www.ThePharmaJournal.com

The Pharma Innovation

Manjunath Patil
Department of Veterinary Surgery and Radiology, Veterinary College, Karnataka Veterinary, Animal and Fisheries Sxciences University, Bidar, Karnataka, India

Dilipkumar D
Professor, Veterinary Surgery and Radiology, Department of Veterinary Surgery and Radiology, Veterinary College, Karnataka Veterinary, Animal and Fisheries Sciences University, Bidar, Karnataka, India

Shivaprakash BV
Professor and Head, Veterinary Surgery and Radiology and Dean, Veterinary College, Department of Veterinary Surgery and Radiology, Veterinary College, Karnataka Veterinary, Animal and Fisheries Sciences University, Bidar, Karnataka, India

Vivek R Kasaralikar
Professor, Veterinary Medicine, Veterinary College, Department of Veterinary Surgery and Radiology, Veterinary College, Karnataka Veterinary, Animal and Fisheries Sciences University, Bidar, Karnataka, India

Vinay P Tikare
Assistant Professor, Veterinary Pharmacology and Toxicology, Veterinary College, Department of Veterinary Surgery and Radiology, Veterinary College, Karnataka Veterinary, Animal and Fisheries Sciences University, Bidar, Karnataka, India

Ramesh BK
Animal Scientist, Krishi Vigyan Kendra, UAS, Department of Veterinary Surgery and Radiology, Veterinary College, Karnataka Veterinary, Animal and Fisheries Sciences University, Bidar, Karnataka, India

Correspondence
Manjunath Patil
Department of Veterinary Surgery and Radiology, Veterinary College, Karnataka Veterinary, Animal and Fisheries Sciences University, Bidar, Karnataka, India

Physiological and haemato-biochemical changes during repair of femur fracture in dogs

Manjunath Patil, Dilipkumar D, Shivaprakash BV, Vivek R Kasaralikar, Vinay P Tikare and Ramesh BK

Abstract
The research was conducted in clinical cases of twenty four dogs with femur fracture and they were randomly divided into three groups *viz.,* group I, II and III with eight animals in each group. Group I, II and III animals were treated with Dynamic compression plating, Locking compression plating and Intramedullary interlocking nailing techniques respectively. Healing of fractures was studied on before operation, immediately after operation, 30th, 60th and 90th post-operative days in 3 groups based on physiological and haemato-biochemical findings. In the present study bradypnoea, bradycardia and hypothermia were found on post-operative days. Progressive increase of packed cell volume, haemoglobin and total erythrocyte count on different post-operative days was seen. Physiological leucocytopenia, neutropenia and relative lymphocytosis was observed on post-operative days. Monocyte and eosinophil count were within physiological range in three groups of animals. Slight hypercalcimia, slight hyperphosphatemia and progressive decrease of serum alkaline phosphatase were found on different post-operative days. However, all physiological and haemato-biochemical findings during repair of femur fracture in dogs were within the normal physiological range in three groups of animals.

Keywords: Physiological, haemato-biochemical, dogs, femur, fracture

Introduction
A fracture is a break in continuity of hard tissues like bone, cartilage. Trauma is the most common cause of fractures in small animals, and can occur due to bending, torsional, shearing and compression forces, eventually resulting in oblique, wedge fragment, spiral or comminuted fractures. Fracture of long bone, particularly femur in dogs is a very painful condition. If timely care is not taken, it may result in complications, i.e., non-union, malunion and damage to muscles, haematoma, seroma, suppuration and gangrene of limb. To avoid such types of complications, cases should be treated at the earliest. Femoral fractures are generally not amenable to conservative repair, and some kind of internal fixation is generally required (Beale, 2004) [10]. The primary goal of fracture treatment is to achieve a healed fracture with normal bone alignment and promote immediate function of the affected limb. To know the healing of fracture presently radiography is routine practice. However, it's difficult in field conditions to take the x-ray every time after orthopedic operation. Also, getting exposure to x-rays every time is harmful both to the patient and radiographer. So, in this context, other parameters *viz.,* physiological and haemato-biochemical findings has any significant role on healthiness of patient and healing of fractures. More ever, there is a paucity of reports available on role of these parameters with respect to healing of fractures. Therefore, the present paper reports the physiological and haemato-biochemical changes during repair of femur fracture in dogs their by determines the healing of fractures.

Material and methods
Twenty four dogs of 2 months to 3 years of age with femoral fractures were presented to Department of Surgery and Radiology, Veterinary College, KVAFSU, Bidar constituted the present study material. Clinical symptoms before operation were non-weight bearing, swelling on thigh region, evinces crepitating sound and pain on palpation and limping. The similar clinical symptoms were observed by Bennet and May (1995) [11] and Johnson *et al.* (1998) [19]. These cases were divided into groups I, II and III consisting of eight dogs in each group and were treated with dynamic compression plating, locking compression plating and Intramedullary interlocking nailing techniques respectively based on the type of fracture.
Food and water were withdrawn for 12 hours and 6 hours respectively before surgery in all the puppies. The affected limb hairs were clipped and shaved from rump region up to hock region

and scrubbed with chlorhexidine solution [1] followed by application of surgical spirit. The puppies were given pre-operative antibiotic amoxycillin sodium and cloxacillin sodium [2] injection @ 20 mg per kg body weight intramuscularly and anti-inflammatory meloxicam [3] injection @ 0.5 mg per kg body weight intramuscularly. The puppies were premedicated with atropine sulphate [4] injection @ 0.045 mg per kg body weight intramuscularly, followed by triflupromazine [5] injection @ 1 mg per kg body weight intravenously. After 15 minutes, anaesthesia was induced with 2.5 % thiopental sodium [6] injection intravenously at dose rate of 12.5 mg/ kg body weight. Anaesthesia was maintained using the thiopental sodium given in incremental doses 'to effect'. The animals were positioned in lateral recumbency with the affected limb above and were adequately draped. Approach of the femur fracture was performed by cranio-lateral incision (Piermatti and Greeley, 1993). Fracture reduction made mechanically by traction and manipulation. Post-operatively, antibiotic amoxycillin sodium and cloxacillin sodium injection @ 20 mg/kg body weight and meloxicam injection @ 0.5 mg/kg body weight intramuscularly for 8-10 days given. Daily surgical wound dressed aseptically and antiseptic cream (Lorexane [7]) applied. The skin sutures were removed 10- 14 days after operation.

Follow up of the animals was done up to 3 months. The physiological and haemato-biochemical observations were recorded on before operation, immediately after operation, 30th day, 60th day and 90th post-operative days in all groups of animals. The respiratory rate was monitored by observing the movement of rib cage. The heart rate was monitored by auscultation using stethoscope. The rectal temperature was recorded by placing clinical thermometer in the rectum of the dog.

For haematological observations, two ml blood was collected in EDTA coated vials using disposable syringes through cephalic or recurrent tarsal vein in all the cases. The estimations of packed cell volume (%), haemoglobin (g/dL), total erythrocyte count (x 10^6/μL) and total leukocyte count (x 10^3/μL) were carried out on fully automated haematology cell counter [7]. Blood smear for differential leukocyte count were stained with Giemsa stain and cells were counted using Battlement method as described by Jain (1993) [18] and individual cells were expressed in percentage.

For biochemical observations, blood samples were collected in clot activator vials to separate the serum. The parameters *viz.,* serum calcium (mg/dL), serum inorganic phosphorus. (mg/dL) and enzyme serum alkaline phosphatase (IU/L) were estimated by using ARTOS [8] biochemical analyzer using respective diagnostic kit. Statistical analysis of data obtained was carried out by employing Student't' test as per the standard procedure outlined by Snedecor and Cochran (1994) [35].

Results and discussion
The details of all the physiological and haemato-biochemical values recorded in the present study were given in table 1.

Physiological Parameters
The physiological parameters like respiratory rate, heart rate and rectal temperature remained elevated on pre-operative day. The respiratory rate and heart rate fluctuated within the normal physiological limits on post-operative days. However, rectal temperature was fluctuated within normal limits from 30th to 90th post-operative days. The elevated physiological parameters on pre-operative days could be attributed to the stress on the animal due to fracture (Kelly, 1974) [21].

Haematological Parameters
The Packed cell volume and haemoglobin level on the 60th and 90th post-operative days were statistically higher when compared to pre-operative intervals in all the groups of animals. However, packed cell volume and haemoglobin were fluctuated within the normal physiological limits. The total erythrocyte count was significantly elevated on 60th post-operative day when compared to pre-operative day in all the groups of animals. However, the total erythrocyte count was within the normal physiological limits. This showed that progressive increase of packed cell volume, haemoglobin and total erythrocyte count on post-operative days indicating erythropoisis, however, within the normal physiological limits. This finding was in agreement with the findings of Singh *et al.* (2008) [33], they observed non-significant variation of packed cell volume, haemoglobin and total protein from the base values.

The total leukocyte count was higher on pre-operative day when compared to post-operative days in all the groups of animals. Physiological leucocytopenia was seen in all the groups of animals suggestive of gradual decrease in inflammatory reaction. This finding was in accordance with Maiti *et al.* (1999) [24] reported the total leukocyte count was increased significantly from the 1st post-operative day and remained elevated up to the 30th post-operative day in all animals and then gradually returned towards normal level at the 60th post-operative day indicating resolution of inflammation and surgical stress. Leucocytosis occurred in condition where there was corticosteroid release in state of stress, pain, anaesthesia, trauma and surgical manipulation.

The differential leuckocyte count like neutrophil was significantly decreased on 60th and 90th post-operative day when compared to before operation in all the groups of animals. Contralaterally, the lymphocyte count was statistically increased on 30th, 60th and 90th post-operative days when compared to pre-operative day in all the groups of animals. This showed that, neutropenia with relative lymphocytosis indicating gradual decrease of inflammatory reaction. However, they were within normal physiological limits on different post-operative days in all the groups of animals.

The monocyte count was higher on pre-operative day when compared to post-operative days in all the groups of animals. However, it was fluctuated within normal physiological limits in all the groups of animals. Also, eosinophil count between different post-operative days fluctuated within normal physiological limits.

These findings were in agreement with the findings of Tembhurne *et al.* (2010) [37] observed that packed cell volume, haemoglobin, total erythrocyte count, total leukocyte count, monocytes, eosinophils and basophils count were within normal physiological range. However, lymphocytopenia was observed on 10th post-operative day on treatment of the femur fracture in canine with the horn-peg prepared from the bovine horn. Also, Hansda *et al.* (2012) [13] observed non-significant changes of haemoglobin, total erythrocyte count, total leukocyte count and differential leukocyte count on 0 day and 20th day.

Our present findings were differed with the findings of Nagaraja (1996) [26], who observed that neutrophilia, lymphopenia, monocytosis and eosinophilia during healing of femur fracture and haemoglobin was in the normal range.

Biochemical parameters

The serum calcium level on the 60th and 90th post-operative days was statistically higher when compared to pre-operative intervals in all groups of animals. However, the serum calcium level was within the higher normal physiological limits. These findings are in agreement with the earlier workers *viz.,* Lauren and Kelly (1969) [23], Pandey and Udapa (1981) [27], Rao (1991) [30] and Vasantha (1991) [38].

The serum inorganic phosphorous level was significantly elevated on 30th, 60th and 90th post-operative days when compared to pre-operative day. However, the serum inorganic phosphorus value was within the normal physiological limits. The results are in agreement with the findings of Henderson and Nobel (1926) [15], Pandey and Udapa (1981) [27], and different from findings of Soliman and Hassan (1964), Vasantha (1991) [38] and Prachasilpchai *et al.* (2003) [29], who observed a non-significant change in serum phosphorus level during fracture healing.

The serum alkaline phosphatase level was significantly higher on pre-operative day when compared to the post-operative days in all groups of animals. These results are in agreement with the earlier workers Hunsberger and Ferguson (1932) [17], Brinker (1965) [12], Shirfin (1970) [32], Katerjian and Arsenia (1975) [20], Singh *et al.* (1976) [34], Hosking (1978) [16], Pandey and Udapa (1981) [27], Volpin *et al.* (1986) [39], Sahay *et al.* (1988) [31], Vasantha (1991) [38], Maiti *et al.* (1999) [24], Manjubala *et al.* (2001) [25], Hegade *et al.* (2007) [14] and Phaneendra *et al.* (2016) [28]. The elevated alkaline phosphatase level could be attributed to proliferation of osteogenic cells and maximum contribution from periosteum of destructed bone, which is a rich source of alkaline phosphatase. In Group III animals higher level of serum alkaline phosphatase was present when compared to group I and II animals, respectively, this might be due to less stable fixation in group III. This finding differed from the findings of Volpin *et al.* (1986) [39] and Komnenou *et al.* (2005) [22].

Table 1: Mean ± SE, values of physiological and haemato-biochemical in different groups of femoral fracture repair in dogs

Sl. No.	Parameter	Groups	Before operation	Immediately after operation	30th day	60th day	90th day
1	Respiratory Rate (breaths/min)	I	36.88±2.15a	24.25±3.78**a	27.25±0.68**a	26.63±0.83**a	25.88±1.13** a
		II	37.25±2.96a	22.13±0.94**b	29.63±1.93*a	28.38±0.83*a	26.25±1.57**a
		III	38.50±1.38a	20.63±1.25**a	31.75±0.90**b	30.38±1.55*a	29.13±1.35*a
2	Heart Rate (beats/min)	I	101.50±5.74a	70.25±2.38**a	83.13±2.29**a	86.38±5.21a	84.25±3.90**a
		II	106.13±2.36a	68.75±2.28**a	82.25±1.87**a	85.50±1.70**a	83.63±3.87**a
		III	111.75±1.66a	73.75±5.07**a	90.63±2.90**b	84.88±1.99**a	82.13±6.18**a
3	Rectal Temperature (0F)	I	102.83±0.31a	98.14±0.70**a	100.40±0.10**a	101.39±0.31**a	101.66±0.24**a
		II	102.29±0.23a	99.38±0.69**a	101.58±0.23*b	100.25±0.14**b	101.31±0.27*a
		III	103.23±0.25ab	98.48±0.68**a	101.89±0.32**bc	102.39±0.29*c	102.54±0.30b
4	Packed Cell Volume (%)	I	31.50±1.00a	34.00±0.73a	44.83±1.42**a	46.23±1.08**a	47.60±0.95**a
		II	33.63±1.90a	37.40±1.44a	42.00±1.13**a	48.13±1.08**a	52.38±1.43**b
		III	30.09±1.30a	32.23±1.18ab	36.06±2.65ab	39.00±1.06**b	42.88±2.40**a
5	Haemoglobin (g/dL)	I	9.29±0.18a	10.13±0.39 a	11.20±0.30**a	12.43±0.38**a	13.15±0.13**a
		II	9.60±0.29a	10.71±0.21**a	11.78±0.36**a	12.85±0.19**a	14.03±0.44**a
		III	10.09±0.16ab	10.72±0.23*a	12.13±0.82*a	13.06±0.77**a	14.28±0.53**a
6	Total Erythrocyte Count (x10^6/μL)	I	5.05±0.18a	5.51±0.76a	6.31±0.22**a	7.66±0.19**a	7.90±0.15**a
		II	5.63±0.25a	6.22±0.11*a	7.03±0.12**b	8.05±0.12**a	9.11±0.09**b
		III	5.07±0.23a	5.37±0.28ab	6.08±0.35*a	6.53±0.42**b	7.22±1.23ab
7	Total Leukocyte Count (x10^3/μL)	I	13.13±0.32a	12.71±0.72a	11.59±0.52*a	10.68±0.21**a	9.32±0.17**a
		II	15.09±0.34b	13.45±0.50a	12.63±0.71**a	10.40±0.59**a	8.84±0.27**a
		III	14.14±0.66ab	13.23±2.24a	12.53±1.28a	11.18±1.34a	9.14±0.55**a
8	Neutrophil (%)	I	78.75±2.76a	71.88±1.58*a	64.63±1.37**a	62.25±0.97**a	61.75±1.35**a
		II	80.13±1.54a	74.13±1.67*a	67.13±1.66**a	61.50±1.86**a	60.38±1.57**a
		III	75.50±3.27a	69.38±1.10ab	63.38±3.18*a	62.13±2.38**a	60.88±0.80**a
9	Lymphocyte (%)	I	18.00±1.01a	25.88±1.33**a	32.63±1.62**a	33.88±1.58**a	34.38±1.46**a
		II	16.38±0.50a	23.13±2.06**a	28.88±0.94**a	34.88±1.67**a	35.88±2.17**a
		III	21.13±1.36ab	28.13±2.33*a	33.38±1.68**ab	33.75±1.93**a	34.75±2.50**a
10	Monocyte (%)	I	1.88±0.48a	0.88±0.37a	0.63±0.17*a	1.13±0.12a	1.38±0.50a
		II	1.50±0.31a	0.88±0.12a	1.38±0.25b	1.25±0.29a	1.13±0.21a
		III	2.13±0.12a	1.38±0.17**ab	2.13±0.21c	3.13±0.51b	3.50±0.53**ab
11	Eosinophil (%)	I	2.13±0.57a	1.25±0.15a	1.88±0.45a	2.63±0.30 a	2.38±0.25 a
		II	2.00±0.35a	1.88±0.28a	2.50±0.40a	2.25±0.52 a	2.63±0.39 a
		III	1.50±0.25a	1.25±0.15a	1.13±0.12ab	1.06±0.06b	0.88±0.12 *b
12	Serum Calcium (mg/dL)	I	8.95±0.09a	9.07±0.19a	9.51±0.15**a	10.31±0.22 **a	11.10±0.17**a
		II	8.79±0.22a	9.10±0.29a	9.81±0.22**a	10.34±0.16**a	10.73±0.21**a
		III	9.14±0.18a	9.29±0.50a	9.83±0.49a	10.90±0.20**b	11.34±0.39**a
13	Serum Inorganic Phosphorus (mg/dL)	I	2.82±0.13a	2.95±0.12a	3.73±0.16**a	4.19±0.17**a	4.68±0.21**a
		II	3.19±0.07b	3.34±0.19a	3.92±0.22**a	4.58±0.10**a	4.95±0.05**b
		III	3.11±0.09a	3.54±0.16*b	3.93±0.09**a	4.44±0.13**a	4.98±0.15**a
14	Serum Alkaline Phosphatase (IU/L)	I	120.18±1.88a	110.35±1.51**a	88.30±1.40**a	78.20±1.31**a	73.19±1.18**a
		II	118.46±1.29a	104.83±0.87**b	82.15±1.19**b	75.10±0.86**a	70.60±1.01**a
		III	122.22±1.79a	111.67±0.76**a	88.63±1.22**a	83.25±1.42**b	76.03±1.69**b

Conclusion

In the present study, respiratory rate, heart rate and rectal temperature were elevated on pre-operative days and there after values declined to normal range suggesting reduction of fracture stress on the animals. Progressive increase of packed cell volume, haemoglobin and total erythrocyte count from immediately after operation to 90th day indicating erythropoisis. Physiological leucocytopenia, neutropenia with

relative lymphocytosis on different post-operative days indicating resolution of inflammation and surgical stress. Monocyte and eosinophil counts were within normal physiological limits in all the groups of animals indicating free from anaphylactic reactions. Slight hypercalcimia, slight hyperphosphatemia and progressive decrease of serum alkaline phosphatase on different post-operative days indicating osteosynthesis of bone. In conclusion, the above physiological and haemato-biochemical findings on different post-operative days helped in knowing the healing of fracture.

References
1. Savlon-Chlorhexidine gluconate solution IP, 1.5 % v/v, Johnson and Johnson Ltd., Baddi
2. Moxel-Amoxycillin sodium and Cloxacillin sodium IP, 300 mg/ml, 3g vial, Alembic pharmaceuticals Ltd., Vadodara.
3. Melonex-Meloxicam IP, 5 mg/ ml, Intas pharmaceuticals Ltd., Ahmedabad
4. Atropine sulphate-Atropine sulphate IP, 1mg/ml, 10ml vial, Domesto Ltd., Vijayawada.
5. Siquil-Triflupromazine HCl IP, 20 mg/ml, 5 ml vial, Sarabhai Chemicals Ltd., Vadodara.
6. Thiosol Sodium- Thiopentone sodium IP 500 mg vial, Neon Laboratories Ltd., Mumbai.
7. Lorexane-Gamma Benzene Hexachloride and Profavine Hemisulphate with Cetrimide cream IP 0.10 % w/w. Virbac Animal Health India Pvt Ltd., Mumbai.
8. Fully automated haematology cell counter-Automatic Blood Cell Counter, Model-PCE 210 (Erma Tokyo).
9. ARTOS® Biochemical analyser and kits, M/s Swemed diagnostics, Bengaluru, India.
10. Beale B. Orthopedic clinical techniques femur fracture repair. Clinical Techniques in Small Animal Practice. 2004; 19(3):134-150.
11. Bennet D, May C. Joint diseases of dogs and cats. Textbook of Veterinary Internal Medicine: Diseases of the Dog and Cat. Edt. Ettinger SJ, Fedman EC. Edn 4, W.B. Saunders Company, Philadelphia, 1995, 2032-2077.
12. Brinker WO. The clinical approach to the fractures in small animals. Veterinary Record. 1965; 76:1412-1422.
13. Hansda P, Gahlod BM, Akhare SB, Dhakate MS, Upadhye SV, Panchbhai VS *et al.* Comparative evaluation of Steinmann pin, Kuntscher nail and interlocking nail for femur fracture repair in dog. Indian Journal of Veterinary Surgery. 2012; 33(1):53-54.
14. Hegade Y, Dilipkumar D, Usturge S. Comparative evaluation of biochemical parameters during fracture healing in dogs. Karnataka Journal of Agricultural Sciences. 2007; 20:694-695.
15. Henderson MS, Noble TP. Serum calcium levels in humans following fractures. Journal of Bone and Joint Surgery. 1926; 8-A:607-611.
16. Hosking DJ. Changes in serum alkaline phosphatase after femoral fracture. Journal of Bone and Joint Surgery. 1978; 60:61-65.
17. Hunsberger AJ, Ferguson LK. Alkaline phosphatase during early fracture healing. Archives of Surgery. 1932; 24:1052-1056.
18. Jain NC. Essentials of Veterinary Hematology. Lea and Fabiger, Philadelphia, 1993, 417.
19. Johnson AL, Smith CW, Schaeffer DJ. Fragment reconstruction and bone plate fixation versus bridging plate fixation for treating highly comminuted femoral fractures in dogs: 35 cases (1987-1997). Journal of American Veterinary Medical Association. 1998; 213(8):1157-1161.
20. Katerjian AY, Arsenia C. Morphological and biochemical studies during differentiation and calcification of fracture callus cartilage. Clinical Orthopaedics. 1975; 107:266-273.
21. Kelly WR. Veterinary Clinical Diagnosis. Edn 2, Bailliere Tindall, London, 1974, 266-270.
22. Komnenou A, Karayannopoulou M, Polizopoulou ZS, Constantinidis TC, Dessiris A. Correlation of serum alkaline phosphatase activity with the healing process of long bone fractures in dogs. Veterinary Clinical Pathology. 2005; 34(1):35-38.
23. Lauren EL, Kelly PJ. Serum calcium in dogs during fracture healing. Journal of Bone and Joint Surgery. 1969; 51:298-301.
24. Maiti BK, Sen TB, Singh B, Sanki S. Haemato-biochemical changes following application of internal fixation technique in treatment of femur fracture in dog. Indian Journal of Animal Health. 1999; 38:133-134.
25. Manjubala RV, Kumar S, Sastry TP. Biocompatibility evaluation of biphasic calcium phosphate ceramics an *in vivo* study. Trends in Biomaterials and Artificial organs. 2001; 14:27-29.
26. Nagaraja BN. Plastic implants for femoral fractures in experimental dogs. M.V. Sc. thesis, University of Agricultural Sciences, Bangalore, India, 1996.
27. Pandey SK, Udapa KN. Effect of growth hormone on biochemical response after fracture in dogs. Indian Journal of Veterinary Surgery. 1981; 1(2):73-78.
28. Phaneendra, Dhana lakshmi N, Devi prasad V, Raju NKB. Evaluation of biochemical parameters for assessment of fracture healing in dogs. Journal of Livestock Science. 2016; 7:111-113.
29. Prachasilpchai W, Bupha-intr T, Kalpravidh M, Sarikaputi M. Serum bone-specific alkaline phosphatase of dogs with various bone condition. Thai Journal of Veterinary Medicine. 2003; 33(3):81-90.
30. Rao NV. Studies on acetabular fractures in dogs. Ph. D. thesis, Andhra Pradesh Agricultural University, Hyderabad, India, 1991.
31. Sahay PN, Moulvi BA, Prasad BA, Prasad RI, Bachoo BA. Studies on serum alkaline phosphatase activity during different stages of fracture healing in goats. Indian Journal of Animal Health. 1988; 11:408.
32. Shirfin IZ. Alkaline phosphatase in fracture healing. Clinical Orthopaedics. 1970; 70:212-216.
33. Singh K, Kinjavdekar P, Aithal HP, Gopinathan P, Amarpal Pawde AM, Singh GR *et al.* Comparison of dynamic compression plate with circular external skeletal fixator for correcting angular deformity after wedge osteotomy of canine antebrachium. Indian Journal of Veterinary Surgery. 2008; 29:87-92.
34. Singh H, Lovell JE, Schiller AG, Kenner GH. Serum calcium, phosphorus and alkaline phosphatase levels in dogs during repair of experimental ulnar defects. Indian Veterinary Journal. 1976; 53:862-865.
35. Snedecor GW, Cochran WB. Statistical Methods. Edn 8, IOWA State University Press, USA. 1994, 138-169.
36. Soliman FA, Hassan SYS. Serum calcium and phosphorus in rabbits during fracture healing with reference to parathyroid activity. Nature London. 1964;

204:693-694.
37. Tembhurne RD, Gahlod BM, Dhakate MS, Akhare SB, Upadhye SV, Bawaskar SS *et al.* Management of femoral fracture with the use of horn peg in canine. Veterinary World. 2010; 3(1):37-41.
38. Vasantha MS. Studies on effect of ultrasonic therapy and short wave diathermy on femoral fracture healing in canine. Ph.D. thesis, Andhra Pradesh Agriculture University, Hyderabad, India, 1991.
39. Volpin G, Rees JA, Ali SY, Bentley G. Distribution of alkaline phosphatase activity in experimentally produced callus in rats. Journal of Bone and Joint Surgery. 1986; 60:629-634.