.086	0.095
	16:30	0.082	0.076	0.084	0.087	0.073	0.077	0.089	0.069	0.062	0.088
	18:30	0.076	0.053	0.070	0.064	0.065	0.079	0.074	0.082	0.070	0.080
	20:30	0.068	0.075	0.065	0.085	0.090	0.098	0.067	0.062	0.097	0.070
CL=Critical levels,*[7].											

The normal serum concentration of Mn is quite low. This could probably due to high turnover of Mn in blood serum. Both liver and serum concentrations are commonly utilized to assess Fe deficiency. When using serum to measure Fe concentration, samples that have evidence of haemolysis should not be used, as they will have artificially increased Fe concentration from the ruptured red blood cells.

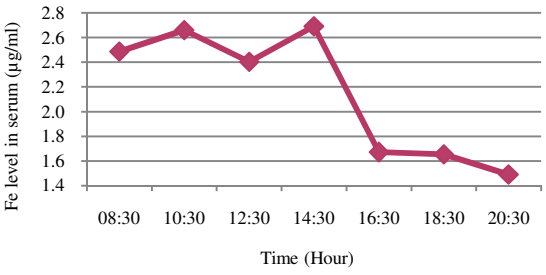

Fig.10. Level of Fe (µg/ml) in blood serum at different time interval of the day

Tissue and serum concentrations of Co are generally quite small, as the vitamin B_{12} is produced in the rumen by the microflora. Since Co concentrations may not truly reflect the vitamin B_{12} concentrations, the most appropriate analysis for Co deficiency is the direct quantification of liver vitamin B_{12}. Liver concentrations of Cu, Mn, Zn and Se provide the best indication of trace mineral status in ruminant animals [1].

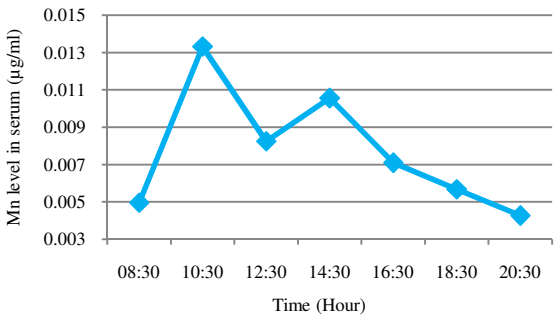

Fig.11. Level of Co (µg/ml) in blood serum at different time interval of the day

Appropriate assessment of mineral status involves thorough evaluation of groups of animals. The evaluation should include a thorough health history, feeding history, supplementation history and analysis of several animals for their mineral status. If minerals are deemed to be adequate in the diet but the animals are found to be deficient, antagonistic interactive effects of other minerals need to be investigated. A variety of factors such as exercise, stress, excitation during blood sampling, hour of the day, feeding time, season, animal's age and level of vitamin supplementation have an influence on the serum mineral levels [10-11].

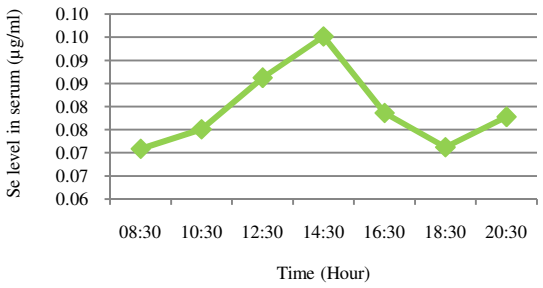

Fig.12. Level of Se (µg/ml) in blood serum at defferent time interval of the day

Serum mineral levels are frequently used in assessment mineral status of dairy animals, which could be misleading. Homeostatic control mechanisms can limit changes in mineral concentrations in blood serum until endogenous reserves are substantially depleted [12].

IV. Conclusion

By taking blood serum sample at random during a particular time of the day, while the levels of only certain minerals may be seen attaining peak levels, others may be at a much lower concentration. Based on such observations, it is misleading to judge the dietary status of minerals, as most of minerals are tightly regulated in the animal system through homeostatic mechanism. Considering total dietary mineral intake from feed resources and minerals requirement for different productive and reproductive functions is the best option for assessing dietary mineral status in ruminant animals.

Acknowledgement

Financial assistance and necessary facilities provided by the management of National Dairy Development Board, Anand, for undertaking this study, are gratefully acknowledged.

References

[1] McDowell, L.R. 1992. *Minerals in Animal and Human Nutrition*. Academic Press, San Diego, CA, pp. 49-51.

[2] Shupe, J.L., Butcher, J.E., Call, J.W., Olson, A.E. and Blake, J.T. 1988. Clinical signs and bone changes associated with phosphorus deficiency in beef cattle. *American Journal of Veterinary Research,* 49:1629-1636.

[3] Mosel, M.V., Wouterse, H.S. and Vant Klooster, A.T.H. 1994. Effects of reducing dietary (Na + K)-(Cl + SO_4) on bone mineral levels in dairy cows at parturition. *Research in Veterinary Science,* 56:270-276.

[4] Hall, J.O. 2006. Appropriate methods of diagnosing mineral deficiencies in cattle. Tri-State Dairy Nutrition Conference, pp. 43-50.

[5] N.R.C. 2001. *Nutrient Requirements of Dairy Cattle*, 7th Edn., National Academy of Sciences – National Research Council, Washington, DC. ISBN-13:978-0-309-06997-7.

[6] Underwood, E.J. and Suttle, N.F. 1999. *The Mineral Nutrition of Livestock*. 3rd ed. CAB International Publishing Co.

[7] McDowell, L.R. 1985. *Nutrition of Grazing Ruminants in Warm Climates*. Academic Press, New York.

[8] Radostits, O.M., Gay, C.C., Hinchcliff, K.W. and Constable, P.D. 2007. *Veterinary Medicine*. 10th edition, Philadelphia: Saunders Elsevier, pp. 795–798.

[9] Mills, C.F. 1987. Biochemical and physiological indicators of mineral status in animals: copper, cobalt and zinc. *Journal of Animal Sciences,* 65:1702-1711.

[10] Beighle, D.E., Boyazoglu, P.A., Hemken, R.W. and Serumaga-Zake, P.A. 1994. Determination of calcium, phosphorus and magnesium values in rib bones from clinically normal cattle. *American Journal of Veterinary Research,* 55:85-89.

[11] Gonul, R., Kayar, A., Bilal, T., Erman, O.R.M., Cagla Parkan, D.V.M., Dodurka, H., Gulyasar, T. and Barutcu, B. 2009. Comparison of mineral levels in bone and blood serum of cattle in Northwestern Turkey. *Journal of Animal and Veterinary Advances,* 8(7):1263-1267.

[12] Miller, W.J. 1975. New concepts and developments in metabolism and homeostasis of inorganic elements in dairy cattle: A review. *Journal of Dairy Science,* 58:1549–1560.

Author's Profile

Dr. Babulal M Bhanderi
PhD. Animal Nutrition
Scientist-II
Animal Nutrition Division
National Dairy Development Board, Anand, Gujarat-388001
Email ID: bhanderi@nddb.coop

Dr. Mangat Ram Garg
PhD. Animal Nutrition
Principal Scientist
Animal Nutrition Division
National Dairy Development Board, Anand, Gujarat-388001
Email ID: mrgarg@nddb.coop

Dr. Pankaj S Sherasia
MVSc. Animal Nutrition
Scientist-II
Animal Nutrition Division
National Dairy Development Board, Anand, Gujarat-388001
Email ID: pankajs@nddb.coop