ISSN: 2320 - 4230

Journal of Drug Discovery and Therapeutics 1 (11) 2013, 18-23

RESEARCH ARTICLE

Evaluation of Serum Calcium, Phosphorus, and Alkaline Phosphatase Levels in Patients with Fracture Healing: A Prospective Study

Dr. Nilesh Loya

Associate Professor, Department of Orthopedic, Annasaheb Chudamam Patil Memorial Medical College, Dhule (A.C.P.M. Medical College, Dhule)

Received 11 September 2013; Revised 10 October 2013; Accepted 17 November 2013

Abstract

Fracture healing is a complex biological process involving inflammation, repair, and remodeling phases. Biochemical markers such as serum calcium, phosphorus, and alkaline phosphatase (ALP) may reflect osteoblastic activity and mineralization during bone repair.

Objectives

1. To assess changes in serum calcium, phosphorus, and ALP levels during fracture healing.
2. To determine the association of these biochemical markers with the rate of fracture union.

Methods

This prospective observational study included adult patients with long-bone fractures managed either conservatively or surgically. Serum calcium, phosphorus, and ALP levels were measured at baseline, 6 weeks, and 12 weeks. Fracture union was assessed radiologically and clinically at follow-up visits. Patients were categorized into normal union and delayed union groups.

Results

A total of 120 patients completed follow-up. Mean ALP levels showed a significant rise at 6 weeks compared with baseline ($p<0.001$) and gradually declined by 12 weeks. Serum calcium and phosphorus demonstrated modest but significant changes during healing ($p<0.05$). Patients with delayed union had lower ALP elevation at 6 weeks and persistently lower phosphorus levels compared with patients achieving normal union ($p<0.05$).

Conclusion

Serum ALP appears to be a useful biochemical marker of fracture healing, while serum calcium and phosphorus provide supportive information regarding mineral metabolism. Early changes in ALP may help identify patients at risk of delayed fracture union.

Keywords: Fracture healing, Alkaline phosphatase, Calcium, Phosphorus, Delayed union, Bone biomarkers.

Introduction:

Fractures are among the most common musculoskeletal injuries encountered in orthopedic practice and represent a significant cause of morbidity worldwide. Bone possesses a remarkable capacity for regeneration, and fracture healing is a highly organized physiological process that restores the structural integrity and mechanical strength of the injured bone. Despite advances in orthopedic management, delayed union and non-union of fractures continue to pose substantial clinical challenges, leading to prolonged disability, increased healthcare costs, and reduced quality of life. [1] Therefore, early identification of factors influencing fracture healing and reliable methods for monitoring the healing process are of considerable clinical importance. [2]

Fracture healing involves a complex sequence of biological events that can be broadly divided into three overlapping phases: the inflammatory phase, the reparative phase, and the remodeling phase. Immediately after a fracture, hematoma formation occurs at the fracture site, followed by the

*Corresponding author: Dr. Nilesh Loya

recruitment of inflammatory cells and release of cytokines and growth factors.[3] These mediators stimulate the proliferation and differentiation of osteogenic cells, leading to the formation of soft callus and subsequently hard callus through endochondral and intramembranous ossification. The final remodeling phase restores the normal architecture and biomechanical properties of bone. Successful fracture healing requires adequate vascular supply, mechanical stability, nutritional support, and coordinated cellular activity. [4]

Biochemical markers of bone metabolism have emerged as valuable tools for studying skeletal physiology and pathology. These markers reflect the activity of bone-forming osteoblasts and bone-resorbing osteoclasts and may provide insight into the dynamic process of fracture healing. [5] Among the various biochemical parameters, serum calcium, phosphorus, and alkaline phosphatase have attracted considerable attention because of their essential roles in bone formation and mineralization. [6]

Calcium is the most abundant mineral in the human body, with approximately 99% stored within the skeletal system. It plays a critical role in bone mineralization, neuromuscular function, and cellular signaling. During fracture healing, calcium is required for the formation of hydroxyapatite crystals, which provide strength and rigidity to newly formed bone. [7] Although serum calcium levels are tightly regulated through hormonal mechanisms involving parathyroid hormone, calcitonin, and vitamin D, subtle changes may occur during active bone repair. Evaluating serum calcium concentrations during fracture healing may provide useful information regarding mineral metabolism and skeletal regeneration. [8]

The present prospective observational study was therefore undertaken to evaluate serial changes in serum calcium, phosphorus, and alkaline phosphatase levels in patients with fractures and to determine their association with the rate of fracture union. By correlating biochemical markers with clinical and radiological outcomes, the study aims to provide insights into the role of these readily available laboratory parameters in monitoring fracture healing and predicting treatment outcomes. The findings may contribute to the development of simple and cost-effective strategies for the assessment of fracture repair in routine orthopedic practice.

Materials and Methods

Study design and setting:

A prospective observational study conducted in the Department of Orthopedics of a tertiary care teaching hospital.

Study population:

Adult patients (18–65 years) presenting with fresh long-bone fractures were recruited consecutively.

Sample Size

A total of 120 patients with radiologically confirmed long-bone fractures were included in this prospective observational study. The sample size was determined based on the average number of eligible fracture patients attending the Orthopedics Department during the study period and those fulfilling the inclusion and exclusion criteria. All enrolled patients were followed prospectively for 12 weeks to assess changes in serum calcium, phosphorus, and alkaline phosphatase levels and their association with fracture healing and union outcomes.

Inclusion criteria

- Patients aged 18–65 years.
- Radiologically confirmed fresh fractures (<7 days).
- Willingness to participate and provide informed consent.

Exclusion criteria

- Pathological fractures.
- Chronic kidney disease.
- Metabolic bone disease.
- Chronic liver disease.
- Use of medications affecting bone metabolism.

Data collection

Clinical details, fracture type, treatment modality, and demographic information were recorded. Venous blood samples were collected at:

1. Baseline (within 48 hours of admission)
2. 6 weeks after fracture
3. 12 weeks after fracture

The following parameters were measured using standard automated methods:

Sample collection: 5 ml of venous blood was collected aseptically from each participant at baseline (within 48 hours of fracture), at 6 weeks, and at 12 weeks during follow-up. Blood samples were collected in plain vacutainer tubes and allowed to clot at room temperature. The samples were then centrifuged at 3000 rpm for 10 minutes to separate serum. The obtained serum was analyzed immediately or stored at 2–8°C until analysis.

Analysis: The biochemical parameters assessed in the present study included serum calcium, serum phosphorus, and serum alkaline phosphatase (ALP). Serum calcium was estimated by the Arsenazo III method, serum phosphorus was measured using the UV molybdate method, and serum alkaline phosphatase activity was determined by the IFCC kinetic method. All analyses were performed on a fully automated clinical chemistry analyzer using standard commercial reagent kits according to the manufacturer's instructions. These parameters were evaluated at baseline, 6 weeks, and 12 weeks of follow-up to assess their role in fracture healing and their association with fracture union.

Result:

Table 1. Demographic and Clinical Characteristics of Study Participants (n = 120)

Variable	Frequency (n)	Percentage (%)
Age Group (Years)		
18–30	35	29.2
31–45	48	40.0
46–60	28	23.3
>60	9	7.5
Gender		
Male	72	60.0
Female	48	40.0
Type of Fracture		
Tibia	48	40.0
Femur	36	30.0
Humerus	24	20.0
Radius/Ulna	12	10.0
Treatment Modality		
Conservative	50	41.7
Surgical	70	58.3

Table 1 shows the demographic and clinical characteristics of the 120 study participants. The majority of patients were aged 31–45 years (40.0%), followed by 18–30 years (29.2%). Males constituted 60.0% of the study population, while females accounted for 40.0%. Tibial fractures (40.0%) were the most common type of fracture, followed by femoral fractures (30.0%). Regarding treatment, 58.3% of patients underwent surgical management, whereas 41.7% received conservative treatment. Overall, the study population was predominantly composed of young to middle-aged males with lower-limb fractures.

Table 2. Serum Calcium, Phosphorus, and Alkaline Phosphatase Levels During Fracture Healing (n = 120)

Parameter	Baseline (Mean ± SD)	6 Weeks (Mean ± SD)	12 Weeks (Mean ± SD)	p-value
Serum Calcium (mg/dL)	9.10 ± 0.60	9.40 ± 0.70	9.30 ± 0.60	0.020*
Serum Phosphorus (mg/dL)	3.50 ± 0.50	3.90 ± 0.60	3.70 ± 0.50	0.010*
Alkaline Phosphatase (IU/L)	102 ± 28	165 ± 40	138 ± 35	<0.001*

Table 2 shows the changes in serum calcium, phosphorus, and alkaline phosphatase levels during fracture healing. Mean serum calcium increased from 9.10 ± 0.60 mg/dL at baseline to 9.40 ± 0.70 mg/dL at 6 weeks and remained slightly elevated at 9.30 ± 0.60 mg/dL at 12 weeks (p = 0.020). Serum phosphorus also showed a significant rise from 3.50 ± 0.50 mg/dL at baseline to 3.90 ± 0.60 mg/dL at 6 weeks, followed by a slight decline at 12 weeks (p = 0.010). Alkaline phosphatase demonstrated the most pronounced change, increasing from 102 ± 28 IU/L at baseline to 165 ± 40 IU/L at 6 weeks and decreasing to 138 ± 35 IU/L at 12 weeks (p < 0.001). These findings indicate significant biochemical changes during fracture healing, particularly increased osteoblastic activity reflected by elevated alkaline phosphatase levels.

Table 3. Comparison of Biochemical Markers Between Normal Union and Delayed Union Groups at 6 Weeks

Parameter	Normal Union (n = 102) Mean ± SD	Delayed Union (n = 18) Mean ± SD	p-value
Serum Calcium (mg/dL)	9.50 ± 0.60	9.20 ± 0.50	0.080
Serum Phosphorus (mg/dL)	4.00 ± 0.50	3.50 ± 0.40	0.004*
Alkaline Phosphatase (IU/L)	172 ± 38	130 ± 32	<0.001*

Table 3 compares biochemical markers between patients with normal union and delayed union at 6 weeks. Serum calcium levels were slightly higher in the normal union group (9.50 ± 0.60 mg/dL) compared to the delayed union group (9.20 ± 0.50 mg/dL), but the difference was not statistically significant (p = 0.080). Serum phosphorus levels were significantly higher in patients with normal union (4.00 ± 0.50 mg/dL) than in those with delayed union (3.50 ± 0.40 mg/dL) (p = 0.004). Similarly, alkaline phosphatase levels were markedly elevated in the normal union group (172 ± 38 IU/L) compared to the delayed union group (130 ± 32 IU/L) (p < 0.001). These findings suggest that higher phosphorus and alkaline phosphatase levels are associated with successful fracture healing and may serve as useful indicators of fracture union.

Discussion:

Present study Table 1 shows the demographic and clinical characteristics of the 120 study participants. The majority of patients were aged 31–45 years (40.0%), followed by 18–30 years (29.2%). Males constituted 60.0% of the study population, while females accounted for 40.0%. Tibial fractures (40.0%) were the most common type of fracture, followed by femoral fractures (30.0%). Regarding treatment, 58.3% of patients underwent surgical management, whereas 41.7% received conservative treatment. Overall, the study population was predominantly composed of young to middle-aged males with lower-limb fractures.

Similar findings were reported by Court-Brown CM and Caesar B, who found that fractures occur more frequently in males and economically productive age groups due to increased exposure to occupational hazards, road traffic accidents, and sports-related injuries. [9]

In the present study, tibial fractures constituted the largest proportion of cases, followed by femoral fractures. This observation is consistent with the findings of Court-Brown CM et al., who reported that lower-limb fractures, particularly those involving the tibia and femur, are among the most common long-bone fractures encountered in orthopedic practice. [10]

Regarding treatment modality, more than half of the patients underwent surgical management. Similar trends have been reported by Giannoudis PV et al., who emphasized that operative stabilization is frequently employed for long-bone fractures to provide mechanical stability, facilitate early mobilization, and promote optimal fracture healing. The demographic and clinical characteristics observed in the present study therefore align well with previously published orthopedic literature. [11]

The present study Table 2 shows the changes in serum calcium, phosphorus, and alkaline phosphatase levels during fracture healing. Mean serum calcium increased from 9.10 ± 0.60 mg/dL at baseline to 9.40 ± 0.70 mg/dL at 6 weeks and remained slightly elevated at 9.30 ± 0.60 mg/dL at 12 weeks (p = 0.020). Serum phosphorus also showed a significant rise from 3.50 ± 0.50 mg/dL at baseline to 3.90 ±

0.60 mg/dL at 6 weeks, followed by a slight decline at 12 weeks ($p = 0.010$). Alkaline phosphatase demonstrated the most pronounced change, increasing from 102 ± 28 IU/L at baseline to 165 ± 40 IU/L at 6 weeks and decreasing to 138 ± 35 IU/L at 12 weeks ($p < 0.001$). These findings indicate significant biochemical changes during fracture healing, particularly increased osteoblastic activity reflected by elevated alkaline phosphatase levels.

Similar findings were reported by Moghaddam A et al., who observed a significant increase in serum alkaline phosphatase during fracture healing, with peak levels occurring during the early stages of callus formation. The authors concluded that alkaline phosphatase is a useful biochemical marker for monitoring bone regeneration and osteoblastic activity. [12]

Likewise, Singh AJ and colleagues reported elevated serum alkaline phosphatase levels in patients with healing long-bone fractures, suggesting that increased enzyme activity reflects active bone formation and mineralization. [13]

The observed rise in serum phosphorus levels during the healing process is also consistent with the findings of Moghaddam A et al., who demonstrated alterations in mineral metabolism during fracture repair, with phosphorus playing a crucial role in hydroxyapatite formation and skeletal mineralization. [1] Although serum calcium is tightly regulated, the slight increase noted in the present study has been described previously and may be related to increased mineral turnover associated with callus maturation. [14]

Overall, the present findings support previous evidence that serum calcium, phosphorus, and particularly alkaline phosphatase undergo significant changes during fracture healing and may serve as useful biochemical indicators of bone repair.

The present study Table 3 compares biochemical markers between patients with normal union and delayed union at 6 weeks. Serum calcium levels were slightly higher in the normal union group (9.50 ± 0.60 mg/dL) compared to the delayed union group (9.20 ± 0.50 mg/dL), but the difference was not statistically significant ($p = 0.080$). Serum phosphorus levels were significantly higher in patients with normal union (4.00 ± 0.50 mg/dL) than in those with delayed union (3.50 ± 0.40 mg/dL) ($p = 0.004$). Similarly, alkaline phosphatase levels were markedly elevated in the normal union group (172 ± 38 IU/L) compared to the delayed union group (130 ± 32 IU/L) ($p < 0.001$). These findings suggest that higher phosphorus and alkaline phosphatase levels are associated with successful fracture healing and may serve as useful indicators of fracture union.

Similar results were reported by Moghaddam et al., who observed that patients with successful fracture healing exhibited significantly elevated levels of bone formation markers, particularly alkaline phosphatase, during the reparative phase of fracture healing. The authors concluded that alkaline phosphatase is a useful biochemical marker for assessing osteoblastic activity and monitoring bone regeneration.[7]

Claes et al. emphasized that effective fracture healing is associated with active mineralization and callus maturation, processes that require adequate phosphate availability and increased osteoblastic function.[3] This may explain the significantly higher serum phosphorus levels observed in the normal union group in the present study.

Conclusion: Serum ALP appears to be a useful biochemical marker of fracture healing, while serum calcium and phosphorus provide supportive information regarding mineral metabolism. Early changes in ALP may help identify patients at risk of delayed fracture union.

Reference

1. Einhorn TA. The science of fracture healing. J Orthop Trauma. 2005;19(10 Suppl):S4–S6.
2. Giannoudis PV, Einhorn TA, Marsh D. Fracture healing: the diamond concept. Injury. 2007;38(Suppl 4):S3–S6.
3. Claes L, Recknagel S, Ignatius A. Fracture healing under healthy and inflammatory conditions. Nat Rev Rheumatol. 2012;8(3):133-143.
4. Marsell R, Einhorn TA. The biology of fracture healing. Injury. 2011;42(6):551-555
5. Seibel MJ. Biochemical markers of bone turnover: part I. Biochemistry and variability. Clin Biochem Rev. 2005;26(4):97-122.
6. Raggatt LJ, Partridge NC. Cellular and molecular mechanisms of bone remodeling. J Biol Chem. 2010;285(33):25103–25108.
7. Boskey AL. Bone mineralization. In: Primer on the Metabolic Bone Diseases and Disorders of

Mineral Metabolism. 8th ed. Ames, Iowa: Wiley-Blackwell; 2013. p. 49-58.

8. Guyton and Hall Textbook of Medical Physiology. 13th ed. Philadelphia: Elsevier; 2016. Chapter 79: Parathyroid Hormone, Calcitonin, Calcium and Phosphate Metabolism, Vitamin D, Bone and Teeth.
9. Court-Brown CM, Caesar B. Epidemiology of adult fractures: A review. Injury. 2006;37(8):691–697.
10. Court-Brown CM, McBirnie J. The epidemiology of tibial fractures. J Bone Joint Surg Br. 1995;77(3):417–421.
11. Giannoudis PV, Einhorn TA, Marsh D. Fracture healing: the diamond concept. Injury. 2007;38(Suppl 4):S3–S6.
12. Moghaddam A, Weiss S, Wentzensen A, et al. Biochemical markers in bone fracture healing. Eur J Trauma Emerg Surg. 2011;37(1):1–11.
13. Singh AJ, et al. Changes in serum alkaline phosphatase during fracture healing. Indian J Orthop. 2013;47(5):487–492.
14. Marsell R, Einhorn TA. The biology of fracture healing. Injury. 2011;42(6):551–555.