

EPIDEMIOLOGY, CLINICO-BIOCHEMISTRY AND EVALUATION OF
TREATMENT MODALITIES FOR THE MANAGEMENT
OF OSTEOMALACIA IN BUFFALOES (*BUBALUS BUBALIS*)

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ABSTRACT

The present study was planned for investigations on epidemiology, clinico-biochemical and radiological aspects and therapeutic management of osteomalacia in buffaloes. Overall hospital incidence of osteomalacia in buffaloes was 8.31% with higher incidence during mid lactation in high yielding buffaloes exclusively fed on dry fodder. The characteristic clinical signs observed were progressive loss of body weight, shifting lameness, stiff gait, arching of back as well as difficulty in lifting chest portion and keeping on knee joint for longer time while getting up. Significant decrease in hemoglobin, total erythrocyte count and haematocrit while increase in MCV and granulocyte count was observed in affected buffaloes as compared to healthy buffaloes. Highly significant decrease in mean plasma inorganic phosphorus, significant decrease in calcium and highly significant increase in alkaline phosphatase levels was observed in osteomalacia affected buffaloes. Radiographic examination revealed proliferation of osteophytes in carpal bones, demineralization of canon bones

and osteolytic changes indicated by decreased radiographic density in last few coccygeal vertebrae. For evaluating the efficacy of different treatment modalities in osteomalacia, ailing buffaloes were divided in to three groups (n=10). Group I, II and III buffaloes were treated with mineral mixture, buffered phosphorus and buffered phosphorus with mineral mixture respectively. The overall recovery period was significantly lower (27.1 ± 4.05 days) in Group III compared to Group II (39.2 ± 4.2 days) and Group I (40.5 ± 3.72 days) buffaloes. On the basis of recovery period and restoration of biochemical parameters combined treatment with parenteral buffered phosphorus preparation and mineral mixture proved most effective in management of osteomalacia in buffaloes.

Keywords: *Bubalus bubalis*, buffalo, osteomalacia, buffered phosphorus, mineral mixture, treatment

INTRODUCTION

Osteomalacia is a disease of adult animals named from one of its characteristic symptom-

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softening and replacement of bone with osteoid tissue which resembles uncalcified bone (Radostits *et al.*, 2010). The disease is common in pregnant or lactating cows due to consistently lower plasma inorganic phosphorus concentration during their peak lactation period, particularly on phosphorus deficient range or during periods of drought (Madsen, 1942). Extreme phosphorus deficiency in ruminants leading to clinical bone disorders is relatively rare under practical conditions while moderate to marginal deficiency is undoubtedly very wide spread which ultimately predisposes animals to diseases (Radostits *et al.*, 2010). Phosphorus deficiency occurs in animals fed on forages or pastures grown on phosphorus deficient soils. This deficiency may also develop in animals fed exclusively on dry roughages, which are naturally poor in phosphorus even when grown upon soil with adequate phosphate content (Smith, 1990). Phosphorus content of pasture recedes as the feed dries off (Rose, 1954).

Different workers have used bone flour, sodium acid phosphate, mineral mixture and organic phosphorus preparations in phosphorus deficiency syndrome in cattle and rheumatism like syndrome in buffaloes with variable results (Kalra *et al.*, 1979; Abdou *et al.*, 1986; Verma 1995; Kishan and Sridhar, 2000; Bhikane *et al.*, 2003; Singh *et al.*, 2012). However, no systematic work has been done on therapeutic aspects of osteomalacia in buffaloes with special reference to use of parenteral buffered phosphorus preparation. Hence, the present investigation was undertaken to study epidemiology, clinical, haemato-biochemical and radiological aspects of osteomalacia in buffaloes and to evaluate therapeutic efficacy of various treatment protocols in management of osteomalacia in buffaloes.

MATERIALS AND METHODS

Study area and selection of animals

The present study was carried in buffaloes admitted to the Teaching Veterinary Clinical Complex, College of Veterinary and Animal Sciences, Udgir, from Latur and Nanded districts of Marathwada region and Bidar district of Karnataka, during June 2015 to May 2016. All the ailing buffaloes admitted to clinics were screened for osteomalacia. The buffaloes showing clinical signs of emaciation, abducted elbows, arched back, difficulty while getting up (kneeling on knee joint) and cautious to stiff gait and having history of good/ high milk yield with exclusive feeding on dry roughages were suspected for osteomalacia. The careful auscultation of heart, lungs, and area of rumeno-reticulum was done to rule out the possibility of pericarditis, pleuro - pneumonia and traumatic reticulitis. The information pertaining to age, stage of lactation, milk yield and feeding status was recorded in all suspected cases of osteomalacia for computing hospital incidence and were subjected to haemato-biochemical investigations.

Complete Blood Count (CBC)

Freshly collected 5 ml blood in EDTA vials by jugular venipuncture from suspected buffaloes was analysed for CBC on automated haematology analyser (Model: Abacus Junior Vet, Diatron GMBH, Austria).

Biochemical analysis

Freshly collected heparinized blood samples (5 ml) by jugular venipuncture from suspected buffaloes were subjected to centrifugation at 3000 rpm for 5 minutes to harvest plasma. Collected plasma samples were analysed

for calcium, inorganic phosphorus, alkaline phosphatase and glucose on semi-automated spectrophotometer (Model: Chemistry Analyzer-CA 2005 B4B Diagnostic division, China, Model no. CA 2005).

Rumen fluid

Rumen fluid from suspected buffaloes was analysed for physical characteristics, pH, rumen protozoa motility and density.

Radiographic examination

Thirty buffaloes were subjected for radiographic examination of reticulothorax, tail bones and long bones of fore leg by using 1000 mA, 150 kVp computed radiographic system (Siemens polydrop LX - 1).

Soil and fodder analysis

Nineteen random soil samples were collected from study area as per method described by Jackson (1973) and subjected to estimation of soil calcium (Piper, 1966) and available phosphate (Olsen *et al.*, 1954). Similarly 39 feed and fodder samples randomly collected from study area were processed for estimation of calcium (Talpatra *et al.*, 1940) and phosphorus (Pathak *et al.*, 1996).

Diagnosis

The confirmative diagnosis of osteomalacia was made based on decreased radiodensity of bones, absence of foreign body syndrome on radiology and decreased levels of plasma inorganic phosphorus, calcium and increased alkaline phosphatase.

Therapeutic trial

Thirty buffaloes with typical clinical signs of osteomalacia and positive for hypophosphatemia

were randomly divided in to three treatment groups (n=10). Group I buffaloes were treated with mineral mixture 50 gm PO BID daily till clinical recovery. Group II buffaloes were treated with buffered phosphorous injection 50 ml/day IV for first two days followed by 25 ml/day IV till clinical recovery. Group III buffaloes were treated with buffered phosphorus injection 50 ml IV/day for 3 days followed by mineral mixture 50 gm BID orally till clinical recovery. Supportive treatment comprised of Dextrose 20% 1 lit IV and vitamin B- complex 10 ml IM for three days.

Clinical, haematological and biochemical parameters of study animals in all three groups were evaluated after clinical recovery. Efficacy of best treatment protocol was evaluated based on clinical recovery period and improvement in biochemical parameters *viz.*, plasma inorganic phosphorus and alkaline phosphatase.

Composition/1.2 kg: Calcium -225 g, Phosphorus -127.5 g, Magnesium -6.0 g, Manganese -1.5 g, Iron -1.5 g, Iodine -325 mg, Copper -4.2 g, Zinc -9.6 g, Cobalt -150 mg, Sulphur -7.2 g, Potassium -100 mg, Sodium -6 mg, Selenium -10 mg, Vitamin A -700000 IU, Vitamin D₃ -70000 IU, Vitamin E -250 mg, Nicotinamide -1000 mg and Chromium -78 mg.

Buffered phosphorus preparation: (Vial A of 30 ml- Sodium dihydrogen phosphate 30 mg/ml and Disodium hydrogen phosphate dodecahydrate -230 mg/ml, Vial B of 20 ml- Inosine -12 mg/ml, Sodium pyruvate -12 mg/ml).

Statistical analysis

Data generated from different treatment groups with reference to haematology, biochemistry and recovery period was subjected to Analysis of Variance (ANOVA) while within group data was analyzed by paired 't' test using SPSS software

(Version: 20).

RESULTS AND DISCUSSION

Incidence of osteomalacia

Out of 758 clinical cases of buffaloes were screened, 63 buffaloes were found clinically positive for osteomalacia indicating an overall hospital incidence of 8.31%. The very high incidence observed could be attributed to severe drought during the study period in the area under study.

Age-wise distribution

The age-wise distribution of osteomalacia in buffalo showed highest occurrence in 6 to 9 year old buffaloes (50.80%) followed by 3 to 6 year (34.92) and >9 year (14.28%). The results of present study are in agreement with Verma (1995) who reported the rheumatism like syndrome with more incidence in 5 to 11 year age group buffaloes. Singh *et al.* (2012) reported higher incidence of hypophosphatemia in mature buffaloes >6 years compared with <3 and 3 to 6 years of age. The higher susceptibility of adult buffaloes to osteomalacia may be attributed to peak production period and decreased absorptive efficiency of phosphorus and bone metabolism with increasing age (NRC, 2001).

Sex-wise distribution

Osteomalacia, being associated with high milk production was observed only in female buffaloes (100%) which is in agreement with findings of Bhikane *et al.* (2003); Verma (1995); Habib *et al.* (2004).

Season-wise distribution

Season-wise highest occurrence of

osteomalacia in buffaloes was recorded during monsoon (53.96%) followed by summer (39.68%) and least in winter (6.36%). No case was recorded during post-monsoon season. On the contrary, several workers have reported higher incidence of the osteomalacia or peg leg during summer in cows (Prodanov, 1952; O' Moore, 1952) and buffaloes (Bhikane *et al.*, 2003). Highest prevalence during monsoon season was attributed to exclusive feeding of dry roughages naturally deficient in phosphorus and non-availability of greens for longer periods due to prevailing drought situation.

Physiological status-wise distribution

Highest incidence of osteomalacia was observed in lactating buffaloes (63.49%) followed by pregnant buffaloes (36.51%). The results of present study are in agreement with those of Barns and Jephcott (1955); Bhikane *et al.* (2003).

Lactation stage-wise distribution

Highest incidence of osteomalacia was observed in mid (75%) lactation (4 to 6 months) followed by early (15%) lactation (0 to 3 months) and least (10%) in late lactation (>6 month). The results of present study are in agreement with those of Bhikane *et al.* (2003). The continuous drain of phosphorus through milk in lactating buffaloes receiving low phosphorus diet (dry forages) for prolonged period might have resulted in depletion of body reserves leading to bone resorption and manifestation of clinical signs during mid to late lactation (Radostits *et al.*, 2010).

Pregnancy stage-wise distribution

Highest occurrence of osteomalacia was observed during mid gestation (52.39%) followed by late gestation (33.33%) and least in early gestation period (14.28%). The present

findings could be attributed to increased need of phosphorus and high-energy demand for growth of fetus during mid to late pregnancy accompanied by poor nutritional status (NRC, 2001).

Milk yield-wise distribution

Milk yield wise highest occurrence of osteomalacia was observed in buffaloes yielding >4 litre/day (93.66%) as compared to those yielding <4 litre/day (6.34%). The results of present study are similar to findings of Abdou *et al.* (1986); Vaudey (1946); McTaggart (1959); Bhikane *et al.* (2003).

Feeding pattern-wise distribution

Feeding pattern wise highest occurrence of osteomalacia was observed in buffaloes fed with dry fodder alone or with minimum concentrate (80.96%) followed by dry fodder with some greens and minimum concentrate (15.87%) and greens *ad. lib.* (3.17%). Similarly earlier workers have mentioned that dry fodder accentuates phosphorus deficiency (Hungerford 1975; Underwood 1981; Bhikane *et al.*, 2003).

Clinical signs

The duration of illness in osteomalacia-affected buffaloes ranged from 15 to 90 days indicated chronic nature of the disease. No significant difference was observed in body temperature (100.69 ± 0.09 vs. 100.14 ± 0.05), heart rate (52.73 ± 0.73 vs. 49.00 ± 0.67) and respiration rate (24.34 ± 0.28 vs. 23.16 ± 0.51) of osteomalacia-affected buffaloes compared to normal healthy buffaloes. Highly significant decrease ($P < 0.01$) in the ruminal motility (2.53 ± 0.09 vs. $4.41 \pm 0.14/5$ minutes) was seen in affected buffaloes in comparison to healthy buffaloes.

The milk yield was found significantly ($P < 0.01$) declined (5.76 ± 0.22 to 2.64 ± 0.17 litre/

day) during illness as compared to before illness. Milk yield was normal in early stage of disease that later declined with progression of disease. In face of dietary deficiency, early to mid-lactation fulfils the phosphorus requirement by its resorption from bones until its saturation (Underwood, 1981). Around 39.68% of ailing buffaloes revealed anoestrus over a period of three months of lactation indicating reduced fertility owing to phosphorus deficiency. The findings are in agreement with those of O'Moore (1952); Brooks *et al.* (1984).

The affected animals were dull, depressed, and preferred recumbency due to weakness. In mild and early cases, the appetite was normal to slightly reduced (42.85%), where as it was severely reduced in long standing cases (20.63%). Pica was observed in 9 buffaloes out of 63 cases of osteomalacia as also reported by Theiler and Green (1932); Morrison (1984); Bhikane *et al.* (2003). The body condition was uniformly poor to emaciated in all buffaloes suffering from osteomalacia. In most cases the loss of body weight was found to be the earliest sign of the disease. In mild cases, the body condition was fair to poor (25.39%), whereas poor to emaciated in moderate cases (28.57%) and highly emaciated / hide bound in severely affected long standing cases (42.85%). The hair coat was invariably, rough, dry and lacked brightness. The loss of body weight observed in this study has been attributed to depression of voluntary feed intake (McTaggart, 1959), reduction in cellulose digestion and microbial protein synthesis (McDowell, 1992) and inefficient utilization of feed (Underwood, 1981).

The shifting lameness was a characteristic sign in affected cases. Mild to severe lameness and varying degree of stiff gait was the frequently observed symptom and its severity varied depending on stage of the disease. Phosphorus along

with calcium provides compactness or rigidity to the skeleton. The spongy bones serves as a reserve store of calcium and phosphorus and these stores are used when the daily intake is insufficient. If the shortage continues unduly long, the reserve in the spongy bone is exhausted and the minerals will be drawn from shafts and other supporting structures of bones. As a result, the bones became porous and weak. The condition is called as 'osteomalacia' (O Moore, 1952; Morrison, 1984).

The affected buffaloes were disinclined to move and showed great difficulty and discomfort when forced to move. The stiffness of body was increased immediately after standing characterized by abducted elbows and arched back posture for long time. During later stage of the disease, the buffaloes remained recumbent and if forced to rise they exhibited pain. Although severely affected buffaloes were able to lift hind portion of the body, faced great difficulty in lifting chest portion and remained on knee joint for longer time, while standing up. The postural change could have occurred to provide architectural strength to structurally weakened fragile skeleton to cope up with the weight of the body by endeavoring to centralize the legs underneath it (Barnes and Jephcott, 1955). The clinical cases of osteomalacia were classified as mild, moderate and severe depending upon stiffness of gait and difficulty in rising.

Haematology

Haemoglobin (10.71 ± 0.24 vs. 11.06 ± 0.32 g/dl) and packed cell volume (31.33 ± 0.70 vs. $37.48 \pm 0.97\%$) values were significantly ($P < 0.05$) decreased in affected buffaloes as compared to healthy animals. Highly significant ($P < 0.01$) decrease in total erythrocyte count was observed in affected ($5.22 \pm 0.20 \times 10^6/\mu\text{l}$) buffaloes

compared to healthy ($6.49 \pm 0.19 \times 10^6/\mu\text{l}$). The mean corpuscular volume was increased ($P < 0.05$) significantly in osteomalacic buffaloes (54.01 ± 1.18 fl) compared to healthy (47.16 ± 1.87 fl) buffaloes. The mean corpuscular haemoglobin (21.62 ± 0.49 vs. 19.68 ± 0.48 pg) and mean corpuscular haemoglobin concentration (34.83 ± 0.50 vs. 31.58 ± 0.26 g/dl) was significantly higher in osteomalacia affected buffaloes compared to healthy buffaloes. The mean Hb, PCV, TEC and MCV values were indicative of macrocytic normochromic anemia. These finding are in agreement with those of Clark (1974); Verma (1995); Bhikane *et al.* (2003); Singh *et al.* (2012). The leukogram revealed highly significant ($P < 0.01$) increase in granulocyte count in ailing buffaloes ($58.99 \pm 1.42\%$) compared to healthy buffaloes ($55.53 \pm 3.50\%$).

Biochemistry

Plasma phosphorus

Highly significant reduction ($P < 0.01$) in mean plasma phosphorus was observed in osteomalacia affected buffaloes (2.54 ± 0.07 mg/dl) as compared to healthy buffaloes (4.81 ± 0.20 mg/dl). The results of present study are in agreement with those of Prodanov (1952); Kalra *et al.* (1979); Verma (1995); Kishan and Sridhar (2000); Bhikane *et al.* (2003); Singh *et al.* (2012). Osteomalacia associated with hypophosphatemia in buffaloes of study region could be attributed to very low soil phosphorus, deficiency of phosphorus and wide Ca:P ratio in diet due to exclusive feeding of dry roughages for prolonged period due to draught resulting in decreased availability of phosphorus, non-supplementation of diet with concentrates and mineral mixture, continuous drainage of phosphorus through milk of lactating buffaloes and increased requirement for growth of foetus in pregnant animals (Prodanov, 1952; Pirzada and

Hussain, 1998; Bhikane *et al.*, 2003).

Plasma calcium

In present study plasma calcium levels were significantly ($P<0.05$) lower in buffaloes affected with osteomalacia (9.65 ± 0.22 mg/dl) as compared to normal healthy buffaloes (10.15 ± 0.26 mg/dl). The results of present study are in agreement with those of Bhikane *et al.* (2003); Soaves and Pevira (1967).

Alkaline phosphatase

Highly significant ($P<0.01$) increase in alkaline phosphatase levels was observed in buffaloes affected (290 ± 12.18 U/L) with osteomalacia compared to healthy buffaloes (77.16 ± 5.49 U/L). The results of present study are in agreement with earlier studies by Hungerford (1975); Underwood (1981); Smith (1990); Kishan and Sridhar (2000); Bhikane *et al.* (2003). Increased serum alkaline phosphatase levels are associated with increased osteoblastic activities in bones of osteomalacia-affected animals (Benjamin, 1978).

Glucose

No significant difference was observed in plasma glucose concentration in buffaloes affected with osteomalacia (54.13 ± 0.81 mg/dl) compared to normal healthy buffaloes (51.25 ± 1.21 mg/dl). The results of present study are in agreement with those of Fishwick *et al.* (1974).

Calcium: phosphorus ratio

Calcium: phosphorus ratio was significantly ($P<0.01$) wide in buffaloes affected with osteomalacia (4.00 ± 0.15) compared to healthy buffaloes (2.12 ± 0.05). The present observations are in agreement with Bhikane *et al.* (2003) who reported significantly higher Ca:P ratio in buffaloes

affected with osteomalacia (4.35 ± 0.17 mg/dl) compared to normal healthy buffaloes (2.01 ± 0.26). The values in-between 1.45 to 2.15 are considered normal (Philipov, 1993). The values either below 1.45 or above 2.15 are considered abnormal and need correction of dietary Ca, P and cholecalciferol.

Radiological examination

Radiographic examination of metatarsal bone and carpal joint was revealed proliferation of osteophytes in carpal bones. Demineralization of canon bones was also observed in some radiographs. Radiographic examination of coccygeal bones revealed osteolytic changes indicated by decreased radiographic density in last few vertebrae. Similar to present study Fishwick *et al.* (1974) have used tail bone radiography for diagnosis of bone deformities in cows and reported decrease in the radiographic density of the tail bones and concluded radiographic examination is better indicator for diagnosis of bone diseases rather than bone ash. Bhikane *et al.* (2003) observed increased radio density particularly at epiphyseal and metaphyseal region and increased inter vertebral space and thinning of coccygeal vertebrae suggestive of demineralization of bone.

Soil and fodder calcium and phosphorus

The soil calcium carbonate (CaCO_3) was found in the range of 6.8 to 15.8% with average value of $11\pm 0.65\%$ from disease prone area indicating calcareous nature of soil with adequate calcium. The available phosphate in analyzed soil sample was found in the range of 6.4 to 11.4 kg/ha with mean value of 8.92 ± 0.36 kg/ha indicating very low soil phosphorus in areas with prevalence of osteomalacia. The mean calcium concentration was $1.40\pm 0.14\%$ in jowar straw 1.36% in dry grass, $2.20\pm 0.21\%$ in soybean straw $1.36\pm 0.32\%$

in pigeon pea straw and $0.67 \pm 0.18\%$ in cotton seed cake indicating adequate calcium status (Ranjhan, 1991). The phosphorus content in the feed and fodder samples analyzed were $0.07 \pm 0.01\%$ in jowar straw 0.078% in dry grass, $0.18 \pm 0.03\%$ in soybean straw $0.22 \pm 0.01\%$ in pigeon pea and $0.54 \pm 0.07\%$ in cotton seed cake indicating deficient status of phosphorus in fodders. The findings are in agreement with those of Rose (1954).

The mean Ca:P ratio was 20.42 in jowar straw 17.43 in dry grass, 33.48 in soybean straw 6.34 in pigeon pea straw and 1.21 in cotton seed cake indicating wide Ca:P ratio. Phosphorus concentration declines dramatically as forages mature, however calcium concentration is less affected by advanced maturity thereby resulting in detrimental widening of the ratio of Ca with phosphorus (Underwood, 1981; Rose, 1954).

Treatment

The mean haematological and biochemical values before treatment and after recovery of Group I, II and III buffaloes ailing with osteomalacia are depicted in Table 1.

Group I

All the treated buffaloes in Group I recovered completely, indicating 100% recovery rate. The recovery period varied between 27 to 67 days. (Mean 40.5 ± 3.72 days). In mild cases recovery was faster than moderate to severe cases. Arneja *et al.* (1987) observed appreciable improvement in buffaloes suffering from osteomalacia like disease following administration of mineral mixture 50 gm orally daily for 30 days. Lau (1988) reported recovery in 6 buffaloes suffering from phosphorus deficiency with mineral mixture containing 57% dicalcium phosphate. Cheng *et al.* (1998) used sodium phosphate and calcium phosphate and

found that sodium phosphate was effective in treating hypophosphatemia in cattle. Singh *et al.* (2012) treated 34 buffaloes showing typical clinical signs of hypophosphatemia with dicalcium phosphate (DCP) 100 g/day orally for 20 days and observed marked improvement in body condition in 20 days.

Group II

All buffaloes treated in Group II received maximum 20 doses of parenteral buffered phosphorus preparation, after that due to incomplete recovery and increasing cost of treatment the animals were shifted to mineral mixture. None of the animal completely recovered by 20 days of treatment however, after continuous mineral mixture supplementation all the buffaloes in Group II were recovered. The recovery period varied between 23 to 68 days (Mean 39.2 ± 4.2 days). No data is available on efficacy of parenteral buffered phosphorus preparation in treatment of osteomalacia-affected buffaloes. However, Haque and Verma, (1989) compared efficacy of organic phosphorus preparation (Tonophosphon) 1 gm (5 ml) IM daily for 15 days and sodium acid phosphate (SAP) 40 gm orally daily for 15 days in induced hypophosphatemia in calves and concluded SAP was found superior to parenteral organic phosphorus preparation on the basis of cure rate, speedy recovery and restoration of serum inorganic phosphorus.

Group III

All the treated buffaloes in Group III recovered completely, indicating 100% recovery rate. The recovery period varied between 11 to 47 days (Mean 27.1 ± 4.05 days). Bhikane *et al.* (2003) compared organic phosphorus (Tonoricin) 2 ml/100 kg body weight IM on alternate days and mineral

mixture 10 gm/ 100 kg body weight orally daily with sodium acid phosphate 10 gm/ 100 kg body weight orally and mineral mixture (Nutrimilk) 20 gm/ 100 kg body weight orally daily for 30 days and revealed faster clinical recovery in group of buffaloes treated with sodium acid phosphate.

In present investigations, among three different treatment modalities recovery period was significantly ($P < 0.05$) lower in Group III buffaloes treated with combined buffered phosphorus and mineral mixture as compared to Group I (mineral mixture alone) and Group II buffaloes (buffered phosphorus alone). Based on the recovery period combined treatment with buffered phosphorus and mineral mixture proved effective and economic treatment modality for successful management of osteomalacia in buffaloes.

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Figure 1. Six years old osteomalacic buffalo showing difficulty while getting up and kneeling on carpal joint.



Figure 2. Emaciation with arched back posture and stiff gait in 8 year sold buffalo suffering from osteomalacia.

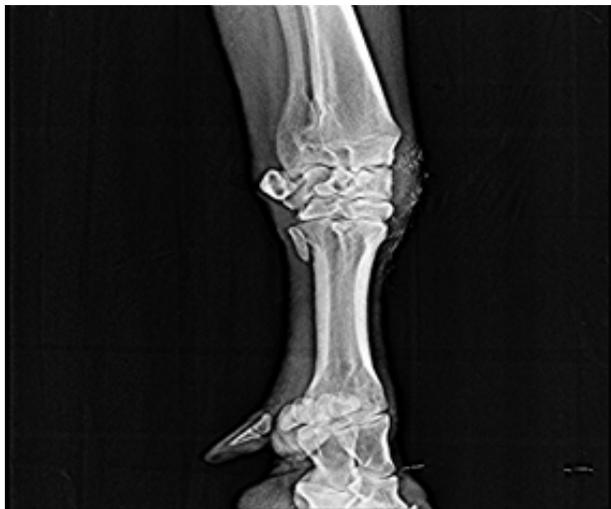


Figure 3. Osteolytic changes seen in carpal bone of buffalo suffering from osteomalacia on radiography.



Figure 4. Osteomalacic buffalo showing reduction in body weight and rough skin coat in Group III (Buffered phosphorus + Mineral Mixture) before treatment.



Figure 5. Restoration of body weight and lustrous skin coat after treatment of 60 days in Group III (Buffered phosphorus + Mineral Mixture).

Table 1. Mean haemato-biochemical values before and after treatment of osteomalacia affected buffaloes with different treatment modalities.

Sr. No.	Parameter	Group I (mineral mixture)			Group II (buffered phosphorus)			Group III (mineral mixture + buffered phosphorus)		
		Before treatment	After treatment	't' value	Before treatment	After treatment	't' value	Before treatment	After treatment	't' value
1.	Hb (g/dl)	10.7±0.83	10.23±0.83	0.692 ^{NS}	9.44±0.39	9.97±0.30	1.294 ^{NS}	9.44±0.39	9.97±0.30	1.294 ^{NS}
2.	PCV (%)	31.49±2.41	29.63±1.28	1.037 ^{NS}	29.91±1.59	28.35±0.87	1.03 ^{NS}	30.61±1.14	29.28±1.29	1.023 ^{NS}
3.	TEC (×10 ⁶ µl)	6.24±0.59	6.99±0.49	1.287 ^{NS}	5.61±0.47	6.39±0.40	1.59 ^{NS}	6.57±0.45	6.74±0.51	0.209 ^{NS}
4.	TLC (×10 ³ µl)	11.39±0.74	07.80±0.53	3.96**	11.00±1.03	08±0.32	2.04 ^{NS}	11.04±0.40	08.41±0.60	3.64**
5.	MCV (fl)	53.70±3.08	45.50±3.33	2.703*	58.2±2.98	49.60±2.09	2.511*	49.50±2.75	44.90±2.39	1.112 ^{NS}
6.	MCH (pg)	18.34±1.19	15.69±1.18	2.101 ^{NS}	18.69±1.34	17.60±1.96	0.747 ^{NS}	17.19±0.87	16.85±1.11	0.672 ^{NS}
7.	MCHC (g/dl)	34.09±0.56	33.36±1.95	0.373 ^{NS}	32.09±1.30	34.66±0.98	1.485 ^{NS}	36.39±1.29	37.04±1.29	0.353 ^{NS}
8.	PLT (× 10 ³ µl)	234±36.61	223.8±21.90	0.317 ^{NS}	266.2±34.61	198.8±23.54	1.586 ^{NS}	252.8±23.62	274.50±25.18	0.594 ^{NS}
9.	Lymphocyte (%)	48.19±2.76	49.17±2.71	0.317 ^{NS}	47.28±2.77	59.24±3.72	2.717*	48.03±3.91	51.47±1.33	0.896 ^{NS}
10.	Granulocyte (%)	48.16±3.19	46.39±3.56	0.428 ^{NS}	49.53±2.78	37.62±3.55	2.607*	48.82±3.42	45.83±1.09	0.812 ^{NS}
11.	Calcium (mg/dl)	9.8±0.45	10.58±0.12	1.856 ^{NS}	10.21±0.28	10.57±0.07	0.657 ^{NS}	10.16±0.53	10.54±0.07	0.661 ^{NS}
12.	Phosphorus (mg/dl)	2.66±0.19	5.4±1.2	15.04**	2.4±0.14	5.56±0.26	15.012**	2.07±0.05	6.06±0.16	24.59**
13.	Ca : P ratio	4.04±0.58	1.96±0.04	3.654**	3.5±0.21	1.94±0.08	7.492**	4.97±0.36	1.745±0.04	9.117**
14.	ALP (U/L)	296.8±15.28	88.2±5.95	13.27**	222.5±17.71	88.1±4.52	16.513**	242.4±29.83	93.9±5.09	4.906**
15.	Glucose (mg/dl)	55.6±1.26	55.5±0.50	0.084 ^{NS}	54.8±0.46	55.51±0.43	0.389 ^{NS}	53.4±1.91	56±0.91	1.364 ^{NS}

s = Non significant

* = Significant (P<0.05)

** = Highly significant (P<0.01)

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