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Etiology of hyperparathyroidism and bone disease during chronic hemodialysis: *I. Association of bone disease with potentially etiologic factors*

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Etiology of Hyperparathyroidism and Bone Disease during Chronic Hemodialysis

I. ASSOCIATION OF BONE DISEASE WITH POTENTIALLY ETIOLOGIC FACTORS

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ABSTRACT The present study was prompted by the observation that, in patients with chronic renal failure being followed at this center, renal osteodystrophy developed almost exclusively in those who were treated by chronic hemodialysis at home rather than in our center. A systematic comparison was made between the 10 patients with roentgenographic evidence of the bone disease and 18 patients without demonstrable bone disease. The two groups were similar in age, sex, nature of renal disease, and duration of dialysis. The mean duration of kidney disease was almost 2 yr longer in the patients without bone disease than in those with bone disease. Other significant differences related to where the hemodialysis was performed and to the calcium concentration in the dialysate (6.0–7.4 mg/100 ml in the hospital and 4.9–5.6 mg/100 ml at home). If the unknown factors related to where the dialysis was performed were of no consequence, the major factor contributing to the production of bone disease observed in these patients was the use of a dialysate with a calcium concentration less than 5.7 mg/100 ml.

INTRODUCTION

Bone disease, calcification of soft tissue, and hypercalcemia are among the most disabling and serious complications in patients being maintained on long-term hemodialysis (1–3). Because the incidence of these disorders of calcium metabolism tends to be proportional to the duration of dialysis (2), it has been suggested that these complications may be a consequence of the prolonged duration of renal insufficiency in the surviving patients. The widely different incidence rates at various dialysis centers, however, indicate that there may be other etiologic factors, perhaps within the dialysis programs themselves. No systematic evaluation of possible factors has been reported despite the suggestion by some investigators that the dialysate calcium concentration may be one such determinant (3–7).

The present study was initiated as a result of the observation that, in patients being followed at this center, clinically significant or roentgenographically demonstrable bone disease occurred almost exclusively in patients being dialyzed at home. The study was undertaken to evaluate those factors which we considered as potentially etiologic, both before and during long-term dialysis, in an attempt to determine why the bone disease developed in the one group of patients and not in the other group. The results indicate that (*a*) the observed bone disease may have been a consequence of dialysis per se and not of factors preceding dialysis or of duration of dialysis and (*b*) the major condition favoring development of bone disease was the use of a dialysate with too low a concentration of calcium.

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METHODS

Patient selection and dialysis conditions. The 28 patients in this study were selected from a group of 43 patients being treated by hemodialysis since 1963. Selection was made on the following basis: (*a*) treatment by hemodialysis for 6 months or more, (*b*) complete information available regarding predialysis blood chemical and roentgenographic data, (*c*) adequate information available regarding calcium and magnesium concentrations in the dialysate, and (*d*) absence of severe complications or repeated surgical procedures which immobilized the patient for prolonged periods. No patient had clinical symptoms or roentgenographic evidence of bone disease before starting long-term dialysis. There was no evidence of calcinosis before dialysis in 23 patients; the data were incomplete in this regard in 4, and 1 had roentgenographic evidence of calcinosis of the shoulder. Steroid therapy had been used in only four patients.

External arteriovenous fistulas and modified Kiil dialyzers were used. Blood flow rates averaged 140–250 ml/min; dialysate flow rates averaged 400–500 ml/min. Dialysate was maintained at 37°C and was not recirculated. Total heparinization was the rule.

The selection of patients for dialysis at home or at the center was based on familial, economic, and geographic factors but not on the nature or the severity of the renal disease; 11 patients were treated at the center. From October 1963 to December 1965, dialysate was prepared from ordinary tap water and contained 6–7 mg of calcium and 1–2 mg of magnesium per 100 ml. Only four patients were dialyzed during this period. After January 1966, a commercial concentrate (Mallinckrodt) was used exclusively, and sufficient calcium chloride was added to bring the concentration of calcium to 6.0 mg/100 ml (SD, 0.5) after dilution with deionized water by a central proportioning system. The magnesium concentration was 1.9 mg/100 ml (SD, 0.1). Dialysate fluoride was measured weekly during the last 3 months of the study and was found to be consistently less than 0.5 μmole/liter. The concentration of manganese was 9 ng/ml and that of iron, 37 ng/ml. Dialysate concentrations of calcium, magnesium, manganese, and iron were measured weekly.

Four patients were treated in an affiliated hospital using a similar proportioning system and soft water. The 13 other patients were treated at home. For these patients, the commercial concentrate (which is expected to provide a final dialysate calcium concentration of 5.0 mg/100 ml after dilution) was mixed individually either with a batch system or with a proportioning pump. Dialysate concentrations of calcium, magnesium, manganese, and iron were measured monthly. Dialysate fluoride was measured twice at 15-day intervals for 12 of the patients on home dialysis and was found to be increased (1 ppm or 53 μmoles/liter) in only 4. The patients for whom no fluoride data are available are those who already had had a kidney transplant at the time of the study.

The only routine medication was a daily multivitamin preparation containing 400 IU of vitamin D. None of the patients was given phosphate-binding agents. Dietary calcium intake was evaluated on the basis of a 7 day dietary history.[1]

Analytical methods. Before the start of long-term dialysis and once monthly during the entire period of dialysis, the following analyses were performed on blood drawn from the cannula before dialysis: (*a*) serum creatinine by the method of Folin and Wu (8) adapted for the AutoAnalyzer, (*b*) serum phosphate by the method of Fiske and SubbaRow (9) adapted for the AutoAnalyzer, and (*c*) total serum calcium and magnesium by complexometric titration (10). Dialysate concentrations of manganese and iron were measured by atomic absorption flame spectrophotometry (11).

Acid-base balance was evaluated bimonthly by the method of Astrup, Andersen, Jørgensen, and Engel (12) on arterial blood drawn before dialysis. Serum protein electrophoresis was performed every 3 months. Serum concentration of fluoride was assessed by a modified diffusion method in 18 patients at the time of their last evaluation (13). The mean value observed in normal persons drinking water containing fluoride at 1 ppm is 0.7 μmole/liter (SD, 0.4); the average difference between duplicate determinations is 0.19 μmole/liter (14). Serum alkaline phosphatase was measured by the method of Kind and King (15) every 3 months. Values were considered as suggestive of bone disease when they were more than 60 U/liter without a concomitant increase in serum glutamic oxaloacetic transaminase.

RESULTS

Factors possibly influencing occurrence of bone disease. The patients were divided into two groups: those with roentgenographic evidence of bone disease and those without such evidence. Clinical symptoms were not taken into account in assigning the patients to the groups. The roentgenographic abnormalities which were considered to represent definite evidence of bone disease were fractures and subperiosteal resorption. Minor changes in bone density were considered to be too subjective.

TABLE I
Characteristics of Groups before Starting Long-Term Dialysis

	Group with bone disease	Group without bone disease
No. of patients	10	18
Age, mean ±SD (*yr*)	33.5 ±13	33.4 ± 9
Sex ratio (males/females)	6/4	11/7
Chronic glomerulonephritis/other renal disease	6/4	11/7
Duration of kidney disease, mean ±SD (*yr*)	11.2 ±13.4	33.4 ±18*
Duration of azotemia, mean ±SD (*yr*)	2.2 ± 3.4	2.5 ±3.6
Serum calcium, mean ±SD (*mg/100 ml*)	7.9 ±1.5	7.7 ±1.3
Serum phosphate, mean ±SD (*mg/100 ml*)	8.2 ±2.6	8.8 ±2.7
Serum alkaline phosphatase, mean ±SD (*U/liter*)	39.1 ±12	45.0 ±27
Roentgenographic evidence of bone disease (no.)	0	0

* For difference between groups, $P < 0.01$.

[1] We appreciate the assistance of Mrs. Joyce D. Margie, who evaluated the dietary calcium of these patients.

TABLE II

Possible Etiologic Factors of Bone Disease during Long-Term Hemodialysis

Etiologic factor	Groups	
	With bone disease	Without bone disease
Dietary calcium, mean ±SD (*mg/day*)	311 ±107	369 ±150
Vitamin D (*IU/day*)	400	400
Phosphate-binding agents	None	None
Corticosteroids (no.)	1/10	3/18
Bilateral nephrectomy (no.)	3/10	6/18
Duration of dialysis, mean ±SD (*month*)	18.6 ±5.0	21.0 ±5.0
Total heparin, mean and range ($U \times 10^6$)	30.12 (14–57)	33.99 (6.5–106)
Predialysis serum creatinine, mean ±SD (*mg/100 ml*)	11.9 ±2.0	12 ±2.0
Predialysis arterial pH, mean ±SD	7.38 ±0.03	7.37 ±0.03
Predialysis standard HCO_3^-, mean ±SD (*mEq/liter*)	21.0 ±2.2	20.4 ±1.6
Using deionized water (no.)		
Regardless of dialysate Ca	2/10	12/18*
Low dialysate Ca only	2/9	1/7
Dialysate Mn, mean and range (*ng/ml*)	13.6 (5–19)	11.06 (6–24)
Dialysate Fe, mean and range (*ng/ml*)	62.4 (28–117)	52.6 (27–232)
Dialysate Mg, mean ±SD (*mg/100 ml*)	1.9 ±1	1.9 ±1
Dialysate Ca		
Range (*mg/100 ml*)	4.9–5.6	5.1–7.4‡
≦5.6 mg/100 ml (no.)	9/10	7/18‡
Dialysate F, range (*μmoles/liter*)		
All patients	5–53	0.5–63§
Low dialysate Ca only	5–52	1–63
Dialysate F, >6 μmoles/liter		
All patients	4/7	2/11
Low dialysate Ca only	3/6	2/5
Serum F, range (*μmoles/liter*)		
All patients	1–36	0–10.2
Low dialysate Ca only	1–10.5	0–10.2

* For difference between groups, $P < 0.02$.
‡ For difference between groups, $P < 0.05$.
§ For difference between groups, $P = 0.05$.

Before starting long-term dialysis, these two groups were comparable in terms of age, sex ratio, nature of the kidney disease, duration of azotemia, and the serum concentrations of calcium, phosphate, and alkaline phosphatase (Table I). The only significant difference was in the duration of kidney disease; since this was longer in the group without bone disease, it cannot be incriminated as etiologic in the bone disease.

In Table II the two groups are compared in regard to possible etiologic factors during the period of dialysis. There was no significant difference between the groups in regard to dietary calcium intake, vitamin D intake, phosphate-binding agents, steroid therapy, duration of dialysis, total amount of heparin received, adequacy of dialysis (assessed by mean predialysis[2] serum creatinine), mean predialysis pH and standard bicarbonate, and dialysate concentration of magnesium, manganese, and iron. No significant association was observed between bilateral nephrectomy and bone disease.

A significant difference between the two groups was found in regard to dialysate calcium concentration; it was significantly lower ($P < 0.05$, by the rank sum test) in the group with bone disease. All but one of the 10 patients with bone disease used a dialysate with calcium concentration ≦ 5.6 mg/100 ml; the single exception was the only patient in this series whose serum fluoride concentration was in the toxic range. In contrast, the majority of the patients (11 of 18) without bone disease were exposed to a dialysate calcium concentration greater than 5.6 mg/100 ml. These two proportions, 1/10 and 11/18, differ significantly ($P < 0.05$).

In regard to the dialysate fluoride concentration, the difference between the two groups was only of borderline significance ($P = 0.05$, by the rank sum test); the group with bone disease had been exposed to a higher level of dialysate fluoride. However, this observation per se is misleading because only one of the patients dialyzed against the higher calcium concentration was exposed to the high dialysate fluoride concentration. When only patients dialyzed against calcium concentrations ≦ 5.6 mg/100 ml are considered, there is no longer a significant difference between the groups with and without bone disease; actually, the mean dialysate fluoride is then higher in the group without bone disease. A chi square analysis was performed after the continuous variable, dialysate fluoride, was transformed into a qualitative one, using a concentration of 6 μmoles/liter for separation. There was no relationship between high dialysate fluoride level (> 6 μmoles/liter) and bone disease for the group of patients dialyzed against a low dialysate calcium concentration. In the group dialyzed against a high calcium concentration, the only patient who developed bone disease was dialyzed against a very high fluoride level.

As regards the treatment of the water used for the dialysate, use of deionized water was significantly associated with bone disease. This association was no longer significant when only the low dialysate calcium group was considered.

Dialysate calcium concentration and incidence of calcification disorders. Because in these patients the dialysate calcium concentration appeared to be the most important factor affecting the development of bone disease, its role in the occurrence of clinical symptoms, calcinosis, and increased serum alkaline phosphatase

[2] In this report, the term "predialysis" means immediately before a dialysis and does not refer to the period before the long-term dialysis regimen was begun.

was assessed by comparing the group of 16 patients dialyzed against a low calcium concentration (4.9–5.6 mg/100 ml) with the group of 12 patients dialyzed against a high calcium concentration (6.0–7.4 mg/100 ml) (Table III).

Clinical symptoms were observed in only one patient of the group dialyzed against a high calcium concentration, whereas they were present in 12 patients of the group dialyzed against a low calcium concentration ($P < 0.001$). Calcinosis was found on the skeletal survey performed at the time of the final examination in one patient of the high calcium group and in five patients of the low calcium group. Because no roentgenograms of the involved areas before long-term hemodialysis were available for four of these patients, it cannot be stated with assurance that this calcinosis developed during dialysis. It should be stressed that the soft tissue calcifications observed in this study were small. In one patient dialyzed against a calcium concentration of 6.0 mg/100 ml, the subacromial calcification of the left shoulder observed before dialysis was absent at the last evaluation. Band keratopathy, assessed by slit lamp examination of the eyes at the time of the final evaluation, was found in six of the patients in the low calcium group and in none of the high calcium group. The difference in over-all incidence of calcinosis at the time of final evaluation, one in the high calcium group and seven in the low calcium group, is of only borderline significance by chi square analysis ($P < 0.10$).

Other possible factors in the development of calcinosis were investigated only in the patients dialyzed against the low calcium concentration because most of the cases occurred in that group (Table IV). There were two statistically significant differences: those who developed calcinosis had a lower mean serum fluoride concentration and a higher mean serum calcium concentration before starting the dialysis program. With one exception, only those who had serum fluoride concentrations less than 3.3 μmoles/liter had evidence of calcinosis.

An increase in serum alkaline phosphatase from normal to abnormal values (> 60 U/liter) was observed during the dialysis period in 3 patients of the high dialysate calcium group and in 10 of the low dialysate calcium group; the increase was at least 16 U/liter. The distribution was different only at a borderline level of significance ($P < 0.10$, by chi square).

No evidence of any disorder of calcification was observed in 7 of the 12 patients in the high calcium group and in only 1 of the 16 in the low calcium group ($P < 0.05$). It is worth noting that this latter patient was the patient dialyzed for the shortest period (6 months).

Relationship between occurrence of bone disease and serum concentrations of total protein, albumin, calcium

TABLE III
Influence of Dialysate Calcium Concentration on Incidence of Calcification Disorders

Calcification disorders	High Ca (6.0–7.4 mg/100 ml)	Low Ca (4.9–5.6 mg/100 ml)	P (chi square)
Patients (no.)	12	18	
Radiographic evidence of bone disease	1	9	<0.05
Clinical symptoms	1	12	<0.001
Chest wall pain	1	7	<0.10
Podagra	0	3	
Arthralgia	1	3	
Severe pruritus	0	2	
Increased alkaline phosphatase	3	10	<0.10
Calcinosis	1	7	<0.10
Roentgenographic	1	5	
Band keratopathy	0	6	
None observed	7	1	<0.05

TABLE IV
Comparison of Patients With and Without Calcinosis

Variable	Calcinosis: With	Calcinosis: Without
Serum creatine (*mg/100 ml*)	12.4	11.6
Serum F (*μmoles/liter*)	2.2	6.1*
pH	7.382	7.385
HCO_3^- (*mEq/liter*)	21.0	21.6
Total protein (*g/100 ml*)	5.7	6.0
Albumin (*g/100 ml*)	2.9	3.1
[Ca] (*mg/100 ml*)		
Before dialysis	8.5	6.5*
First 3 months	9.4	9.1
Last 3 months	9.4	9.1
[P] (*mg/100 ml*)		
Before dialysis	7.8	9.0
First 3 months	7.5	6.7
Last 3 months	6.6	7.9
[Mg] (*mg/100 ml*)		
Before dialysis	2.7	3.7
First 3 months	3.1	3.5
Last 3 months	3.5	3.4
Alkaline phosphatase (*U/liter*)		
Before dialysis	40.5	35.0
First 3 months	40.2	39.5
Last 3 months	66.4	57.6
[Ca] × [P]		
Before dialysis	64	56
First 3 months	70	59
Last 3 months	63	70

* For difference between groups, $P < 0.05$.

TABLE V

Relationship of Dialysate Calcium Concentration and Occurrence of Bone Disease to Serum Concentrations of Total Protein, Albumin, Calcium, Phosphate, and Magnesium

	Serum concentrations*				
	Total protein	Albumin	Ca	P	Mg
	(g/100 ml)	*(g/100 ml)*	*(mg/100 ml)*	*(mg/100 ml)*	*(mg/100 ml)*
Dialysate calcium					
4.9–5.6 mg/100 ml	5.7	3.0	9.1	7.4	3.3
	(4.9–6.6)	(2.5–3.5)	(7.4–10.5)	(4.8–9.6)	(2.6–4.5)
6.0–7.4 mg/100 ml	5.7	3.0	8.3	6.6	3.4
	(5.0–6.3)	(2.6–3.4)	(6.7– 9.3)	(5.2–9.0)	(2.9–3.9)
Significance‡	NS	NS	$P < 0.01$	NS	NS
Bone disease					
Present	5.7	2.9	9.4	7.3	3.2
	(4.9–6.6)	(2.5–3.5)	(8.3–9.9)	(6.3–8.3)	(2.6–3.8)
Absent	5.8	3.0	8.5	6.9	3.4
	(5.0–6.3)	(2.6–3.4)	(6.7–9.9)	(4.8–9.6)	(2.8–4.5)
Significance‡	NS		$P < 0.01$	$P < 0.10$	NS

* Mean and range. These are predialysis values.

‡ Rank sum test.

([Ca]), phosphate ([P]), and magnesium ([Mg]). Serum total protein and serum albumin concentrations were comparable in the low dialysate calcium group and the high dialysate calcium group (Table V). Before start of the dialysis regimen, there was no significant difference in [Ca] between the groups, although the mean [Ca] was slightly lower in the group subsequently dialyzed against a low calcium concentration. The mean predialysis [Ca] for the entire period of dialysis was significantly higher ($P < 0.01$) in the low dialysate calcium group. This significant difference also was found during the first 3 months of dialysis. Only one patient had hypercalcemia ([Ca] > 10.1 mg/100 ml) during the last 3 months of dialysis. No significant difference in [P] existed between the two groups either before or during dialysis. There was a slight tendency (not significant) for [P] to decrease in the last 3 months in the high dialysate calcium group. The only statistically significant difference in [Ca] × [P] product was that the low dialysate calcium group had a higher product in the final 3 months.

Serum concentrations of total protein and albumin were comparable between the group with bone disease and the group without bone disease. The mean [Ca] for the entire period of dialysis was significantly higher ($P < 0.01$) in the group with bone disease although no significant difference was observed between the two groups before starting dialysis. The mean [P] for the entire period of dialysis was higher in the group with bone disease but only at a borderline level of significance ($P < 0.10$). The difference in mean [Mg] was not significant between the groups.

DISCUSSION

The present study attempted to evaluate those factors which might favor the development of bone disease and extraskeletal calcinosis in patients with chronic renal disease who were undergoing long-term dialysis. None of the 28 patients in this study had roentgenographic evidence of bone disease before beginning dialysis, but 10 manifested evidence (fractures or subperiosteal resorption) of bone disease during the course of 6–60 months of hemodialysis. Because there were no significant differences in predialysis status between the 10 in whom bone disease developed and the 18 in whom it did not, one or more factors operative during dialysis must be implicated. Since duration of dialysis was not significantly different between the groups with and without bone disease, this bone disease can no longer be considered to be a consequence of prolonged survival of the patients, but it may be a complication of long-term dialysis per se.

The concentrations of calcium and fluoride in the dialysate and the nature of water treatment (use of a deionizer vs. use of a water softener) were the only factors which yielded statistically significant differences between the two groups. Because the patients who developed bone disease were exposed to a dialysate of significantly lower calcium concentration and, reciprocally, because the patients dialyzed against a low calcium concentration had a significantly higher incidence of bone disease, it appears that a low dialysate calcium concentration favors development of bone disease whereas a

dialysate calcium concentration of 6 mg/100 ml or more may be protective.

Because all but one of the patients exposed to dialysate of high fluoride concentration were in the low calcium dialysate group and because most of the patients in the low dialysate calcium group used softened water, the influence of dialysate calcium level is confounded with both that of dialysate fluoride and that of the nature of water treatment when the whole group is considered. Statistical analysis of the correlation of bone disease with dialysate fluoride and the nature of water treatment in the low dialysate calcium group did not demonstrate significant association. However, there were only four patients whose dialysate fluoride concentrations were greater than 14 μmoles/liter (0.3 ppm). Two of these did not have bone disease and had been treated by dialysis only 6 and 12 months. Furthermore, of these four patients, one was the only patient who was exposed to a high dialysate calcium concentration and had bone disease. Thus, the present data do not conflict with the conclusion that fluoride was complicating the renal osteodystrophy seen in the patients at Ottawa General Hospital who had been treated by fluoridated dialysis for 1–3 yr (dialysate calcium concentration, 6 mg/100 ml).[3]

It is not surprising that the lower dialysate calcium concentrations in this study were associated with a significant incidence of bone disease because one would expect that both calcium balance and secretion of parathyroid hormone (PTH) during dialysis should be responsive to the calcium concentration of the dialysate. It has been reported (4, 5, 7, 16) that calcium balance during dialysis is positive when the dialysate calcium concentration is greater than the diffusible fraction of serum calcium. Although diffusible serum calcium was not measured in the present study, it can be estimated from the total calcium (5). Positive balance would be predicted in 14 of the 18 patients without bone disease but in only 3 of the 10 with bone disease ($P < 0.05$). Gain of calcium during dialysis assumes major importance because of the demonstrated negative calcium balance between dialyses in patients on long-term hemodialysis (5, 7). The patients are unable to absorb calcium normally from the gastrointestinal tract, apparently as a result of a disturbance in the metabolism of vitamin D (17). Thus, it is essential to their long-term calcium balance that the losses be counterbalanced by gains during dialysis.

Parathyroid function is likewise affected by the calcium concentration of the dialysate. Secretion of PTH may not be suppressed during dialysis against a low calcium concentration so that a greater degree of parathyroid hyperplasia would be expected in the low calcium group. The apparent paradox of the predialysis serum calcium concentration being significantly higher in the low calcium group than in the high calcium group may be explained by the lack of adequate suppression of PTH secretion during dialysis and the resultant increase in basal level of plasma PTH. This interpretation agrees with the previous observation by Stanbury (18) that serum calcium concentration was significantly higher in uremic patients with the greatest parathyroid hyperplasia. The fact that patients with bone disease have significantly higher predialysis serum calcium values suggests that they have more severe parathyroid hyperfunction and that this is one pathophysiologic mechanism of the bone disease.

Extraskeletal calcification was seen almost exclusively in the patients dialyzed against a low calcium concentration. Because, in most of the cases, absence of calcinosis was not documented before the start of long-term dialysis, it cannot be stated with assurance that low dialysate calcium favors calcinosis. On the other hand, because a dialysate calcium concentration of 6 mg/100 ml was associated with only one case of vascular calcification and with the disappearance of shoulder calcification in another case, use of this calcium level appears to be safe in regard to the hazard of metastatic calcification. This may not be the case with calcium concentrations greater than 6.5 mg/100 ml, which have been reported to be associated with a high incidence of calcinosis (3, 19, 20).

Since in the low dialysate calcium group there was a significant difference, between patients with and without calcinosis, in the serum calcium concentration before the start of long-term dialysis, it is possible that calcinosis was predetermined or had begun before dialysis. However, the significant negative correlation between calcinosis and serum fluoride concentration suggests that a high serum fluoride level may inhibit soft tissue calcification. This finding is similar to that of Bernstein, Sadowsky, Hegsted, Guri, and Stare (21) in residents of the Dakotas, where high fluoride intake was associated with less aortic calcification.

It is concluded that bone disease is a complication of long-term hemodialysis per se and that the major factor favoring bone disease is the use of too low a calcium concentration in the dialysate (< 5.7 mg/100 ml). Because it has been reported that a dialysate calcium level of 6.5 mg/100 ml or greater may be associated with complications such as hypercalcemia (22) and calcinosis (3, 19, 20), the optimal range for dialysate calcium concentration may be between 5.7 and 6.5 mg/100 ml. It is possible, however, that patients whose serum calcium concentration has already begun to increase, as a result of secondary hyperparathyroidism, may require higher initial concentrations of dialysate calcium in order to

[3] Posen, G. A., D. R. Taves, J. R. Marier, and Z. F. Jaworski. 1968. Read at the meeting of the American Society of Nephrology, Washington, D. C., November, 1968.

suppress secretion of PTH and to prevent negative calcium balance during dialysis.

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REFERENCES

1. Pendras, J. P., and R. V. Erickson. 1966. Hemodialysis: a successful therapy for chronic uremia. *Ann. Intern. Med.* **64**: 293.
2. Kim, D., N. H. Bell, W. Bundesen, P. Putong, N. M. Simon, C. Walker, and F. del Greco. 1968. Renal osteodystrophy in course of periodic dialysis for chronic uremia. *Trans. Amer. Soc. Artif. Intern. Organs.* **14**: 367.
3. Traeger, J., P. Zech, J. P. Bernhardt, A. Frederich, J. F. Moskovtchenko, P. Meunier, N. Pozet, and G. Laurent. 1968. Le métabolisme calcique chez les malades en dialyse chronique. *Actual. Nephrol. Hop. Necker.* 87.
4. Verberckmoes, R. 1967. Dosage de la fraction ultrafiltrable du calcium par une méthode utilisant le rein artificiel de type twin coil. *Proc. Conf. Replacement Renal Funct.* **3**: 28.
5. Kaye, M., R. Mangel, and E. Neubauer. 1967. Studies in calcium metabolism in patients on chronic haemodialysis. *Proc. Conf. Replacement Renal Funct.* **3**: 17.
6. Ritz, E., H. E. Franz, and E. Jahns. 1968. The course of secondary hyperparathyroidism during chronic hemodialysis. *Trans. Amer. Soc. Artif. Intern. Organs.* **14**: 385.
7. Wing, A. J. 1968. Optimum calcium concentration of dialysis fluid for maintenance haemodialysis. *Brit. Med. J.* **4**: 145.
8. Folin, O., and H. Wu. 1919. A system of blood analysis. *J. Biol. Chem.* **38**: 81.
9. Fiske, C. H., and R. SubbaRow. 1925. The colorimetric determination of phosphorus. *J. Biol. Chem.* **66**: 375.
10. Jones, J. D., and W. F. McGuckin. 1964. Complexometric titration of calcium and magnesium by a semiautomated procedure. *Clin. Chem.* **10**: 767.
11. Slavin, W. 1968. Atomic Absorption Spectroscopy. Interscience Publishers, Inc., New York.
12. Astrup, P., O. S. Andersen, K. Jørgensen, and K. Engel. 1960. The acid-base metabolism: a new approach. *Lancet.* **1**: 1035.
13. Fry, B. W., and D. R. Taves. 1970. Serum fluoride analysis with the fluoride electrode. *J. Lab. Clin. Med.* **75**: 1020.
14. Taves, D. R. 1966. Normal human serum fluoride concentrations. *Nature (London).* **211**: 192.
15. Kind, P. R. N., and E. J. King. 1954. Estimation of plasma phosphatase by determination of hydrolysed phenol with amino-antipyrine. *J. Clin. Pathol. (London).* **7**: 322.
16. Ogden, D. A., and J. H. Holmes. 1966. Changes in total and ultrafilterable plasma calcium and magnesium during hemodialysis. *Trans. Amer. Soc. Artif. Intern. Organs.* **12**: 200.
17. Avioli, L. V., S. Birge, S. W. Lee, and E. Slatopolsky. 1968. The metabolic fate of vitamin D_3-^3H in chronic renal failure. *J. Clin. Invest.* **47**: 2239.
18. Stanbury, S. W. 1968. Bone disease in uremia. *Amer. J. Med.* **44**: 714.
19. Parfitt, A. M., A. C. Winfield, J. De Palma, A. Gordon, and S. G. Massry. 1969. Calcium and phosphorus metabolism during intermittent hemodialysis for uremia: effect of high dialysate calcium concentration. *Proc. Int. Congr. Nephrol. 4th.* (Abstr.)
20. Vantelon, J., D. Perrin, J. Zingraff, and J. L. Funck-Brentano. 1967. Calcifications kérato-conjonctivales au cours des traitements par hémodialyse périodique. *Proc. Conf. Replacement Renal Funct.* **3**: 24.
21. Bernstein, D. S., N. Sadowsky, D. M. Hegsted, C. D. Guri, and F. J. Stare. 1966. Prevalence of osteoporosis in high- and low-fluoride areas in North Dakota. *J. Amer. Med. Ass.* **198**: 499.
22. Freeman, R. M., R. L. Lawton, and M. A. Chamberlain. 1967. Hard-water syndrome. *N. Engl. J. Med.* **276**: 1113.