COMPARATIVE EVALUATION OF SURGICAL MANAGEMENT OF LONG BONE FRACTURE WITH INTERLOCKING NAILING ALONE AND ALONG WITH -TRICALCIUM PHOSPHATE IN DOGS

MANIKANT, SANDEEP SAHARAN*¹, AMIT KUMAR, ANJU POONIA, DEEPAK KUMAR TIWARI and MANEESH SHARMA¹

Department of Veterinary Surgery and Radiology, ¹Department of Veterinary Clinical Complex, Lala Lajpat Rai University of Veterinary and Animal Sciences, Hisar-125 004 Haryana, India

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ABSTRACT

The goal of the current study was to assess the effectiveness of - tricalcium phosphate (-TCP) in healing of bone in dogs with long bone fractures repaired using interlocking nail technique. Twelve dogs regardless of age, breed, or sex were chosen and divided into two groups at random. Six canines were enrolled in group I, and interlocking nailing was done on them while six dogs were included in group II where -TCP was placed over fractured site after the fracture was stabilized with interlocking nailing. Postoperatively, clinical and radiographic examinations were done on days 15^{th} , 30^{th} and 60^{th} . At different times during the study, group II performed better than group I in terms of mean weight bearing and mean bone union scores. The results of the current study indicate that -TCP efficiently speeds up bone healing in dogs who have long bone fracture.

Keywords: -Tricalcium Phosphate, Fracture, Interlocking nailing

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A fracture will always result in pain and suffering, in addition to the injured limb losing its function (Vardhan *et al.*, 2017). Weight-bearing long bone fracture typically results from severe trauma, such as a vehicle accident (Huang *et al.*, 2012). The goal of fracture treatment is to facilitate the patient's earliest ambulation and the fastest healing feasible (Shahar, 2000). Long bone fracture in dogs is repaired using both conservative and operative techniques (Brinker *et al.*, 1994). Compared to repair methods used outside of the bone, the interlocking nail's placement within the medullary canal offers a biomechanical benefit because it is aligned with the construct's neutral axis (Dueland *et al.*, 1996).

Alloplasts, often known as "bone substitute materials," are inert synthetic graft materials that comprise tricalcium phosphate (TCP) and synthetic hydroxyapatite ceramics. These substances only have osteoconduction as their mode of action. They serve as a scaffold for improved bone tissue growth and healing (Liu and Kerns, 2014). Ceramics made of calcium phosphate, which are common in native human bone, have started to be recognised as appropriate biomaterials (Kanazawa *et al.*, 1975). It has been noted that calcium phosphates have osteoconductive and osteoinductive properties, and they support mesenchymal stem cells' osteogenic development (Shih *et al.*, 2014). This ceramic's ability to increase adenosine signalling in phosphate metabolism and provide

MATERIALS AND METHODS

The 12 dogs used in this study were brought to the department for treatment of long bone fractures. They were separated into two groups at random, regardless of their age, breed, sex, or body weight, as shown in Table 1.

- tricalcium phosphate (-TCP): -tricalcium phosphate (-TCP) granules having a particle size range of 355 to 500µ were employed as an osteoconductive material to fill the bone defect at the fracture site. Prior to surgery, all the animals were administered intramuscular injections of prophylactic antibiotic Ceftriaxone @ 25 mg/kg body weight, @ 0.04 mg/kg of Atropine Sulphate, and @ 0.2 mg/kg of Meloxicam as pre-emptive analgesia. After waiting for five minutes, an intramuscular injection

osteoinductive growth factors has been proposed as a way to enhance osteogenesis (Hoppe *et al.*, 2011; Shih *et al.*, 2014). The function of osteoclasts is inhibited by agonists of the Adenosine A2A receptor, which is strongly related to bone metabolism. Adenosine A2A receptor agonists can also activate osteoblasts and attract mesenchymal stem cells to the bone marrow (Lopez *et al.*, 2019). Additionally, it is believed that -TCP has a favourable impact on the expression of the gene for bone morphogenetic protein (BMP-2) (Tang *et al.*, 2017). So, the present study was undertaken to evaluate the effects of Beta-tricalcium phosphate on bone healing along with interlocking nail in canines.

^{*}Corresponding author: saharan007@gmail.com

of a combination of xylazine hydrochloride (1 mg/kg body weight) and ketamine hydrochloride (5 mg/kg body weight) was given. After the swallowing reflex was lost, the patient underwent endotracheal intubation, and isoflurane anaesthesia was maintained for the remainder of the surgery at a flow rate of 1.0-3.0%.

Surgical technique: The femur, tibia, and humerus were approached for interlocking nailing in the current study. Fracture fragments were lifted with a bone hook and held with bone holding forceps once the fracture site had been made visible. A tampon was used to remove the haematoma at the fracture site and detach the muscles' adhesion to the fracture fragments. For the femur, humerus, and tibia in both groups, interlocking nails of suitable diameter were placed normogradely through the medullary cavity. An L-shaped structure with a threaded knob is called an external aiming device (Jig), and it can be used to align screws with holes in nails. Additionally, intraoperative C-Arm use was used to ensure anatomical reduction and the correct pin position.

In group II, 1 cc of sterile beta-tri calcium phosphate bone graft was deposited in a sterile petridish mixed with NSS and administered using a spatula at the fracture gap after anatomical reduction and internal fixation in a similar manner. As per conventional procedure, the skin was closed with silk no. 1 and the muscles with Vicryl no. 1.

Post-operatively meloxicam was advised @ 0.3 mg/kg body weight I/M and Ceftriaxone @ 25 mg/kg body weight I/M for 5 days. After the operation, a modified Robert Jones bandage was put on to support the operated limb, and it was suggested that you walk on a leash for two weeks. After 15 days following surgery, the skin sutures were taken out.

Post-operative evaluation: Clinical and radiographic evaluations were performed at day 0, 15th, 30th and 60th following surgery to assess postoperative bone healing. Weight bearing while standing and walking was evaluated according to the method recommended by Sahu *et al.* (2017) and has been presented in Table 2. In accordance with Lane and Sandhu (1987), radiographic assessments were performed to assess bone formation scores and bone union scores (Table 3), and the stage of bone union (Table 4) was assessed in accordance with Hammer *et al.* (1985). A classification system adopted by Fox (1995) was used to evaluate clinical outcomes based on functional limb usage (Table 5).

Statistical analysis: The statistical analysis of data was done by one-way-ANOVA with linear repeated measure by SPSS software. All the data values were expressed as Mean \pm Standard error of mean (Mean \pm S.E.). P-value less

than 0.05 considered as statistically significant.

RESULTS AND DISCUSSION

In dogs with long bone fractures fixed using interlocking nails, this study was done to determine the

Table 1. Design of the study

Table I.	Desig	gn of the study		
Group		Treatment p	lan	
Group I (1 Group II (n=6) (n=6)	Internal fixa Internal fixa along with	tion using interloc ation using inter -TCP at fracture si	king nailing locking nailing ite
Table 2.	Weig walk	ht bearing sco ing (Sahu <i>et al</i>	oring system whil (., 2017)	e standing and
Score	Desc	cription		
	V	Weight bearin	g while standing	
0	Carr	ying the limb o	ff the ground	
1	Touc	ching the toe on	the ground	
2	Touc	ching the paw o	n the ground	
3	Full	weight bearing	5	
		Weight bearin	ng while walking	
0	Carr	ying the limb o	ff the ground	
1	Occa	asional touchin	g of toe/paw on ea	ch step
2	Freq	uent touching of	of toe/paw on each	step
3	Touc	ching the toe on	every step	
4	Touc	ching the paw o	n every step	
Table 3.	Radi 1987)	ographic scor)	ing system (Lan	e and Sandhu,
Score	Desc	cription		
		Bone Forn	nation Scores	
0	Noe	vidence of bon	e formation	
1	Bone	e formation in 2	25% of the gap	
2	Bone	e formation in 5	50% of the gap	
3	Bone	e formation in 7	75% of the gap	
4	Bone	e formation in 1	00% of the gap	
		Bone Ur	ion Scores	
0	With	complete frac	ture trace	
2	With	i incomplete fra	acture trace	
4	Abse	ence of fracture	trace	
Table 4.	Stage (Han	e of bone unio nmer <i>et al.</i> , 198	on radiographic s 35)	scoring system
Grade C	allus Fo	ormation	Fracture	Stage of

Grade	Callus Formation	Fracture Line	Stage of Union
1	Homogenous bone structure	Obliterated	Achieved
2	Massive-Bone trabeculae crossing the fracture line	Achieved	Barely discernible
3	Apparent-Bridging of fracture line	Discernible	Uncertain
4	Trace-No bridging of fracture line	Distinct	Not Achieved
5	No callus formation	Distinct	Not Achieved



Fig. 1. Internal fixation of fracture using, A) Interlocking nailing, B) Application of -TCP



Fig. 2. Radiographs showing fracture healing in group I (upper row) and group II (lower row) Immediately after surgery, B. On 15th day, C. On 30th day, D. On 60th day)





Fig. 3. Weight bearing while standing in group I (upper row) and group II (lower row) (A. On 15th day, B. On 30th day, C. On 60th day)

effectiveness of -tricalcium phosphate (-TCP) in promoting bone healing. At various time intervals, the bone formation scores (BFS) in group II were not significantly higher than those in group I (Table 6). Similar to this, group II's bone union scores (BUS) were not significantly higher than group I's (Table 7). Group II has reached the stage of union earlier than Group I (Table 8 and Fig. 2). The administration of the osteoconductive substance, -TCP, at the fracture site has resulted in an

Table 5. Functional limb usage assessment (Fox et al.)			
Grade	Description		
	Functional limb usage		
Excellent	Weight bearing without lameness		
Good	Slight Lameness		
Fair	Slight to moderate lameness principally after exercise		
Poor	Intermittent or consistent non-weight bearing lameness		

Table 6.	Bone formation scores at different time intervals in both the groups (Mean±S.E.)
14010 00	bone for mation scores at anner ent time meet vals in soun the groups (iftean-size)

Time interval	0 th day	15^{th} day	30 th day	60 th day	
Group I	$0.00^{a} \pm 0.00$	0.67 ^b ±0.21	1.67°±0.21	$3.00^{d} \pm 0.26$	
Group II	$0.00^{a} \pm 0.00$	$0.83^{b} \pm 0.31$	$2.00^{\circ}\pm0.26$	3.33 ^d ±0.33	

(Means with different superscripts (a, b)/(A, B) varies significantly (p<0.05) within group/ between the group)

 Table 7.
 Bone union scores at different intervals in both the groups (Mean±S.E.)

Time interval	0 th day	15 th day	30 th day	60 th day	
Group I	$0.00^{a} \pm 0.00$	$0.67^{ab}\pm 0.42$	$2.00^{cd} \pm 0.00$	$0.67^{d} \pm 0.42$	
Group II	$0.00^{a} \pm 0.00$	$0.67^{b}\pm 0.42$	$2.00^{abc} \pm 0.00$	$3.00^{c} \pm 0.45$	

(Means with different superscripts (a, b) varies significantly (p<0.05) within group)

Table 8. Stages of bone union at different time intervals in group I and group II

Group I	0 th day	15 th day	30 th day	60 th day
IA	NotAchieved	Not Achieved	Uncertain	Achieved
IB	Not Achieved	Not Achieved	Uncertain	Uncertain
IC	NotAchieved	Not Achieved	NotAchieved	Uncertain
ID	NotAchieved	Uncertain	Uncertain	Achieved
IE	NotAchieved	Not Achieved	Uncertain	Achieved
IF	Not Achieved	Not Achieved	Uncertain	Uncertain
Group II				
IIA	Not Achieved	Uncertain	Achieved	Achieved
IIB	Not Achieved	Not Achieved	Uncertain	Uncertain
IIC	NotAchieved	Uncertain	Uncertain	Achieved
IID	Not Achieved	Not Achieved	NotAchieved	Uncertain
IIE	NotAchieved	Uncertain	Achieved	Achieved
IIF	NotAchieved	Uncertain	Achieved	Achieved

increase in the rate of bone formation, which has led to an increase in the mean bone union scores in group II. The osteoconductive properties of -TCP were also described by Goel *et al.* (2013) and Preethi *et al.* (2021), demonstrating that it was a secure and efficient therapeutic choice for

fracture repair in significant osteo-periosteal abnormalities.

The mean weight bearing score when standing (Table 9) gradually increased over time in both groups, moving from 0.00 ± 0.00 on day 0 to 2.67 ± 0.21 on day 60^{th} in group I and 0.00 ± 0.00 to 2.83 ± 0.17 on day 60^{th} in group

 Table 9.
 Weight bearing scores while standing at different time intervals in both the groups (Mean ± S.E.)

0	0	0	0		
Group	0 th day	15 th day	30 th day	60 th day	
Group I	$0.00^{a} \pm 0.00$	$0.67^{b}\pm 0.21$	1.50°±0.22	$2.67^{d} \pm 0.21$	
Group II	$0.00^{a} \pm 0.00$	0.83 ^b ±0.17	1.67°±0.21	$2.83^{d} \pm 0.17$	
(Means with diffe	erent superscripts (a, b)/(A	, B) varies significantly	(p<0.05) within group/betw	veen group, respectively)	
Table 10. Weigh	t bearing scores while wa	alking at different time	intervals in both the grou	ps (Mean±S.E.)	
Group	0 th day	15^{th} day	30 th day	60 th day	
Group I	$0.00^{a} \pm 0.00$	0.83ª±0.40	2.00 ^b ±0.52	3.17°±0.48	
Group II	$0.00^{a} \pm 0.00$	1.50 ^b ±0.43	2.50 ^b ±0.43	3.50°±0.34	
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(Means with different superscripts (a, b) varies significantly (p < 0.05) within group)

II, respectively (Fig. 3). For the entire post-operative period, group II's mean weight bearing scores were higher. Also, on the day of the presentation, all animals in both groups displayed non-weight bearing lameness. Because a fracture results in pain in the affected area from inflammation and injury to the surrounding muscles, animals often lift the injured limb off the ground (Gupta, 2015). On the fifteenth day, group II's weight bearing scores non-significantly increased in comparison to group I. In addition, all of the animals in group II were observed contacting the toe or paw of the ground, but most of the animals in group I were carrying the limb off the ground. On the 30th day, the majority of the animals were discovered touching a toe or paw on the ground, showing that group I's weight bearing score had improved, whereas in group II, majority of the animals only began fully bearing weight on the injured limb on the 30th day. On the 60th postoperative day, animals from both groups displayed full weight bearing.

Similarly, the weight bearing scores while walking (Table 10) was non-significantly higher in group II as compared to group I at different intervals. Singh *et al.* (2020) noted related results as well. The early and better fracture healing in dogs of group II, which was supported by radiographic scores and was probably connected to local administration of -TCP at the fracture site, was the cause of the improvement in the weight bearing scores (when standing and walking). The group that received osseomold (DMB and Calcium sulphate hemihydrate) and autogenous cancellous bone graft at the fracture site had the lowest mean lameness scores, which was a sign of early fracture healing, according to Kumar (2020).

In group I, there was one case where the functional use of the limb was excellent, three cases where it was good, one case where it was fair, and one case where it was bad. In group II, three examples were excellent, two were good, and one was fair. The timing of the recovery of limb functions with complete range of motion in group II animals who had early callus development and weight bearing is consistent with this finding. Singh *et al.* (2020) also reported similar results.

Due to its osteoconductive properties and improved *in vivo* degradation, -tricalcium phosphate (-TCP) has emerged as a viable material for applications involving bone regeneration. Through controlling osteogenic processes like the differentiation of mesenchymal stem cells into osteoblasts, the development of new blood vessels, the release of angiogenic growth factors, and blood clot formation, -TCP promote bone regeneration (Lu *et al.*, 2021). In the group of animals receiving -TCP treatment, the weight bearing scores, bone union scores, and functional limb usage were all improved. Thus, -TCP helps dogs heal faster. As a result, it is determined that interlocking nailing in -TCP is superior to interlocking nailing alone for the treatment of long bone fractures in dogs.

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