



EFFECT OF KIDNEY TRANSPLANT ON CERTAIN BONE PARAMETERS

Biochemistry

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ABSTRACT

The key function of kidney is to remove waste material from body via filtration. If this ability of kidney is lost the resulting condition is called End Stage Renal Disease (ESRD). This condition often recovered by kidney transplantation.

Kidney is one of the key organs in synthesis of Vitamin D (Calcitriol). Calcitriol plays vital role in bone metabolism by regulating minerals like calcium and phosphorous. Beside these two minerals alkaline phosphatase also considered as key indicator in bone health.

As a result of transplant besides reinstating the main function of kidney to remove waste material, changes in bone parameters are also expected. The current study was undertaken to study these parameters to uncover the changes after kidney transplantation.

Present study included 31 patients who had undergone kidney transplant. Serum levels of Calcium, Phosphorus and Alkaline phosphatase were estimated before and after transplant for the period of 6 months.

Result: The present study has revealed the rise in the study parameters. The hypercalcemia is evident among the transplant patients. Phosphorus levels normalized at the end of study period where as alkaline phosphatase showed initial rise and then a gradual decline.

KEYWORDS

Kidney transplant, Calcium, Phosphorus, Alkaline phosphatase, Bone parameters.

INTRODUCTION

Chronic kidney disease (CKD) is graded based on damage to the kidney occurred due to any reasons and is measure by Glomerular filtration rate (GFR). GFR \leq 15ml/min is considered as stage 5 CKD which is often called End Stage Renal Disease (ESRD) (1). This can only be treated either by dialysis or transplant. The former needs to be done for life time while the latter is a one-time surgical procedure depending on survival graft and comes with life time medication(2).

ESRD induces a dysregulated metabolism which causes alterations in many biochemical parameters (3). Kidney is a major organ involved in homeostasis of calcium (Ca) and phosphorus (P) beside bone. Damage to kidney eventually causes asynchronous changes in bone parameters. It mainly affects Calcium, phosphorus metabolism and activity of alkaline phosphatase, with certain other biochemical parameters which have direct or indirect effect on bone metabolism. (4)

Vitamin D is a key molecule in calcium metabolism. It increases the absorption of Ca and P by active transcellular pathway. Vitamin D exists mainly in two forms viz calcidiol and calcitriol. The former is synthesized in liver which then moves to kidney to be converted to active form (5,6). Given the fact that calcitriol is synthesized in kidney and is strategic molecule to influence Ca and P, dysfunction of kidney can immensely affect on these two parameters. In normal circumstances Ca and P show reciprocal relation. Like Ca, P levels are also regulated by kidney via parathyroid hormone (PTH), Vitamin D and fibroblast growth factor-23 (FGF-23) (7)

As Ca and P are constituted structural part bone, variation in their levels involves changes in bone turn over (8). Among the various steps of mineralization of bone, the first is to form hydroxyapatite. Alkaline phosphatase (ALP) is the enzyme which belongs to hydrolase catalyzes reaction to form inorganic phosphorus from bound organic form. (9) This indicates that ALP ensures constant supply of P in building up hydroxyapatite and thereby bone.

The current study had been planned to evaluate sequential changes in these bone parameters before and after kidney transplantation. After transplantation it will be noteworthy to evaluate the bone metabolism through these parameters and its relation with functional graft.

MATERIAL AND METHOD

It was conventional sampling with observation and analytical study.

31 ESRD patients whose eGFR \leq 15ml/min, who were awaiting kidney transplant were selected for study. The study was approved by Institutional ethical committee. Informed consent was taken from each participants. Also, for those patients who were not in condition to provide self-consent, the consent was taken from their family head/members. After transplantation, patients were receiving immunosuppressants (Tacrolimus) therapy as per Kidney Disease: Improving Global Outcomes (KDIGO) (7) guidelines and was part of routine patient management. Study has also included 31, age and sex matched healthy controls selected from same socioeconomic group.

Inclusion and exclusion criteria: The patient who lost the follow-up have been excluded. Those who were having habits of smoking, tobacco chewing, alcohol have also been excluded. Subjects in both the groups were not under any medication which could directly affect the study parameters. The known cases of bone deformities or disorder related to parathyroid gland, were also excluded.

Sample collection and procedure: 5 ml of blood sample was collected before transplant and then for the period of 6 months after transplant from each subject. The intervals of sample collected and analyzed has been depicted in Table. 1. Samples were processed as per requirement of the standard method used for determination of biochemical parameters.

All the parameter were estimated calorimetrically by Erba Diagnostics Mannheim kits. Serum Ca was measure by using Arsenazo III end point method (10). Arsenazo III forms coloured chromophore when combines with Ca at 6.75 pH. Serum inorganic Phosphorus was analyzed by Ammonium Molybdenum Method (11) In acidic medium P reacts with molybdate to form color complex of Phosphomolybdate. Serum ALP activity was estimated by kinetic method adapted from Wilkinson (12).

STATISTICAL ANALYSIS: After collection of data the statistical analysis was done by using MS-Excel-2019 software. Values were expressed in mean \pm SD and were compared by using students' t test. Correlation coefficient was calculated by using Pearson's method.

RESULTS:

Results obtained in this study are expressed in mean \pm SD. Values obtained from Kidney transplant recipients (Group II) was subdivided as per the follow-up intervals as shown in Table 1. Pre-transplant (Pre-

Tx) values were compared with healthy controls (Group I) by unpaired t test. Whereas Group II values were compared with paired t test. Pearson's Correlation formula was used to establish correlation

Table 1 : Serum Calcium, Phosphorus and alkaline Phosphatase levels in Control and transplant recipients.

| Parameters | Controls (Group I) (n=31) | Transplant recipients (n=31) (Group II) | | | | | |
|----------------------------|---------------------------|---|---------------------|---------------------|------------|-------------------------|-------------------------|
| | | Pre-Tx | 1 st day | 7 th day | 1 Month | 3 Months | 6 Months |
| Calcium (mg/dl) | 9.97±1.27 | 13.50±1.68* | 9.84±1.84 | 12.61±1.18 | 10.94±2.05 | 11.28±2.07 [#] | 12.03±2.27 [#] |
| Phosphorus (mg/dl) | 3.63±0.76 | 4.64±0.68 | 4.48±0.47 | 3.08±0.45 | 2.83±0.38 | 3.00±0.43 | 2.99±0.58 |
| Alkaline phosphatase (U/L) | 92.29±20.95 | 245.7±63.67* | 281.5±101.3 | 230.6±49.3 | 202.5±49.3 | 172.2±32.5 | 137.0±35.1 |

* p value ≤ 0.05 when compared with control, # p value ≤ 0.05 when compared with Pre-Tx

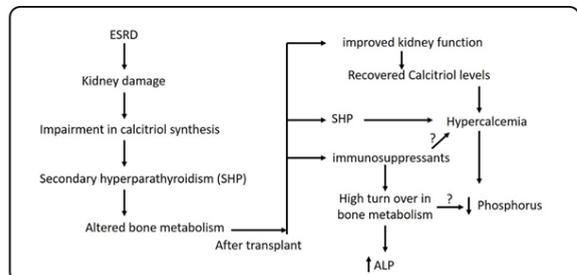


Diagram 1. Effect of ESRD, kidney transplant and immunosuppressant on bone parameters. (↑-increased, ↓-decreased; ?-role uncertain; ALP - alkaline phosphatase; ESRD-End stage renal disease)

DISCUSSION

The current study on bone parameters in KTx had revealed that the hypercalcemia is common among transplant recipients. However, the resulted values raised a bit higher than the control. (Table 1). These values had remained on higher side for 6 months with occasional insignificant fall. There are consistent variations in the various studies done so far. In fact, within the study groups the incidence of hypercalcemia (HC) was not a common trend but a majority patient showed HC (4,13). Many long duration studies have proven that HC lasts for first year after transplant, which latter gets normalized. (13) Our study has revealed that the Calcium levels have normalized in first few weeks after transplant, whereas the little increase was observed at the end of 6 months.

Secondary Hyperparathyroidism (SHP) is considered to be the reason for HC after transplantation. The occurrence of SHP is associated with uremic state preceding KTx. The other significant reason would be the improvement in the synthetic function of kidney which recovers circulating levels of calcitriol. (13, 14). The possible role of immunosuppressant drugs has remained unclear in previous studies. However, the steroids are potential factors to alter calcium metabolism. There are some evidences which suggest that tacrolimus levels cause bone resorption leading to HC. (15,16)

In the physiological state the phosphorus levels show reciprocal relation with calcium. In our study the similar observations were registered though the change is not significant. (Table 1). The phosphorus levels normalize gradually during 6 months. This is in contrast to Calcium levels which kept on fluctuating. Our study is at par with observations made by Reinhardt W *et al* (17) and as detailed in review published by Messa *et al* (13).

Parathyroid hormone is a regulator of bone mineral levels especially calcium & phosphorus. SHP exerts its phosphaturic effect on transplanted kidney. Further the normalization of levels of phosphorus can also be considered as effect of reduced doses of steroid administered as immunosuppressant regimen. (17)

Our study demonstrated increased levels of Alkaline Phosphatase (ALP) in first month of translation. Then after gradual reduction was observed (Table 1). There is significant change in ALP levels from pretransplant status to post 6 months of transplant, though not completely equal to control. Many studies have registered similar observations. (13, 17, 18).

Mechanism of effect of immunosuppressants on ALP level is unclear, however the immunosuppression results in increased bone metabolism as bone cells are the main source of immune cells. With the high turnover of bone metabolism as observed with calcium and phosphorus the ALP levels showed the normal association.

Our study thus revealed that there is high turnover in bone metabolism causing altered levels of the study parameters in first 6 months of

between these parameters. p value ≤ 0.05 was considered as significant. Study had produced following results for various parameters in Group I and II.

kidney transplant (Diagram 1). The study also supports the fact that the high turnover in bone metabolism is having no relation with graft function. Further the recovered calcitriol synthesis can be considered as one of the factors causing alteration in levels of these parameters. However, there is considerable effect of immunosuppressant regimen.

CONCLUSION:

We observed hypercalcemia in KTx patients. In first 6 months the calcium levels remained fluctuating on higher side showing fall at the end. Whereas the phosphorus levels have gradually found to be decreased and normalized after 6 months. ALP levels have registered initial rise then started declining. This is indicative of high turnover of bone metabolism during first 6 months of KTx. The study has similar findings as registered by earlier researchers showing no correlation between graft function. The study is indicative that the immunosuppressant regimen has effect on these parameters resulting in their alteration.

LIMITATIONS

We have studied these parameters for the period of 6 months after transplant. Many researchers have registered the normal bone metabolism after 1 year of transplant. So, study period could have been extended, which was not possible for us due to limited resources and time constraint. We couldn't study the PTH levels and calcitonin levels. In addition to the studied parameters PTH and calcitonin could have added more information on present study.

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