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COMPREHENSIVE STUDY OF SOME PLASMA BIOCHEMICAL PARAMETERS IN RELATION TO BONE DISEASE IN HEMODIALYSIS PATIENTS IN ISFAHAN

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ABSTRACT

The concentrations of serum calcitonin, parathormone (PTH), amylase, alkaline phosphatase (ALKP), calcium and phosphorus were studied in pre-and post-dialysis patients with chronic renal failure. All patients had extremely elevated serum urea and creatinine concentrations with the mean values of 143.8 and 10.5 mg/dL respectively. The plasma amylase activity was higher than normal with a mean of 174 IU/L, but showed no significant changes following hemodialysis. The mean values for calcium and phosphorus in predialysis plasma were 7.2 and 7.5 mg/dL respectively, whereas it changed to 8.6 and 3.8 mg/dL post-dialysis. The extent of increase in PTH and alkaline phosphatase levels depended on the duration of dialysis in the majority of the patients. The longer the period of the dialysis, the higher the concentrations of PTH and ALKP. On the basis of this study the relationship between bone disease and hemodialysis has been discussed.

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INTRODUCTION

Chronic renal failure patients on long term dialysis may develop a series of disorders including neurological disease such as Alzheimer's disease,[1] encephalopathy,[2] anemia,[3] and imbalanced calcium homeostasis leading to the appearance of bone disease such as osteomalacia.[4] The ability of the kidney to hydroxylate 25-hydroxycholecalciferol is impaired in renal failure,[5] with subsequent reduction in intestinal absorption of calcium. The reduction in calcium absorption leads to hypocalcemia and stimulates the secretion of parathormone (PTH) causing secondary hyperparathyroidism[6] and consequent bone resorption.

Ellis and Peart in 1975 found that osteitis fibrosa due to secondary hyperparathyroidism occurred in 93%, osteomalacia in 40% and osteosclerosis in 30% of the patients with chronic renal failure.[7]

Accumulation of aluminum in bone occurs in most uremic patients undergoing hemodialysis, producing dialysis osteomalacia which is resistant to vitamin D therapy.[8]

Aluminum has been reported to pass across the dialysis membrane, enter blood circulation and interfere with bone mineralization.[9] The major aim of the present project was to study some plasma parameters in relation to bone metabolism comprehensively in a number of patients undergoing hemodialysis in the Shariati Hospital dialysis center in Isfahan.

MATERIALS AND METHODS

Chemicals: All chemicals were of reagent grade and purchased from Sigma Chemical Company unless otherwise stated.

Table I: Pre-and post-dialysis concentrations of calcium (Ca), phosphorus (P), amylase, urea and creatinine in 30 hemodialysed patients. The concentrations of Ca, P, urea and creatinine are expressed in mg/100 mL of serum and the activity of amylase is indicated as IU/L.

Patient No.	Pre-dialysis					Post-dialysis				
	Ca	P	Amylase	Urea	Creatinine	Ca	P	Amylase	Urea	Creatinine
1	7.5	6.2	128	129	7.9	8.5	4.1	135	59	5.1
2	4.8	6.3	148	135	7.5	7.7	2.5	155	64	4.2
3	10.2	3.8	274	148	11.8	11.4	6.8	198	74	6.9
4	8.1	7.4	191	139	10.4	10.2	2.7	205	79	6.5
5	8.3	7.6	124	158	9.9	8.0	5.2	121	84	7.3
6	6.7	12.2	127	143	10.4	8.7	5	147	63	6.2
7	7.2	13.1	185	159	11.5	8.9	5.1	179	72	6.6
8	7.6	5.8	187	148	7.7	8.6	3.9	194	58	4.3
9	7.1	8.6	114	138	9.2	9.0	4	122	80	5
10	6.8	7.1	195	150	9.8	7.2	3.6	183	65	6.2
11	6.3	6.4	123	163	13.5	8.3	3.4	148	72	6.9
12	5.4	7	200	147	10.4	6	3.1	144	76	5.5
13	7.7	4.3	136	134	9.8	8.6	2.1	139	62	5.3
14	7.4	7.4	190	139	9.1	9.8	9.5	183	73	4.5
15	8.1	8.3	117	150	9.1	8.4	5.4	166	77	6.5
16	7.2	6.6	270	118	11.2	10.4	2.6	259	82	6.3
17	7.6	4.0	151	150	9.6	9.8	3.7	169	65	6.4
18	8	5.4	135	155	14.6	6.4	3.1	111	79	8.6
19	8.3	8.5	116	139	10.2	8.3	5.5	129	67	5.9
20	6.4	3.7	154	89	3.8	10.3	3.4	171	47	2
21	10.6	9.9	105	193	11.9	9.8	3.9	128	59	4
22	5.4	6.2	145	158	14.7	7.9	3.4	220	65	7.5
23	6.7	8.5	200	169	12.9	7.8	2.7	198	79	7.2
24	4.7	8.1	189	163	11	7.7	2.5	197	72	7.9
25	7.6	6.6	217	146	10.5	10.8	3	209	71	6.3
26	8.4	10.4	266	123	9.7	9.9	2.9	274	61	6.5
27	6.9	7.1	197	148	11.2	7.1	3.2	189	85	6.9
28	5.8	9.4	221	139	11.9	9.9	8.6	230	74	7.1
29	6	8	194	133	12.8	7.7	5.3	207	91	8.1
30	7	6.2	242	141	11.5	7.7	3	263	63	4.5
Mean	7.2	7.5	174.7	143.8	10.5	8.6	3.8	179.1	70.6	6.0
±SD	1.4	2.1	52.2	15.6	2.2	103	104	43.3	9.7	104

Patients: 56 patients (39 male and 17 female) who were regularly admitted to the hospital were under investigation. Most of them had usually been dialysed two times weekly. For the determination of plasma biochemical parameters, blood samples were collected both pre-and post-dialysis in order to compare the changes during dialysis except otherwise stated. Plasma samples were separated by centrifugation and stored at 4°C for analysis, or used immediately.

Plasma biochemistry: Plasma urea, creatinine, calcium and phosphorus concentrations were determined by routine experimental methods used in clinical laboratories.

Alkaline phosphatase activity was determined by the method of Bessy, et.al. using p-nitrophenyl phosphate as substrate.[10] Amylase activity was assayed by the method of Caraway on the basis of starch hydrolysis. Plasma PTH and calcitonin were determined by immunoradiometric assay using commercial laboratory kits purchased from Diagnostic Products Corporation (Los Angeles, CA 90045) and radiation was measured by LKB 1275 gamma counter.

RESULTS

The concentrations of plasma calcium, phosphorus and amylase were determined in our patients maintained on regular hemodialysis who were identified to be uremic with the mean plasma urea and creatinine concentrations of 143.8 and 10.5 mg/dL respectively (Table I). It can be seen that following hemodialysis the concentration of calcium was elevated by 20%, whereas the phosphate level decreased by 49%. Although most of our patients showed an elevated plasma amylase activity (mean 174.7 IU/L compared with the normal values of up to 160 IU/L) but no significant changes occurred following hemodialysis (mean value 179.1 IU/L).

Since PTH, calcitonin and alkaline phosphatase are involved in bone metabolism, the relationship between

Table II: Plasma parathormone (PTH), calcitonin (Cal) and alkaline phosphatase (ALKP) concentrations in hemodialysed patients classified on the basis of the dialysis period. PTH and Cal concentrations are expressed in ng/mL and pg/mL respectively and the activity of ALKP is indicated in IU/L.

Less than 2 years			2-4 years			more than 4 years		
PTH	Cal	ALKP	PTH	Cal	ALKP	PTH	Cal	ALKP
734	13	51	864	44	99	789	42	384
666	11	58	954	39	175	518	85	91
150	37	32	687	68	90	1129	22	174
351	44	18	551	14	62	163	18	39
459	57	59	709	56	88	341	13	21
179	64	71	458	11	94	431	30	76
245	18	97	320	18	27	323	8.4	44
191	21	24	317	35	46			
288	10.4	70	274	13	64			
389	6.3	123	191	21	24			
166	18	123	454	66	29			
602	19	37	155	20	50			
107	69	51						
235	23	70						
238	12	51						
203	20	24						
323	8.4	44						
264	15	24						
371	32	56						
84	55	19						
77	62	102						
73	110	75						
78	65	118						
34	5.4	24						
266.7	33.1	59.2	487	33.7	74.6	527	31.2	119.8

these parameters and the period of hemodialysis was then investigated. For this purpose the patients were divided into three groups (Table II). The first group were those who had received hemodialysis for less than two years. The mean values for PTH, calcitonin and alkaline phosphatase in this group were 266. 75 ng/dL, (normal: up to 27 ng/dL) 33.1 pg/mL (normal: up to 50 pg/mL) and 59.20 IU/L (normal: 13-46 IU/L) respectively.

The second group of patients were those undergoing dialysis for 2-4 years in which about an 80% increase in PTH (487 ng/dL against 266.75 ng/dL) and 26% increase in alkaline phosphatase activity (74.6 IU/L against 59.2 IU/L) were observed. No significant changes have been seen in the calcitonin level. The results obtained from the third group of patients who were under regular hemodialysis for more than four years showed that although plasma calcitonin level again remained unchanged PTH and alkaline phosphatase activity showed further elevation of 10 and 40 percent respectively in comparison to the second group and almost a 100% increase when compared with the first group of our patients (Table II).

DISCUSSION

The high incidence of osteomalacia in dialysis patients motivated us to conduct a comprehensive survey on plasma bone related parameters in a number of dialysis patients in Isfahan. The results obtained showed that there was a significant increase (20%) in the plasma calcium level following hemodialysis, which may be due to the high concentration of this element in the Isfahan water supply. Kerr et.al. reported that no significant changed occurred in plasma calcium following dialysis in the New Castle dialysis center.[12] Thus in order to regulate the calcium level in plasma of these patients the concentration of this element in water should be controlled prior to the dialysis process. The elevation of plasma calcium however does not seem to be harmful for these patients because of the existence of hypocalcemia. In contrast to the calcium concentration the level of plasma phosphorus was reduced by 45% following hemodialysis. Data from another dialysis center indicates a 5% increase in plasma phosphorus concentration following hemodialysis.[12] This reduction in plasma phosphorus concentration may be a

benefit to the patients who are suffering from hyper-. phosphatemia.

The reduction in plasma calcium due to the failure of the kidney to hydroxylate 25-hydroxycholecalciferol may lead to the stimulation of parathormone secretion which causes bone resorption.[6] Due to the bone resorption process the alkaline phosphatase activity has been elevated significantly in the majority of the patients (Table II).

Amylase activity was elevated in the majority of our patients which is in agreement with the observation of Najjar, et.al.[13]

Although their results are somehow consistent with the present study but the elevation in serum amylase activity was not observed in all of our patients in Isfahan. Hence the mean value for post-dialysis amylase activity remained unchanged when compared with that of pre-dialysis. Amylase is a calcium dependent enzyme the activity of which could be influenced by serum calcium concentration.[14] Therefore, it seems that the slight elevation of serum calcium following hemodialysis may not be enough to influence amylase synthesis. The elevated activity of the enzyme in hemodialysis patients in comparison to the control may be due to the secondary hyperparathyroidism and in turn to altered bone metabolism.

However, more investigation is needed in order to elucidate the exact mechanism by which these changes occur. With regard to the duration of hemodialysis and the simultaneous elevation of serum PTH and alkaline phosphatase activity in our patients and considering the occurrence of different forms of bone disorders in hemodialysis patients including aluminum bone disease, osteitis fibrosa, etc. it may be concluded that the changes observed in PTH and alkaline phosphatase levels are attributed to the alteration in bone metabolism.[15] Observations of Noris, et.al. indicated that PTH and alkaline phosphatase concentrations have been elevated in different forms of bone diseases and this elevation is more significant in osteitis fibrosa than in aluminum bone disease.[15] However more investigation is needed in order to find out the exact relationship between the duration of hemodialysis and the occurrence of bone diseases.

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