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Effects of autologous bone marrow on the healing of long bones fractures reduced by external skeletal fixators in goats

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Abstract

This study aimed to assess the effect of autologous bone marrow on metacarpal and metatarsal fractures in goats presented to the indoor surgery clinic at the University of Veterinary and Animal Sciences (UVAS) in Lahore, Pakistan. A total of 20 female Beetal goats weighing 20-22.5 kg and aged 8-12 months were allocated into equal groups of five animals each: G1, G2, G3, and G4. Animals in G1 were fixed with a linear external skeletal fixator, and animals in G3 were fixed with a circular fixator and treated with multiple aspirates from the bone marrow, while animals in G2 were fixed the same way as G1, and animals in G4 were fixed the same way as G3 and treated with normal saline on days 0, 14, and 28. The radiographic union scale (RUS) and weight-bearing score (WBS) were used to evaluate the post-treatment rate of repair, and sero-biochemical changes were observed on days 0, 7, 14, 28, and 45. The results showed a significant difference ($P<0.05$) in bone healing among groups treated with bone marrow aspirates (BMA) and groups treated with normal saline. RUS and WBS scoring in G1 and G3 animals were significantly higher ($P<0.05$) than in G2 and G4 animals on days 7, 14, and 28. Moreover, animals in G1 and G3 had significantly ($P<0.05$) greater levels of alkaline phosphatase, total hydroxyproline, and free hydroxyproline than animals in G2 and G4, respectively. In conclusion, the application of BMA at the site of fracture aids in the healing of fractured metacarpal and metatarsal bones aligned with external (linear and circular) fixators in goats.

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Introduction

Goats are widely recognized as animal models for orthopedic research (1) due to their docile nature, low cost, availability, ease of handling and housing, and compliance (2). Long bone fractures are one of the key orthopedic conditions encountered in small ruminants (3). The use of external skeletal fixators for treating long bone fractures is a common practice in humans, and due to the benefits of this intervention, researchers are being compelled to find new ways to treat fractures in veterinary surgery. External fixators can be easily installed without the use of radiographs (4). Fractures reduced by external fixators have several advantages, including immediate support by the treated limb in the animal, fewer chances of infection, less postoperative pain, less implantation damage, and free mobility of the joint, which contributes to a greater degree of movement and thus enhances self-healing processes (5). The healing of a fracture depends on successful callus formation; therefore, many studies that concern fracture healing focus on alternative bone matrices (6). However, alternatives to this have not been accomplished due to several reasons, with the histochemical effects on the host tissue being one of the major reasons (7). A reasonable approach to inducing progenitor cells has been recommended when applying many orthopedic settings in animals (8). Bone marrow aspirate (BMA) is a reasonable source of progenitor cells for skeletal tissue (9). Bone marrow (BM) is supposed to promote osteogenesis in bones during fracture repair and healing (10). The healing of a fracture can be assessed clinically, radiographically, and by bone turnover markers (11). The bone turnover markers are proteins in nature that show bone absorption (12) and are distributed among collagenous markers for bone development (13). Efforts are being made the development of new technologies to enhance fracture repair in animals (14).

To the best of our knowledge, no studies on fracture treatment of the metacarpal and metatarsal bones in goats using external skeletal fixation in conjunction with BMA application have been conducted in Pakistan. Therefore, this is the first study to evaluate the effects of autologous BMA, along with external (linear and circular) fixators, on fracture treatment of the metacarpal and metatarsal bones in goats. It was hypothesized that autologous BMA, along with external (linear and circular) fixators, could be an effective fracture treatment in goats for metacarpal and metatarsal fractures.

Materials and methods

Ethical Approve

This study and all techniques were conducted following the rules and regulations of the Ethical Review Committee (Ethical Approval No. DR/27; Dated: 01/10/2019) at the Department of Veterinary Surgery and Pet Sciences, University of Veterinary and Animal Sciences, Lahore, Pakistan.

Experimental design and treatments

A total of 20 clinical cases of long-bone metacarpal and metatarsal fractures in Beetal goats were selected for the study. These cases were presented at the University of Veterinary and Animal Sciences (UVAS), Lahore. All the selected animals were female, with an average body weight of 20-22.5 kg and an age of 8-12 months. All animals were brought in on the same day of metacarpal and metatarsal fracture occurrence. The animals with metatarsal and metacarpal fractures were divided into 4 groups, with 5 animals in each group. The first treatment group had fracture fixation using an external linear fixator, and BMA was injected at the site of the fracture line. The second treatment group had fracture fixation using an external linear fixator, and normal saline (NS) was injected at the site of the fracture. In the third treatment group, fracture fixation was done using an external circular fixator, and BMA was inserted at the fracture line. In the fourth treatment group, fracture fixation was done using an external circular fixator, and NS was injected at the fracture line. All the techniques used in this study were conducted under the rules and regulations of the Ethical Review Committee at the Department of Veterinary Surgery and Pet Sciences, University of Veterinary and Animal Sciences, Lahore, Pakistan.

Bone marrow aspirate

Animals were placed in lateral recumbency with the donor limb placed upward. The humerus was brought forward cranially, and the elbow was rotated medially. A notch was made between the greater tubercle and the head of the humerus. Using a sternal needle, a puncture was made by slowly twisting the needle clockwise and anticlockwise. The needle was fitted with a 10 mL syringe and inserted into the marrow. After the aspiration of 3 mL of bone marrow, the bone marrow aspirate was centrifuged at 3200 rpm for 15 minutes. The BMA was then injected into the fracture sites and subcutaneously on the 14th and 28th days post-surgery (15).

Preoperative preparation and sedation

Animals selected for surgery fasted for 18-20 hours before surgery. The surgical site was prepared aseptically by clipping, shaving, and scrubbing with a 5% povidone-iodine antiseptic solution, and isopropyl alcohol was used as a degreasing agent. In all treatment groups, animals were sedated with diazepam (Domosedan injection, Orion Corporation, Finland) at a dosage of 0.25 mg per kg of body weight. After sedation, induction was performed using a combination of propofol and ketamine (Fresofol 1% injection, Fresenius Kabi, Austria). Anesthesia was maintained with a constant rate of infusion using a volumetric syringe-driving pump with the combination of diazepam at a rate of 0.02 mg/kg per hour, ketamine at a rate of 0.04 mg/kg per minute, and propofol at a rate of 0.016 mg/kg per minute.

Fracture repair

A close surgical reduction of the fracture was performed in all cases. External skeletal fixation was utilized as the treatment method for metacarpal or metatarsal fractures. For transverse fractures, a linear external skeletal fixator was employed, while a circular external skeletal fixator was used for comminuted fractures in the selected animals. In group G1, a linear external skeletal fixator was applied using a closed approach in all cases (Figure 1). External manipulation of the fracture was performed to align and oppose the bone, and autologous BMA was injected at the site of the fracture. An orthopedic drill (less than 300 rpm) was used to percutaneously drill the pins into each bone. Two pins of 3 mm were inserted in the proximal and distal fragments of the fractured bone, which were then connected using two connecting bars of 3.5 mm on both sides. The bars were secured with stainless steel clamps. The pins were first inserted at the upper and most distal parts of the bone. The clamps were then connected to the connecting bars, and fracture stability was ensured by tightening the clamps and verifying that the fractured fragments were correctly aligned and opposed. The same surgical procedure as described above was performed in group G2, but with the inoculation of NS instead of BMA at the fracture site.

In group G3, following fracture reduction, one ring of a fixator was introduced at the proximal fragment of the fractured bone, and one ring was introduced at the distal portion (Figure 1). All the rings were made of aluminum and had a diameter of 10 cm. Kirschner pins of 2 mm in diameter were used in all cases, with each ring being fixed with two pins. A 4 mm skin incision was made for the placement of pins using forceps. A blunt dissection of the soft tissue was performed, and a drilling sleeve was used to protect the neurovascular and tendinous structures. Trans-fixation bits were placed around 2 cm distal to the joint associated with the fracture. The boundaries between the pin and skin were washed with a 0.5% to 20% solution of chlorhexidine gluconate, and a gauze soaked with the solution was applied between the skin, fixator, and pin tracts. The dressings were replaced after 24 hours. The same surgical procedure as described for group G3 was performed in group G4, but with NS instead of BMA at the fracture site.

Blood sampling and biochemical evaluation

Blood samples were collected from the jugular vein of goats, approximately 5 mL on day 0 before surgery and thereafter on days 7, 14, 28, and 45 post-surgeries. The blood was collected in sodium citrate vacutainer tubes to prevent coagulation and then transferred to the Diagnostic Laboratory at the University of Veterinary and Animal Sciences in Lahore. The tubes were centrifuged at 3000 rpm for 15 minutes at 4°C using a temperature-controlled centrifuge machine (HARRIER 18/80). The serum was separated and stored at -20°C for further procedures. Samples for the analysis of calcium (Ca), phosphorus (P), alkaline phosphatase (ALKP), free hydroxyproline (FHP), and total hydroxyproline (THP) were stored at -20°C until further analyses. Samples for FHP and THP were analyzed with an automatic analyzer (Biosystems S.A. Costa Brava, Barcelona, Spain), while samples for Ca (MTD Diagnostics, IVD, Italy), P (Spectrum, SAE, Cairo, Egypt), and ALKP (Sigma Aldrich® St. Louis, Missouri, USA) were analyzed using serum diagnostic kits.

Clinical evaluating parameters

Weight-bearing and radiographic union scoring were performed to assess the severity of lameness and the degree of recovery, respectively. Weight-bearing grades of the affected limb were determined as follows according to Pierson (2002): 0 - full weight bearing, 1 - weight bearing as tolerated, 2 - partial weight bearing, 3 - touch-down weight bearing, and 4 - no weight bearing (16). To assess the degree of recovery, radiographs of fractured bones were obtained and graded according to the scoring system designated by Lane and Sandhu (17): 0 points for no evidence of callus formation, 1 point for callus formation occupying 25% of the gap, 2 points for callus formation occupying 50% of the gap, 3 points for callus formation occupying 75% of the gap, and 4 points for callus formation occupying the full gap formation (17).

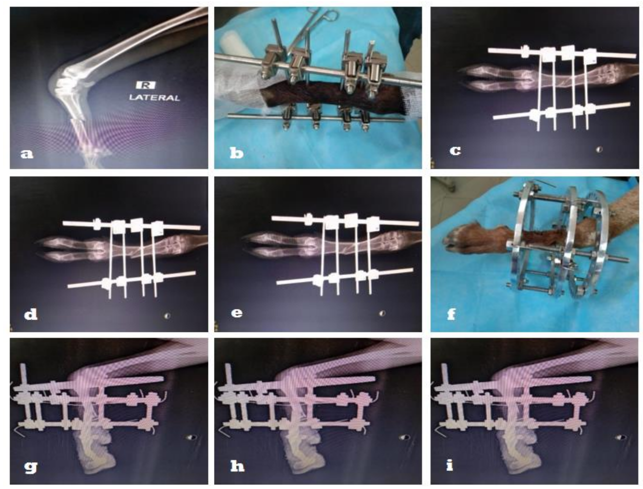

Figure 1: Clinical photograph (a) and lateral radiograph of complete metacarpal fracture in goat; Immediate postoperative lateral radiographs of metacarpal bone; (b) linear skeletal fixator; (c) Linear skeletal fixator on day 0; (d) radiographs for metacarpal bone on day 14, postoperative radiograph for linear fixator + bone marrow; (e) radiographs for metacarpal bone on day 45, postoperative radiograph for linear fixator + bone marrow; (f) circular skeletal fixator; (g) circular skeletal fixator at day 0; (h) radiographs for metacarpal bone on day 14), postoperative radiograph for circular fixator + bone marrow; (i) radiographs for metacarpal bone on day 45, postoperative radiograph for circular fixator + bone marrow

Statistical analysis

The statistical analyses were conducted using IBM SPSS version 20 (IBM Corp., Armonk, NY, USA). Quantitative data were summarized using mean±SD or median and range, while qualitative data were organized using frequencies and percentages. The mean differences of continuous variables between the groups (G1, G2, G3, and G4) were compared using analysis of variance (ANOVA), while the median differences were compared using the Mann-Whitney U test. A p-value > 0.05 indicated no significant difference, while a p-value < 0.05 indicated a significant difference.

Results

Weight-bearing Scoring

The WBS was observed to be the same among the groups at day 0, while at days 7 and 14, there was a statistically significant difference (P<0.05). The results for the remaining days showed similar WBS values (Table 1).

Table 1: Weight-bearing score of animals treated with linear and circular external fixators along with bone marrow aspirate and normal saline

Time	Treatments Median (Range)				
	G1	G2	G3	G4	p-value
Day-0	4 (3 - 4)	4 (3 - 4)	4 (3 - 4)	4 (4 - 4)	1.000
Day-7	3 (3 - 4)	4 (3 - 4)	2 (2 - 3)	3 (2 - 3)	0.009
Day-14	2 (2 - 3)	3 (2 - 3)	1 (1 - 2)	2(1 - 2)	0.002
Day-28	1 (1 - 2)	2 (1 - 2)	1 (0 - 1)	1 (1 - 2)	0.086
Day-45	0 (0 - 1)	1 (0 - 1)	0 (0 - 1)	1 (1 - 2)	0.066

G1: Animals treated with a linear external fixator and bone marrow aspirate; G2: Animals treated with a linear external fixator and normal saline; G3: Animals treated with a circular external fixator and bone marrow aspirate; G4: Animals treated with a linear external fixator and normal saline; differences were considered significant at P<0.05.

Radiographic scoring

The radiographic scoring results for the goats showed that there were improved RUS outcomes at days 7, 28, and 45 in animals treated with BMA compared to those treated with NS (P<0.05). The results for the remaining days showed similar RUS values (Table 2).

Table 2: Radiographic union score of animals treated with linear and circular external fixators along with bone marrow aspirate and normal saline

Time	Treatments Median (Range)				
	G1	G2	G3	G4	p-value
Day-0	0 (0 - 0)	0 (0 - 0)	0 (0 - 0)	0 (0 - 0)	---
Day-7	1 (0 - 0)	0 (0 - 0)	1 (0 - 0)	0 (0 - 0)	0.040
Day-14	2 (1 - 2)	1 (1 - 2)	2 (1 - 2)	1 (1 - 1)	0.040
Day-28	3 (2 - 3)	2 (1 - 2)	3 (2 - 3)	2 (1 - 1)	0.048
Day-45	4 (3 - 4)	3 (2 - 4)	4 (3 - 4)	3 (3 - 4)	0.143

G1: Animals treated with a linear external fixator and bone marrow aspirate; G2: Animals treated with a linear external fixator and normal saline; G3: Animals treated with a circular external fixator and bone marrow aspirate; G4: Animals treated with a linear external fixator and normal saline; differences were considered significant at P<0.05.

Blood parameters

There was no statistically significant difference in the sero-biochemical findings regarding phosphorus and calcium (p > 0.05). However, ALKP was significantly higher (P<0.05) in the BMA groups on days 7, 14, and 28 compared to the NS groups on days 14, 28, and 45 (P<0.05). FHP and THP were significantly higher (P<0.05) in the BMA groups on days 7, 14, 28, and 45 (Table 3).

Discussion

Efforts are being made to develop new techniques for accurate diagnosis of complications of bone fractures and bone healing (18). Progressive improvement in weight-bearing was noticed for all the animals during the observation period. Weight-bearing was the same among the groups on day 0, while on days 7 and 14, there was a statistical difference among the groups. The results of the present analysis indicated that groups G1 and G3 showed better healing compared to groups G2 and G4. Due to the positive effect of BMA, which contains osteogenic cells and osteo-conductive factors that enhance the preparation of an inorganic scaffold and provide an easy regenerative system (19). This demonstrates the positive osteogenic properties of BMA (11).

The RUS showed improved outcomes on days 7, 28, and 45 in animals treated with BMA compared to those treated with NS. The results on the remaining days were similar regarding RUS. The results indicate that groups G1 and G3 showed better callus formation compared to groups G2 and G4. Similar results were noticed by (11), which are due to the positive effect of BMA on the radiographic characteristics of newly produced calluses at the fracture line. This positive effect is due to the delivery of connective tissue at concentrations higher than those naturally present in BM, which results in a great increase in the degree of bone combination capacity and machine-driven toughness of fractures (20).

No significance was observed regarding serum calcium among the groups. However, an increasing trend in serum calcium levels up to day 28 was observed. Increased levels of serum calcium in the initial stages are recognized to increase osteoclastic activity. Using bone plates for the reduction of bone fractures in goats and also observed a rise in serum calcium levels (21). Umarani and Ganesh observed a decrease in calcium level on the seventh day after the reduction of a femur fracture in goats, followed by a significant increase in concentration from day 15 to reach the normal level by day 60 of healing (22). The initial increasing trend of serum calcium levels is recognized to increase osteoclastic bone resorption during the initial stages of fracture healing and during the level of remodeling, which results in calcium mobilization into blood (23). There was an increasing trend in serum phosphorous levels. The elevation of serum phosphorous levels after the fixation of long bone fractures in goats (24). The increase in serum phosphorous is attributed to osteoclastic activity leading to the resorption of dead bone, thus increasing the levels of phosphorous (23).

Table 3: Sero-biochemical findings of animals treated with linear and circular external fixators along with bone marrow aspirate and normal saline

Parameter	Days	Treatments				
		G1	G2	G3	G4	p-value
Phosphorus (mg/dl)	0	6.01±1.63	6.61±1.083	6.50±1.77	7.38±2.20	0.662
	7	6.00±1.82	8.02±1.24	6.34±1.60	5.56±1.95	0.149
	14	6.52±1.04	7.88±1.14	6.96±0.98	6.82±1.79	0.402
	28	5.88±1.14	7.50±1.01	6.28±1.36	6.72±1.50	0.255
	45	5.75±1.17	6.62±0.88	7.36±3.17	7.50±1.40	0.459
Calcium (mg/dl)	0	8.40±1.140	8.80±1.30	8.40±0.89	8.80±1.30	0.899
	7	10.00±0.70	10.00±1.00	10.60±1.14	10.20±1.30	0.786
	14	11.60±1.51	11.40±0.54	10.80±1.30	11.20±0.83	0.710
	28	11.80±1.09	12.60±0.54	11.20±0.83	12.20±0.83	0.111
	45	9.60±1.14	9.20±0.83	9.40±0.89	9.60±1.14	0.909
ALKP (IU/L)	0	88.58±11.60	119.72±34.93	86.74±10.13	99.96±39.82	0.250
	7	176.11±30.43	161.29±31.34	227.36±43.45	138.85±51.15	0.019
	14	288.26±51.66	221.44±36.15	317.09±44.92	212.71±53.98	0.007
	28	310.16±63.06	212.49±35.09	371.64±52.13	272 51±64.38	0.003
	45	304.81±59.48	273.15±72.54	350.70±44.61	273.52±55.50	0.166
FHP (mg/day)	0	8.42±0.03	8.16±0.17	8.49±0.02	8.46±0.02	0.001
	7	10.62±0.42	9.72±0.42	10.08±0.55	9.67±0.44	0.019
	14	11.78±0.97	11.02±0.23	11.30±0.37	10.86±0.06	0.073
	28	12.39±0.32	11.18±0.33	12.23±0.61	11.18±0.33	0.001
	45	13.45±0.34	12.56±0.32	13.60±0.40	12.48±0.50	0.001
THP (mg/day)	0	10.42±0.04	10.29±0.27	10.46±0.05	10.46±0.03	0.240
	7	11.62±0.42	10.51±0.53	11.05±0.56	10.42±0.02	0.002
	14	12.70±0.46	11.67±0.48	12.38±0.77	11.41±0.13	0.004
	28	14.23±0.38	12.48±0.43	14.27±0.72	12.69±0.75	0.001
	45	15.19±0.42	13.63±0.45	14.27±0.72	12.69±0.75	0.001

G1: Animals treated with a linear external fixator and bone marrow aspirate; G2: Animals treated with a linear external fixator and normal saline; G3: Animals treated with a circular external fixator and bone marrow aspirate; G4: Animals treated with a linear external fixator and normal saline; differences were considered significant at $P<0.05$.

Alkaline phosphatase had significantly higher values among the groups (G1, G2, G3, and G4), while FHP was observed to be higher in group G1 and group G3 compared to other groups, G2 and G4. Similar patterns were observed for THP. Scholar Manjulkar also observed similar biochemical changes during the healing of long bone fractures in goats, along with an increased level of serum ALKP (25). The rise in ALKP levels is due to increased osteoblastic activity as osteoblasts release a large quantity of ALKP that participates in the formation of bone matrix. Bone mineralization is also due to the property of BMA to induce osteoblastic proliferation and a rise in the activity of the alkaline phosphatase enzyme (26). Further studies have also mentioned an increased level of urinary excretion of FHP and THP following fractures compared to healthy individuals (27). This is because the definite phase of resorption lasts for 50 days due to the elevation of the residue as a result of continuing collagen synthesis (28).

Hydroxyproline is mainly present in collagen fibers and comprises almost 13% of an amino acid (29). It is derived from proline and is formed as a result of hydroxylation after translation that occurs in the peptide chain. Due to the degradation of collagen, FHP is released, which cannot be reutilized for the synthesis of collagen (30). Furthermore, about 50% of collagen exists in bone, where its turnover can be faster than that of soft tissues, which are eliminated in the urine. Therefore, it is considered a marker of bone resorption (31). Further studies have also observed increased levels of urinary FHP and THP following fractures compared to healthy animals (27). The measurement of sero-biochemical markers for bone healing during the process of fracture healing could augment the accuracy of the assessment of the bone healing stage (32). The results of this study are associated with the stimulation of osteoblasts, which play a vital role in the vigorous synthesis and maturation of the extracellular matrix of bone during the process of fracture healing (11).

Conclusion

In conclusion, the use of bone marrow aspirate shows promising osteogenic effects on the repair of metacarpal and metatarsal fractures in goats, especially when combined with external fixation.

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Conflict of interest

All authors have declared no conflicts of interest.

References

1. Grzeskowiak RM, Rifkin RE, Croy EG, Steiner RC, Seddighi R, Mulon PY, Adair HS, Anderson DE. Temporal changes in reverse torque of locking-head screws used in the locking plate in segmental tibial defect in goat model. Front Surg. 2021;8:637268. DOI: 10.3389/fsurg.2021.637268
2. Cachon T, Hassoun R, Odet M, Massenzio M, Carozzo C, Ronel S, Lafon Y, Viguier E. Morphometric dimensions of the goat thoracolumbar vertebrae using digitized CT images. Comput Methods Biomech Biomed Eng. 2019;22(I):S123-5. DOI: 10.1080/10255842.2020.1713505
3. Basith MA, Sreenu M, Chandra Sekhar EL, Rajendranath N. Management of long bone fractures by free-form external skeletal fixation using epoxy putty in sheep and goat. J Livstock Sci. 2018;9(1):51-5. [available at]
4. Nagar S, Singh R, Shahi A, Vaish R. External fixation using acrylic versus epoxy as a connecting bar for repair of compound fracture in goat. J Anim Res. 2019;9(1):147-51. DOI: 10.30954/2277-940X.01.2019.20
5. Hadeed MM, Evans CL, Werner BC, Novicoff WM, Weiss DB. Does external fixator pin site distance from definitive implant affect infection rate in pilon fractures?. Injury. 2019;50(2):503-7. DOI: 10.1016/j.injury.2018.10.041
6. Doijode V. Internal fixation in goats for long bone fracture repair with low cost veterinary cuttable plate. J Pharm Innov. 2018;7(8):538-42. [available at]
7. Zhao R, Yang R, Cooper PR, Khurshid Z, Shavandi A, Ratnayake J. Bone grafts and substitutes in dentistry: A review of current trends and developments. Mol. 2021;26(10):3007. DOI: 10.3390/molecules26103007
8. Jiang M, Liu R, Liu L, Kot A, Liu X, Xiao W, Jia J, Li Y, Lam KS, Yao W. Identification of osteogenic progenitor cell-targeted peptides that augment bone formation. Nat Commun. 2020;11(1):1-4. DOI: 10.1038/s41467-020-17417-9
9. Lana JF, da Fonseca LF, Azzini G, Santos G, Braga M, Cardoso Junior AM, Murrell WD, Gobbi A, Purita J, Percope de Andrade MA. Bone marrow aspirate matrix: A convenient ally in regenerative medicine. Int J Mol Sci. 2021;22(5):2762. DOI: 10.3390/ijms22052762
10. Kassem MS. Percutaneous autogenous bone marrow injection for delayed union or non-union of fractures after internal fixation. Acta Orthop Belg. 2013;79(6):711-7. [available at]
11. Al-Sobayil F, Sadan MA, El-Shafaey ES, Ahmed AF. Can bone marrow aspirate improve mandibular fracture repair in camels (*Camelus dromedarius*)? A preliminary study. J Vet Sci. 2020;21(6). DOI: 10.4142/jvs.2020.21.e90
12. Sousa CP, Lopez-Peña M, Guzón FM, De Abreu HV, Luís MR, Viegas CA, Camassa J, De Azevedo JT, Cabrita AS, Reis RL, Gomes ME. Evaluation of bone turnover markers and serum minerals variations for predicting fracture healing versus non-union processes in adult sheep as a model for orthopedic research. Injury. 2017;48(8):1768-75. DOI: 10.1016/j.injury.2017.05.025
13. Coelho CA, Bordelo JP, Camassa JA, Barros VA, Babo PS, Gomes ME, Reis RL, Azevedo JT, Requicha JF, Faisca P, Carvalho PP. Evaluation of hematology, general serum biochemistry, bone turnover markers, and bone marrow cytology in a glucocorticoid-treated ovariectomized sheep model for osteoporosis research. An Acad Bras Cienc. 2020;92(4). DOI: 10.1590/0001-3765202020200435
14. Cox G, Einhorn TA, Tzioupis C, Giannoudis PV. Bone-turnover markers in fracture healing. J Bone Joint Surg. 2010;92(3):329-34. DOI: 10.1302/0301-620X.92B3.22787
15. Theophilus ON, Innocent N, Nnenna U, Chioma O. Evaluation of single and multiple use of bone marrow aspirate in management of tibia fractures of Nigerian indigenous dogs, J Appl Anim Res. 2018;46:828-834. DOI: 10.1080/09712119.2017.1408466
16. Pierson F. Principles and techniques of patient care. 3rd ed. Philadelphia: WB Saunders Company; 2002. 208 p.
17. Lane JM, Sandhu HS. Current approaches to experimental bone grafting. Orthop Clin N Am. 1987;18(2):213-225. DOI: 10.1016/s0030-5898(20)30385-0
18. Sun Y, Helmholz H, Willumeit-Roemer R. Preclinical in vivo research of magnesium-based implants for fracture treatment: A systematic review of animal model selection and study design. J Magnes Alloy. 2021;9(2):351-361. DOI: 10.1016/j.jma.2020.09.011
19. Elkhenany H, El-Badri N, Dhar M. Green propolis extract promotes in vitro proliferation, differentiation, and migration of bone marrow stromal cells. Biomed Pharmacotherap. 2019;115:108861. DOI: 10.1016/j.biopha.2019.108861
20. Iaquinta MR, Mazzoni E, Manfrini M, D'Agostino A, Trevisiol L, Nocini R, Trombelli L, Barbanti-Brodano G, Martini F, Tognon M. Innovative biomaterials for bone regrowth. Int J Mol Sci. 2019;20(3):618. DOI: 10.3390/ijms20030618
21. Kumar D. Efficacy of bone substitutes for fracture healing in goats [Ph.D. dissertation]. India: Nanaji Deshmukh Veterinary Science University, Jabalpur; 2016.
22. Uma Rani R, Ganesh TN. Study of serum calcium, phosphorus, and alkaline phosphatase during fracture healing of femur in goats. Indian Vet J. 2003;80(4):377-8. [available at]
23. Venugopalan A. Essentials of veterinary surgery. 8th ed. India: Oxford and IBH Publishing Co; 2009. 166-173 p.
24. Daron J. Comparision of fiber glass with plaster cast for treatment of long bone fracture in goats [master's thesis]. India: Karnataka Veterinary Animal and Fisheries Science University, Bidar; 2013.
25. Manjulkar GP. Effect of various immobilizations with special reference to injection placentrex for healing of fracture in caprine [master's thesis]. India: Dr. Panjabrao Deshmukh Krishi Vidyapeeth, Akola; 2000.
26. Zhang X, Hirai M, Cantero S, Ciubotariu R, Dobrila L, Hirsh A, Igura K, Satoh H, Yokomi I, Nishimura T, Yamaguchi S. Isolation and characterization of mesenchymal stem cells from human umbilical cord blood: Reevaluation of critical factors for successful isolation and high ability to proliferate and differentiate to chondrocytes as compared to mesenchymal stem cells from bone marrow and adipose tissue. J Cell Biochem. 2011;112(4):1206-18. DOI: 10.1002/jcb.23042
27. Veronesi F, Giavaresi G, Tschon M, Borsari V, Nicoli Aldini N, Fini M. Clinical use of bone marrow, bone marrow concentrate, and expanded bone marrow mesenchymal stem cells in cartilage disease. Stem Cells Dev. 2013;22(2):181-192. DOI: 10.1089/scd.2012.0373
28. Vilquin JT, Rosset P. Mesenchymal stem cells in bone and cartilage repair: Current status. Regen Med. 2006;1(4):589-604. DOI: 10.2217/17460751.1.4.589
29. Çetinkaya M, Demirutku A. Interfragmental fixation of the rostral mandibular fracture with cerclage wire in a thoroughbred English horse. Turk J Vet Anim Sci. 2012;36:67-71. DOI: 10.3906/vet-1102-792

30. Ahn H, Gong DJ, Lee HH, Seo JY, Song KM, Eom SJ, Yeo SY. Mechanical properties of porcine and fish skin-based collagen and conjugated collagen fibers. Polymers. 2021;13(13):2151. DOI: 10.3390/polym13132151
31. Min L. Effect of carbon dioxide laser (10600nm) on hydroxyproline in mouse skin. Pak J Pharm Sci. 2019:32. [available at]
32. Yoon BH, Yu W. Clinical utility of biochemical marker of bone turnover: Fracture risk prediction and bone healing. J Bone Metab. 2018;25(2):73-78. DOI: 10.11005/jbm.2018.25.2.73

تأثيرات نخاع العظم الذاتي على شفاء كسور العظام الطويلة التي تم تقليلها بواسطة المثبتات الهيكلية الخارجية في الماعز

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الخلاصة

تهدف هذه الدراسة إلى تقييم تأثير نخاع العظم الذاتي على كسور المشط ومشط القدم في الماعز الواردة إلى عيادة الجراحة الداخلية في جامعة العلوم البيطرية والحيوانية (UVAS) في لاهور، باكستان. تم توزيع ما مجموعه ٢٠ أنثى من الماعز الشمندر تزن ٢٠-٢٢,٥ كغم وتتراوح أعمارها بين ٨-١٢ شهرا في مجموعات متساوية من خمسة لكل منها: G1 و G2 و G3 و G4. تم تثبيت في G1 باستخدام مثبت هيكلي خارجي خطي، وتم تثبيت في G3 بمثبت دائري وعولجت بعدة دفعات من نخاع العظم، بينما تم تثبيت في G2 بنفس طريقة G1، وتم تثبيت في G4 بنفس طريقة G3 وعولجت بمحلول ملحي طبيعي في الأيام ٠ ، ١٤ و ٢٨. تم استخدام مقياس الاتحاد الشعاعي (RUS) ودرجة تحمل الوزن (WBS) لتقييم معدل الإصلاح بعد المعالجة، ولوحظت تغيرات كيميائية حيوية مصلية في الأيام ٠ و ٧ و ١٤ و ٢٨ و ٤٥. أظهرت النتائج وجود فرق معنوي (P<0.05) في التئام العظام بين المجموعات المعالجة بشفط نخاع العظم (BMA) والمجموعات المعالجة بمحلول ملحي طبيعي. كانت درجات RUS و WBS في G1 و G3 أعلى بكثير (P<0.05) مقارنة بالحيوانات G2 و G4 في الأيام ٧ و ١٤ و ٢٨. علاوة على ذلك، كان لدى G1 و G3 مستويات أكبر بكثير (P<0.05) من الفوسفاتيز القلوي، والهيدروكسي برولين الكلي، والهيدروكسي برولين الحر في G2 و G4، على التوالي. في الختام، يساعد تطبيق BMA في موقع الكسر في التئام عظام المشط والمشط المكسورة المتوافقة مع المثبتات الخارجية (الخطية والدائرية) في الماعز.