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## Evaluation of tibial fracture healing by estimation of biochemical and physiological parameters in dogs

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#### Abstract

The objective of the study was to evaluate tibial fracture healing by estimation of serum parameters such as calcium, phosphorus and alkaline phosphatase as well as physiological parameters such as heart rate, respiratory rate and rectal temperature in dogs. The present study included a total of 12 dogs with tibial fractures which were divided into 2 groups of 6 dogs each. In group-I, the tibial fractures were repaired using the intramedullary pinning technique. In group-II, the technique of fracture repair used was the intramedullary interlocking nailing technique. In both groups, on the 0<sup>th</sup> day pre-operatively and on the 15<sup>th</sup>, 30<sup>th</sup> and 60<sup>th</sup> day post-operatively, whole blood was collected and physiological parameters were recorded. Thus, calcium, phosphorus, alkaline phosphatase, heart rate, respiratory rate and rectal temperature were estimated at different stages of fracture healing. Biochemical and physiological parameters alone were insubstantial in the assessment of fracture healing and should be correlated clinically and radiologically for an all-encompassing, accurate evaluation of fracture healing.

Keywords: tibia, calcium, phosphorus, alkaline phosphatase, heart rate, respiratory rate, temperature

## Introduction

Bones are metabolically active organs. Their reconstruction is crucial for the proper functioning of the skeletal system during bone growth and remodelling, fracture healing and maintaining calcium–phosphorus homeostasis <sup>[1]</sup>. Calcium levels present some changes in different periods of the fracture healing process <sup>[2]</sup>. The fluctuation of serum calcium levels may represent the capability of calcium transportation, reservation, bone metabolism and restoration <sup>[3]</sup>. Phosphorus is the second, next to calcium, basic component of bone tissue <sup>[4]</sup>. The appropriate level of inorganic phosphorus is crucial for the activity of osteoblasts and osteocytes in the process of matrix mineralization <sup>[5]</sup>. Alkaline phosphatase is a marker of bone formation. Alkaline phosphatase is widely found in many organs, including liver, kidney and bone. Liver and osteoblasts are the main sources of serum alkaline phosphatase, accounting for about 50% each <sup>[6, 7]</sup>. The physiological status varies individually, however, the relation of physiological parameters such as heart rate, respiratory rate and rectal temperature were studied with respect to fracture healing.

## **Materials and Methods**

The study included twelve cases of tibia fractures in dogs which were stabilised using the intramedullary pinning technique in group-I or the intramedullary interlocking nailing technique in group-II. In both groups, whole blood was collected and physiological parameters were recorded on the 0<sup>th</sup> day pre-operatively and on the 15<sup>th</sup>, 30<sup>th</sup> and 60<sup>th</sup>, day post-operatively. The whole blood was collected aseptically from the cephalic or saphenous veins of the dogs in clot activator vacutainers and serum was separated. Calcium (mg/dL), phosphorus (mg/dL) and alkaline phosphatase (IU/L) was estimated on a semi-automated clinical chemistry analyser. Heart rate (beats/minute) was noted by observation of the chest movement and rectal temperature (°F) was recorded by placing a clinical thermometer in the rectum of the dogs in both groups. The data obtained was tabulated and statistically analysed using student's t-test <sup>[8]</sup>.

## **Results and Discussion**

## Calcium levels (mg/dL)

In group-I animals, serum calcium level significantly decreased ( $p \le 0.01$ ) from the 15<sup>th</sup> to 30<sup>th</sup> day when compared to pre-operative day 0. It came back to normal by the 60<sup>th</sup> day. In group-II animals, the serum calcium level decreased ( $p \le 0.01$ ) on day 15 followed by a rising trend from day 30 to day 60 with a significant rise ( $p \le 0.01$ ) on the 60<sup>th</sup> day. Comparison between groups revealed that the serum calcium level was significantly ( $p \le 0.05$ ) higher on day 15 in group-I as compared to group-II, whereas, vice-versa was observed on day 30. There is a paucity in literature in accordance with these findings. The Mean  $\pm$  SE values for calcium (mg/dL) are given in figure 1 for both groups.

#### Phosphorus levels (mg/dL)

Post-operative hyperphosphatemia ( $p \le 0.01$ ) was observed from the 15<sup>th</sup> day to the 60<sup>th</sup> day in both groups of animals. From day 15 to day 30, a rising trend was observed in both the groups, whereas, it showed a declining trend on the 60<sup>th</sup> day in both groups <sup>[9]</sup>. Comparison between the two groups revealed that from the pre-operative day to the 30<sup>th</sup> day there was significantly ( $p \le 0.05$ ) higher serum phosphorus level in group-II when compared to group-I animals, whereas, on day 60 there was non-significant difference between group-I and group-II animals. The Mean  $\pm$  SE values for phosphorus (mg/dL) are given in figure 2 for both groups.

## Alkaline phosphatase levels (IU/L)

Alkaline phosphatase enzyme level was significantly ( $p \le 0.01$ ) increased from day 15 to day 60 in both the groups of animals when compared to the pre-operative level. Comparison between both groups revealed that group-II animals had a significantly higher ( $p \le 0.05$ ) alkaline phosphatase level on day 0 and day 60 when compared to the group-I animals. The levels of serum alkaline phosphatase increased from the preoperative day to the 14<sup>th</sup> day and there after decreased and reached the normal values by the 60<sup>th</sup> day <sup>[10]</sup>. Highest value of serum alkaline phosphatase was observed on the 14<sup>th</sup> postoperative day <sup>[11]</sup>. Serum phosphorus followed a proportional pattern to alkaline phosphatase changes; however, serum calcium values followed an inverse pattern to alkaline phosphatase changes <sup>[12]</sup>. The Mean ± SE values for alkaline phosphatase (IU/L) are given in figure 3 for both groups.



**Fig 1:** Mean ± SE calcium (mg/dL) on different days postoperatively in both groups of dogs

Means bearing superscript\*\* differ significantly ( $p \le 0.01$ ) from interval 'before (Day 0)' within the group

Means bearing superscript a, b differ significantly ( $p \le 0.05$ ) between the groups at corresponding intervals



**Fig 2:** Mean ± SE phosphorus (mg/dL) on different days postoperatively in both groups of dogs

Means bearing superscript\*\* differ significantly  $(p \le 0.01)$  from interval 'before (Day 0)' within the group Means bearing superscript a, b differ significantly  $(p \le 0.05)$  between the groups at corresponding intervals



Fig 3: Mean  $\pm$  SE alkaline phosphatase (IU/L) on different days post-operatively in both groups of dogs

Means bearing superscript\*\* differ significantly  $(p \le 0.01)$  from interval 'before (Day 0)' within the group Means bearing superscript a, b differ significantly  $(p \le 0.05)$  between the groups at corresponding intervals

### Heart rate (beats/minute)

In group-I animals, there was an overall decrease  $(p \le 0.01)$  in heart rate when compared to the day 0 value. Fluctuating trends were observed from the 15<sup>th</sup> day to the 60<sup>th</sup> day. On the 15<sup>th</sup> day there was decreased heart rate followed by slight increase on the 30<sup>th</sup> day and the heart rate decreased once again on the 60<sup>th</sup> day. Group-II animals showed significant tachycardia ( $p \le 0.01$ ) from the 15<sup>th</sup> day to the 60<sup>th</sup> day when compared to the 0<sup>th</sup> day. Comparison between groups revealed that group-I animals had a higher ( $p \le 0.05$ ) heart rate as compared to group-II animals at all intervals of study. The Mean  $\pm$  SE values for heart rate (beats/minute) are given in figure 4 for both groups.

## **Respiratory rate (breaths/minute)**

In group-I, the respiratory rate fluctuated within the normal

physiological range. The respiratory rate in group-I animals decreased slightly on the 15<sup>th</sup> day followed by a rising trend from day 30 to day 60 when compared to day 0. In group-II animals, the respiratory rate significantly decreased ( $p \le 0.05$ ) between the 15<sup>th</sup> to 30<sup>th</sup> day intervals when compared to day 0. By the 60<sup>th</sup> day, respiratory rate came to normal. Comparison between the groups revealed that group-II animals had a higher respiratory rate ( $p \le 0.05$ ) than group-II animals on day 0. However, there was no significant difference between the 15<sup>th</sup>, 30<sup>th</sup> and 60<sup>th</sup> day intervals of study. The Mean  $\pm$  SE values for respiratory rate (breaths/minute) are given in figure 5 for both groups.

## Rectal temperature (°F)

In both groups, the rectal temperature fluctuated within the normal physiological limits when compared to pre-operative day 0. However, there was significant hyperthermia ( $p \le 0.05$ ) on day 60 when compared to day 0 in group-I animals. Comparison between groups revealed that there was no significant difference (p>0.05) between all the intervals of the study. The Mean  $\pm$  SE values for rectal temperature (°F) are given in figure 6 for both groups.



Fig 4: Mean ± SE of heart rate (beats/minute) on different days postoperatively in both groups of dogs

Means bearing superscript\*\* differ significantly  $(p \le 0.01)$  from interval 'before (Day 0)' within the group

Means bearing superscript a, b differ significantly ( $p \le 0.05$ ) between the groups at corresponding intervals



Fig 5: Mean  $\pm$  SE of respiratory rate (breaths/minute) on different days post-operatively in both groups

Means bearing superscript\* differ significantly  $(p \le 0.05)$  from interval 'before (Day 0)' within the group

Means bearing superscript a, b differ significantly ( $p \le 0.05$ ) between the groups at corresponding intervals



Fig 6: Mean  $\pm$  SE of rectal temperature (°F) on different days postoperatively in both groups of dogs

Means bearing superscript\* differ significantly ( $p \le 0.05$ ) from interval 'before (Day 0)' within the group

## Conclusions

In conclusion, the biochemical and physiological parameters alone were insubstantial in the assessment of fracture healing and should be correlated clinically and radiologically for an all-encompassing, accurate evaluation of fracture healing.

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