



ISSN (E): 2277- 7695
ISSN (P): 2349-8242
NAAS Rating: 5.23
TPI 2021; SP-10(10): 482-485
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www.thepharmajournal.com
Received: 06-08-2021
Accepted: 18-09-2021

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Haemato-biochemical observations on treatment of femoral diaphyseal fracture using titanium alloy (Ti-6Al-7Nb) nails with elastic stable intramedullary nailing technique in dogs

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Abstract

A clinical study on elastic stable intramedullary nailing (ESIN) was conducted on six skeletally immature dogs of either gender irrespective of breed with transverse or short oblique mid diaphyseal femoral fractures using Titanium alloy (Ti-6Al-7Nb) nails. Hemato-biochemical parameters were evaluated before surgery, immediately after surgery and on 7th, 14th, 28th and 45th post-operative day. Haematological evaluation revealed no significant variations in all the six dogs. The variations in serum alanine amino transferase and serum creatinine values were statistically non-significant and were within the normal physiological range. Statistically significant elevation in Alkaline phosphatase values was observed from 7th post-operative day with peak value observed on 14th post-operative day and values came back normal by 45th post operative day. There was non-significant increase in the serum calcium and serum phosphorus levels of dogs upto 28th postoperative day and 14th postoperative day respectively and were within normal physiological range.

Keywords: Fracture, hemato-biochemical parameters, alkaline phosphatase, elastic stable intramedullary nailing

Introduction

Occurrence of diaphyseal fracture of long bones like femur, humerus, tibia, radius and ulna is a frequent orthopaedic condition in dogs. Femur was the most common long bone involved in canine fractures (Singh *et al.*, 2015) [21]. The highest number of femoral fractures were reported in dogs less than six months of age (Kallianpur *et al.*, 2018) [8]. Surgical management with Elastic Stable Intramedullary Nailing (ESIN) using titanium elastic nails is a minimally invasive technique and becoming popular for treatment of femoral diaphyseal fractures in pediatric patients in human medicine (Flynn *et al.*, 2004) [4]. Ti-6Al-4V alloy was used as implant because of its excellent specific strength, corrosion resistance, and biocompatibility characteristics (Niinomi, 2001) [15]. Titanium elastic nailing technique for long bone fracture management has advantages of stable fixation with faster healing in dogs (Singh *et al.*, 2014) [21]. Due to the elastic nature of the implant and its three point fixation, it could withstand compressive, angular and rotational forces at the fracture site (Hunter, 2005) [6]. Chincholi (2016) [3] reported that Elastic Stable Intramedullary Nailing (EISN) was advantageous over other surgical methods particularly in pediatric patients because it was simple and was a load sharing internal splint that did not violate open physis, allowed early mobilization, maintained alignment, micromotion at the fracture site was confirmed by the elasticity of the fixation and promoted faster external bridging callus formation. The periosteum was not disturbed and being a closed procedure, there was no disturbance of fracture haematoma there by reduced the risk of infection. Additionally, the ESIN technique had advantages like aesthetic post-operative scars, facile removal, short inpatient period, good cost-effectiveness ratio and less psychological impact on the patient (Ulici *et al.* 2020) [23].

Materials and methods

The clinical study on Elastic Stable Intramedullary Nailing (ESIN) was conducted on six skeletally immature dogs with femur fracture that were presented for the treatment at Department of Surgery and Radiology, Veterinary College Hebbal, Bengaluru.

Routine clinical, orthopaedic, neurological and radiographic examinations were conducted to diagnose the femur fractures. Only the cases confirmed with transverse mid diaphyseal and short oblique mid diaphyseal fractures of femur were selected for the study. The dogs with neurological signs and the dogs with open wounds at fracture site were excluded from the study. Inj. Ceftriaxone sodium @ 25 mg/kg body weight intravenously) was administered as an antimicrobial prophylaxis and Inj. Tramadol @ 2 mg/kg body weight intravenously was administered as pre-emptive analgesic. The dogs were pre-medicated with Inj. Atropine sulphate @ 0.04 mg/kg body weight subcutaneously followed by Inj. Xylazine hydrochloride @ 1 mg/kg body weight intramuscularly as pre-anaesthetic medication. General anaesthesia was induced and maintained with (2.5%) solution of Thiopentone Sodium @ 25 mg/kg body weight intravenously. Fracture was reduced and fixed by elastic stable intramedullary nailing technique. Postoperatively antibiotic Cephalexin @ 25 mg/kg body weight orally b.i.d for 10 days, surgical wound dressing and antiseptic cream application were followed. Carprofen @ 2 mg/kg body weight orally s.i.d for 3 days was used as analgesic. For hematological evaluation, two ml of blood sample was collected in EDTA vacutainer tubes. Hematological parameters viz. total erythrocyte count, total leukocyte count, differential leukocyte count and hemoglobin were estimated evaluated before surgery, immediately after surgery and on 7th, 14th, 28th and 45th post-operative day using mindray BC-2800Vethaemo-analyser. Two ml of blood sample was collected in serum vacutainer tubes and allowed to clot at room temperature for separation of serum. Biochemical parameters viz., serum calcium, serum phosphorus, serum alkaline phosphatase, serum alanine amino transferase (ALT) and serum creatinine were evaluated before surgery, immediately after surgery and on 7th, 14th, 28th and 45th post-operative day using Micro Lab Instruments RX-50 biochemical analyzer using respective diagnostic kit as per manufacture's instruction. All the results were statistically analyzed by unpaired t-test, using computer based statistical programme, Graph pad prism, and interpreted as per the procedure described by Snedecor and Cochran (1996) to arrive at a conclusion.

Results and discussion

The variations in the mean values of haemoglobin and total erythrocyte count were statistically non-significant and the values were within the normal physiological range. Non significant variation in haemoglobin and total erythrocyte count within the normal physiological range recorded during the procedure might be due to minimal hemorrhage during operative procedure and post surgery. Similar findings were observed by Nagaraja (1996) [14] and Chaurasia *et al.* (2019) [2]. However, non significant mild increase in haemoglobin and total erythrocyte count values till 45th post-operative day were seen, it might be due to erythropoiesis. Similar findings were recorded by Patil (2017) [16]. The variations in the mean

values of total leucocyte count were statistically non-significant and the values were within the normal range. However, leucocytosis was observed in the initial post-operative days which could be due to surgical stress and inflammatory process, but was decreased gradually from the 7th post-operative day. These Similar findings were reported by Mahesh, (2009) [12], Manjunatha (2010) [13] and Jain *et al.* (2018) [7]. The variations in the mean values of neutrophils, lymphocytes, monocytes and eosinophils percentage were statistically non-significant and the values were within the normal range (Table. 1). Similar findings were reported by Rathod (2013) [18] and Patil *et al.* (2017) [16].

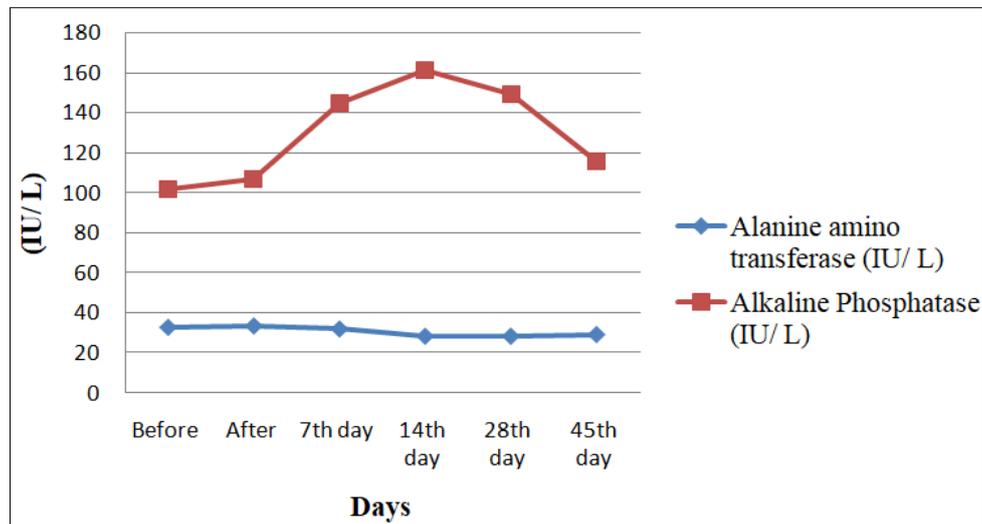
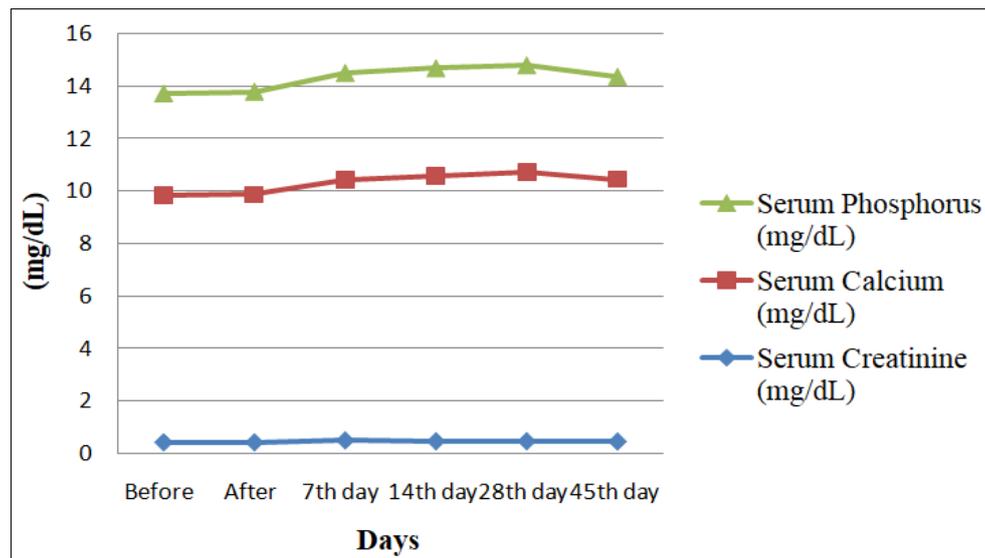
The variations in mean values of serum alanine amino transferase were statistically non-significant and were within the normal range (Fig. 1). This suggest that the procedure, implants, anesthetics and medication did not cause any damage to the liver. These findings were in accordance with Mahesh (2009) [12] and Tembhrne *et al.* (2010) [22]. There was significant ($P<0.05$) elevation of mean serum alkaline phosphatase values on 7th post operative day with peak value on 14th post-operative day and there was significant decrease from 14th to 45th post operative day (Table. 2). This might be attributed to the increased levels of serum alkaline phosphatase level to the increased chondroblastic proliferation and deposition of the calcium salts at the fracture site during initial bone healing process. The findings were in accordance with Chandy (2000) [1], Komnenou *et al.* (2005) [9], Mahendra *et al.* (2007) [11], Manjunatha (2010) [13], Phaneendra *et al.* (2016) [17], Kumar *et al.* (2018) [10], Jain *et al.* (2018) [7] and Reddy *et al.* (2020) [19]. However, Hegade *et al.* (2007) [5] and Patil *et al.* (2017) [16] recorded higher levels of serum alkaline phosphatase levels on pre-operative days when compared to the post-operative days and Mahesh (2009) [12] recorded non-significant variations throughout the study period. The variations in the mean values of serum creatinine level were statistically non-significant and the values were within the normal range (Fig. 2). This suggest that the procedure, implants, anesthetics and medication did not cause any damage to the kidney. There was non-significant increase in the serum calcium levels of dogs upto 28th post-operative day and the values were within the normal range (Fig. 2). The similar observations were made by Patil *et al.* (2017) [16], Jain *et al.* (2018) [7] and Reddy *et al.* (2020) [19]. However, Chaurasia *et al.* (2019) [2] found that significantly low serum calcium on 15th post-operative day and increase on 45th and 60th day and Kumar (2019) [10] found significant rise of the serum calcium on 14th post-operative day and reached normal physiological level on 45th day. A statistically non-significant increase in serum phosphorus was observed upto the 14th post-operative day and later varied non-significantly upto the 45th day (Table. 2). The values were within the normal range. The similar observations were observed by Chandy (2000) [1], Mahendra *et al.* (2007) [11], Mahesh (2009) [12], Jain *et al.* (2018) [7], Chaurasia *et al.* (2019) [2], Kumar (2019) [10] and Reddy *et al.* (2020) [19].

Table 1: Pre and post-operative mean \pm SE values of Haematological parameters of dogs subjected to ESIN

Days	Before Surgery	Immediately after Surgery	7th day	14th day	28th day	45th day
Haemoglobin (g%)	11.02 \pm 0.77	10.72 \pm 0.58	10.62 \pm 0.74	11.03 \pm 0.70	11.03 \pm 0.65	11.28 \pm 0.69
Total Erythrocyte Count (10 ⁶ Cells/mm ³)	5.52 \pm 0.33	5.39 \pm 0.27	5.63 \pm 0.23	5.57 \pm 0.26	5.88 \pm 0.21	6.39 \pm 0.25
Total Leucocyte Count (10 ³ Cells/mm ³)	12.35 \pm 0.33	12.66 \pm 0.49	14.71 \pm 3.14	14.21 \pm 2.01	13.83 \pm 0.70	12.56 \pm 0.56
Neutrophils %	72.47 \pm 1.07	72.77 \pm 0.74	75.38 \pm 1.58	74.79 \pm 2.10	71.71 \pm 0.93	71.10 \pm 0.34
Lymphocytes %	24.04 \pm 1.12	23.51 \pm 0.84	21.46 \pm 1.44	21.79 \pm 1.91	24.99 \pm 1.05	25.41 \pm 0.61
Monocytes %	2.39 \pm 0.28	2.64 \pm 0.36	2.15 \pm 0.30	2.29 \pm 0.26	2.15 \pm 0.26	2.38 \pm 0.38
Esinophils %	1.07 \pm 0.04	1.06 \pm 0.15	1.06 \pm 0.17	1.16 \pm 0.17	1.10 \pm 0.15	1.10 \pm 0.11

Table 2: Pre and post-operative mean \pm SE values of Serum Biochemical parameters of dogs subjected to ESIN

Days	Before	After	7th day	14th day	28th day	45th day
Alanine amino transferase (IU/ L)	32.66 \pm 3.56	33.33 \pm 3.43	31.83 \pm 1.13	28.16 \pm 2.62	28.16 \pm 2.50	29.00 \pm 1.39
Alkaline Phosphatase (IU/ L)	102.03 \pm 1.07	107.16 \pm 1.34	145.33 \pm 9.10	161.46 \pm 5.56	149.27 \pm 4.41	115.95 \pm 6.57
Serum Creatinine (mg/dL)	0.44 \pm 0.11	0.44 \pm 0.12	0.51 \pm 0.07	0.48 \pm 0.05	0.48 \pm 0.10	0.47 \pm 0.06
Serum Calcium (mg/dL)	9.40 \pm 0.16	9.43 \pm 0.15	9.91 \pm 0.22	10.09 \pm 0.26	10.26 \pm 0.30	9.99 \pm 0.21
Serum Phosphorus (mg/dL)	3.89 \pm 0.05	3.93 \pm 0.09	4.08 \pm 0.14	4.11 \pm 0.12	4.05 \pm 0.04	3.90 \pm 0.12

**Fig 1:** Changes in serum alanine amino transferase and alkaline phosphatase values during the study period**Fig 2:** Changes in serum creatinine, calcium and phosphorus values during the study period

Conclusion

The fracture treatment with ESIN did not cause any significant variation in haemato-biochemical parameters. However, significant elevation in alkaline phosphatase from 7th post-operative day with peak value on 14th post-operative day indicates increased chondroblastic proliferation and deposition of the calcium salts at the fracture site during initial bone healing process and gradual decrease to normal physiological values by 45th post operative day indicates completion of osteosynthesis. Haemato-biochemical findings on different post-operative days helped in knowing the healing of fracture.

Conflict of interest

Authors have no conflict of interest with any one about this manuscript.

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