RENAL OSTEODYSTROPHY IN PATIENTS ON LONG-TERM HEMODIALYSIS WITH FLUORIDATED WATER

by

G. A. Posen, J. R. Marier and Z. F. Jaworski
Ottawa, Canada

SUMMARY: Serum and bone fluoride concentrations of ten patients maintained on long-term hemodialysis with fluoridated water (1 ppm, i.e., 50 μM) were correlated with duration of treatment and the occurrence of clinical, radiological, and histological manifestations of bone disease. Two patients had symptomatic renal osteodystrophy when accepted on the program, whereas six others developed the disease within a year of fluoridated dialysis. However, in all patients, the disease progressed despite recommended therapy (including high doses of vitamin D). The mean pre-dialysis serum fluoride level was 16 ± 4 μM which rose to 28 ±3 post-dialysis. The bone fluoride content ranged from 800 to 22,500 ppm on a dry fat-free basis. Toxic effects have been reported at these levels and could complicate underlying renal osteodystrophy. Further studies are required to delineate the role of fluoride in this condition.

In our experience, renal osteodystrophy has been a common and disabling complication of maintenance hemodialysis. Some investigators (1, 2, 3) have reported improvement of this condition by treatment with various doses of vitamin D, calcium supplements, and phosphorus-binding gels. However, the bone lesions in our patients have developed or continued to progress despite these measures.

In our hemodialysis center, opened in April 1964, fluoridated dialysis began with the fluoridation of the city water supply in November 1965. Our subsequent therapeutic failure was completely unexpected and a possible explanation was suggested by the observation of Taves et al. (4, 5) that the serum fluoride (i.e., F^-) levels in patients chronically hemodialysed with fluoridated water are comparable to those that cause fluorotic bone disease (6). Thus, the study of fluoride levels in our patients became of particular interest because several of them had been on fluoridated dialysis for much longer periods than those patients reported by Taves et al. (4, 5).

From the Dept. of Medicine, Ottawa General Hospital, and University of Ottawa; also, Division of Biology, National Research Council, Ottawa, Canada.

***

Presented at the Second Annual Conference of I.S.F.R., Barcelona, 1/27-28/69.

During hemodialysis with fluoridated water, serum fluoride uptake comes about because fluoridated water contains about 50 μM (1.0 ppm) of fluoride, whereas human serum normally contains only about 1 μM (0.02 ppm) of free fluoride ion (5). Thus, there can be a 50:1 gradient favoring diffusion of fluoride ion from the dialysis water into the patient's blood, and ensuing deposition of fluoride into bones increasing with the duration of fluoridated dialysis.

Materials and Methods

The study group comprised ten patients (Table 1) dialysed with fluoridated water for periods ranging from one to thirty-one months. Three of the patients (G.B., R.N., and C.W.) were treated for ten to twelve months with non-fluoridated dialysate prior to fluoridation of the Ottawa water supply. The patients ranged in age from 16 to 61 years; four were female and six were male; eight had glomerulonephritis as their primary diagnosis and two had polycystic disease.

TABLE 1

Patient	Age	Sex	Diagnosis	Duration Dialysis (months)	
				Non F	F*
1. G.B.	28	F	G.N.	10	30.5
2. C.W.	50	M	P.C.	10	31
3. R.N.	36	M	G.N.	12	30
4. L.C.	42	M	G.N.	0	28
5. W.B.	25	M	G.N.	0	26
6. Y.L.	17	F	G.N.	0	16
7. R.V.	40	F	P.C.	0	12
8. D.C.	16	F	G.N.	0	9
9. H.R.	48	M	G.N.	0	1
10. A.D.	61	M	G.N.	0	1

* = When clinical tests were made
G.N. = Glomerulonephritis
P.C. = Polycystic disease

The patients were dialysed on a single-pass Kiil system. The duration of each dialysis varied from eight-to-ten hours, three times a week, to twelve-to-fourteen hours twice a week. Thus, the program involved a minimum of twenty-four hours of fluoridated dialysis per week. The dialysate contained 3 meq calcium and 1.5 meq magnesium per liter. The fluoride content of the dialysis water was between 0.9 and 1.0 ppm, i.e., 50 μM, after November 1965.

In September 1967, all patients began receiving phosphorus-binding gels, 800 mg of elemental calcium, and 5,000 units of vitamin D per day. In four

patients (G. B., R. N., R. V., and C. W.) who were becoming disabled by their bone disease, vitamin D was increased in 50,000-unit increments to a total of 200,000 units daily.

The skeletal status of our patients was studied before the initiation of dialysis and then serially by biochemical, histological and radiological methods. Serial radiological bone surveys, including skull, clavicles, ribs, vertebrae, pelvis, hands, and long bones, were performed before the patients were placed on the hemodialysis program and at 6-month intervals thereafter.

Serum and dialysate calcium was measured by atomic absorption spectrophotometry, serum phosphorus by a standard photocolorimetric procedure, and serum alkaline phosphatase by the Bessy-Lowry method.

Serum ionic fluoride was measured by Taves' diffusion method (7, 8) modified by releasing fluoride directly from serum by means of hexamethyldisiloxane in HCl (9). The fluoride was trapped in sodium bicarbonate, and measured by a decrease in fluorescence of a morin-thorium complex. Serum fluoride levels were determined before and after dialysis treatments. The approximate uptake of fluoride during a dialysis was determined by averaging the change in serum fluoride levels entering and leaving the dialyser, and multiplying this by the blood-flow and the length of time of the dialysis. The approximate value multiplied by 0.8 gives the fluoride uptake. The value of 0.8 is the correction found necessary when applying this method to patient D. M. (5), where a more accurate graphical integration method could be applied. A correction is needed because the arterio-venous difference is a non-linear function with time.

Trephine biopsies of the iliac crest were obtained under local anesthesia in 7 patients and prepared for histological examination in the undecalcified state by the method of Bohatirchuk (10).

Bone fluoride levels were estimated by the Marier and Rose Zr-SPADNS method (11), but with the volumes scaled-down to permit determination of fluoride in the range of 0 to 4 μg per test. This modification was necessary because of the small size of the bone-biopsy samples (1.0 to 3.5 mg), and involved the use of a 2.0 ml aliquot of test solution along with 0.4 ml additions of the two reagents; in all other respects, the procedure was as previously described (11). The procedure involves drying and defatting of the tissue, ashing in the presence of 200 mg of MgO fixative (containing 0.003% F), microdistillation, then colorimetric estimation. With each group of samples, known fluoride standards containing the MgO fixative were processed simultaneously through all phases of the procedure, so as to correct for any variation in the "baseline" fluoride correction substracted from the total result. Preliminary studies had indicated an accuracy ranging between 87 and 123%, averaging 99%. Although the small size of the bone samples precluded duplicate ashings etc., the 15 ml volume of distillate was large enough to allow replicate colorimetric determinations, and each value reported in this paper represents an average of (at least) triplicate determinations.

Results

The patients' serum calcium, phosphorus, alkaline phosphatase, and Ca x P products, obtained before and after the initiation of the dialysis program, are shown in Table 2.

TABLE 2

Patient	Calcium(Mg%) B	Calcium(Mg%) A	Phosphorus (Mg%) B	Phosphorus (Mg%) A	Alk. Phos.(Bl) B	Alk. Phos.(Bl) A	Post-treatment Ca x P Product
1. G.B.	8.4	9.1	6.3	6.8	2.5	2.4	61.9
2. C.W.	7.0	10.1	5.7	5.0	1.2	3.5	60.6
3. R.N.	7.2	8.6	12.7	9.0	2.5	2.4	77.4
4. L.C.	8.9	10.0	9.0	6.6	1.2	2.7	66.0
5. W.B.	6.7	9.5	7.0	5.0	1.8	3.6	47.5
6. Y.L.	3.5	9.5	13.5	7.9	1.6	3.1	75.1
7. R.V.	7.6	9.4	7.5	4.5	5.6	4.5	42.3
8. D.C.	7.2	8.9	8.5	7.9	1.9	2.7	70.3
9. H.R.	9.8	9.5	3.8	4.9	3.5	2.8	46.6
10. A.D.	9.0	9.6	5.6	5.5	2.6	2.3	52.8

B = Before dialysis program
A = After dialysis program and treatment with Ca, Vit. D. and P-binding gels
Normal values: Ca 8.9 to 10.3 mg%; P 2.8 to 4.5 mg%; alkaline phosphatase 0.8 to 2.3 BU (Bessy-Lowry Units); Ca x P Product = 24.9 to 46.3.

Prior to dialysis treatment, the serum calcium concentrations were subnormal in all except two patients (A. D. and H. R.). These levels became normal within six months after starting treatment, except for R. N. whose calcium level, although increased, was below the lower limit of normal. The serum phosphorus levels, which were elevated prior to treatment in all except patient H. R., fell after starting the dialysis therapy; however, only in R. V. did it reach the normal range. The alkaline phosphatase values were normal in five of the ten cases at the beginning of the dialysis program; but after one year of treatment all except one were above the upper limit of the normal range. Similarly, all but one of the Ca x P products remained higher than normal after dialysis therapy.

On radiological examination (Table 3), eight of the ten patients showed varying degrees of hypomineralization, seven had looser zones, two had subperiosteal reabsorption, and one had osteosclerosis. These radiological changes were associated with bone pains, arthralgias and fractures in seven of the patients.

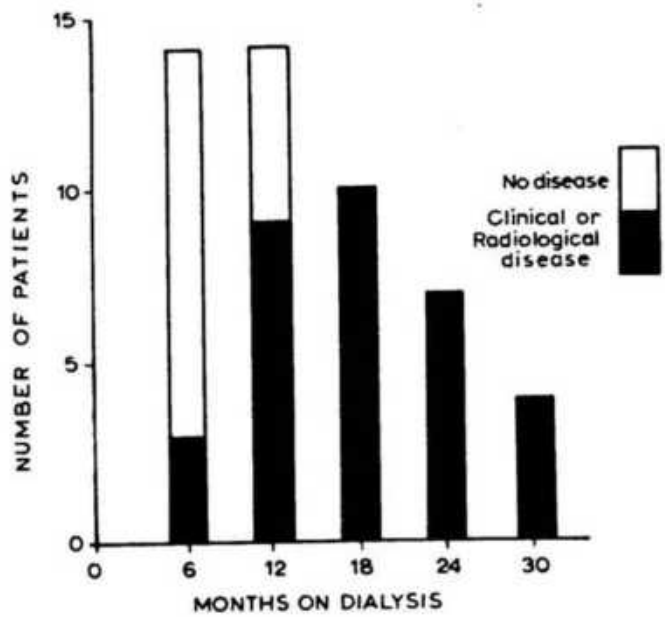
TABLE 3

Patients	Radiological Disease				Clinical Features		
	Hypomineralization	Looser Zones	Sub-Periosteal Reabsorption	Osteosclerosis	Bone Pain	Arthralgia	Fractures
1. G. B.	+	++	-	-	++	++	++
2. C. W.	++	+++	-	-	+++	+++	+++
3. R. N.	++	+++	-	-	+++	+++	+++
4. L. C.	++	+	-	-	++	++	+
5. W. R.	++	+	-	-	++	+++	+
6. Y. L.	++	++	-	-	+++	+	+++
7. R. V.	++	++	+	-	+++	+	+
8. D. C.	-	-	++	+	-	-	-
9. H. R.	-	-	-	-	-	-	-
10. A. D.	+	-	-	-	-	-	-

- Absent; + mild; ++ moderate; +++ severe

Fig. 1

Frequency of Bone Lesions in the Dialysis Program
General Hospital, Ottawa, Canada

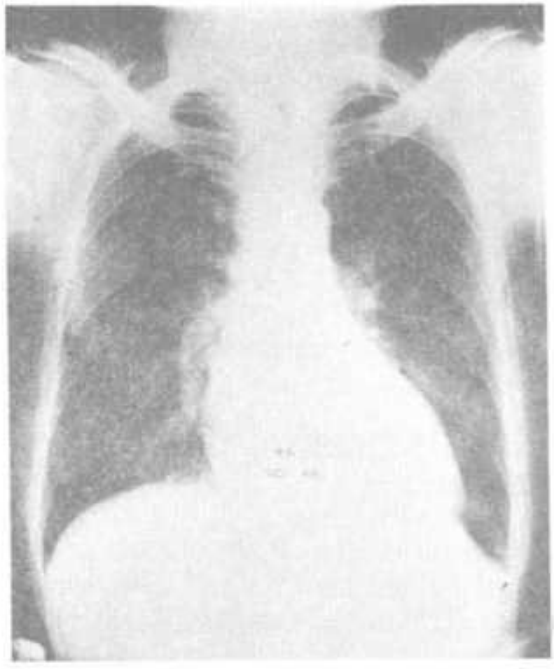


The frequency of clinical and radiological evidence of bone disease at various times after starting dialysis is depicted in Figure 1. There was a progressive increase in the frequency of bone disease with the passage of time until, after eighteen months of dialysis, evidence of bone disease was present in all patients.

Figures 2 and 3 show the radiological progression of the bone disease in patient R. N., who was on dialysis since November 1964. The patient was dialyzed for approximately two years before he developed signs of bone disease (Fig. 2).

Fig. 2

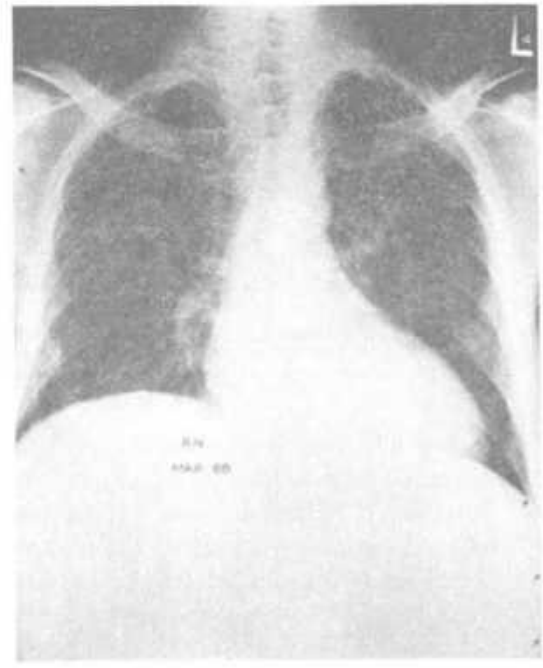
Chest Film, Patient R.N. (Nov. '66)



Early looser zone in 9th lt. rib, but no loss of alignment, no periosteal reaction at site of the radiolucent band.

Fig. 3

Same Patient (March '68), Progression of the Osteodystrophy with Looser Zones in Numerous Ribs, Including Lt. 6th, 8th (2 lesions), 9th and 10th Ribs.

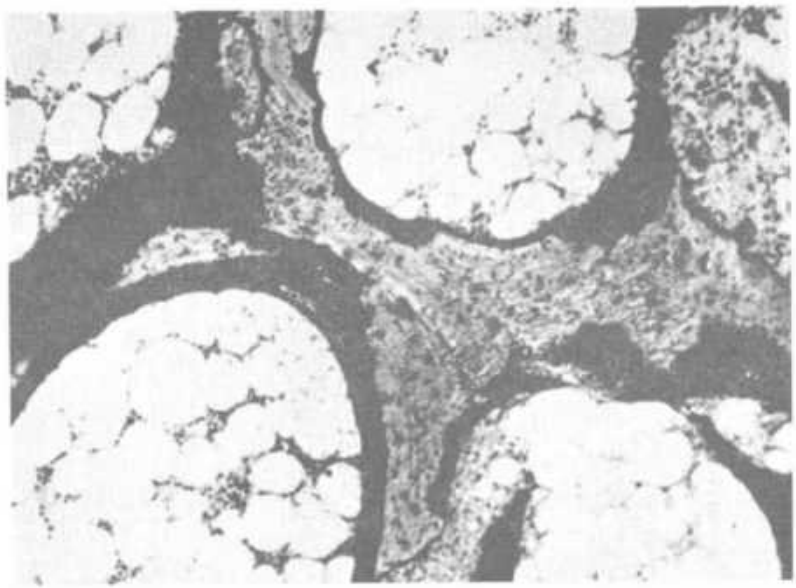


Several ribs show slight displacement, indicating true fracture through the looser zone, but no evidence of healing with periosteal new bone although granular calcification is present adjacent to bone lesion. Calcification in the right rotator cuff.

However, Ottawa water was not fluoridated until November 1965, so that patient R. N. was dialyzed with fluoridated water for only one year. Significantly, unlike our other patients who developed bone disease after only one year of dialysis, he did not show signs of disease for two years. The patient was placed on vitamin D 50,000 units per day, 800 mg of elemental calcium per day, and phosphorus-binding gels. In November 1967, because of progression of the disease, vitamin D was increased to 100,000 units daily, but with no benefit (Fig. 3). Iliac crest biopsy taken in November 1967 (Fig. 4) demonstrated wide uncalcified osteoid seams and areas of bone resorption.

Fig. 4

Undecalcified Iliac Crest Bone Histology of Patient R. N. (Nov. '67)

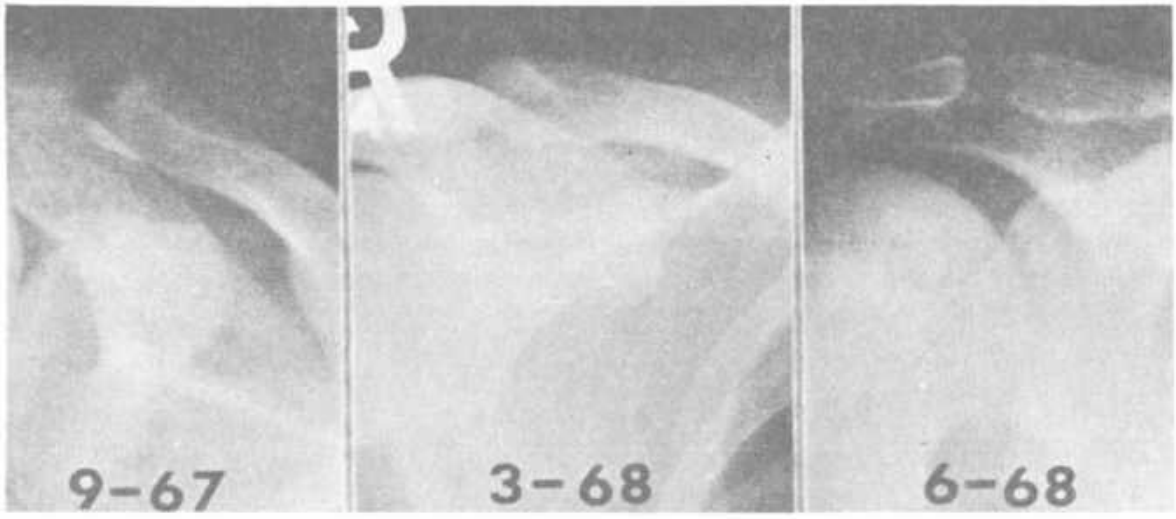


The radiological features of patient D. C. are shown in Fig. 5. When the patient started treatment, her serum calcium was 7.2 mg per 100 ml and the lateral ends of her clavicles showed resorption, a sign of secondary hyperparathyroidism. After six months of dialysis, vitamin D and calcium supplements, and phosphorus-binding gels, her serum calcium rose to the lower limit of normal and her clavicles had undergone remineralization. After continuing the same therapy for an additional three months, she again showed resorption of the lateral end of her clavicles.

The increasing severity of histological bone changes with the duration of fluoridated dialysis is shown in Fig. 6. Vitamin D and calcium therapy had no apparent effect on the bone disease.

Fig. 5

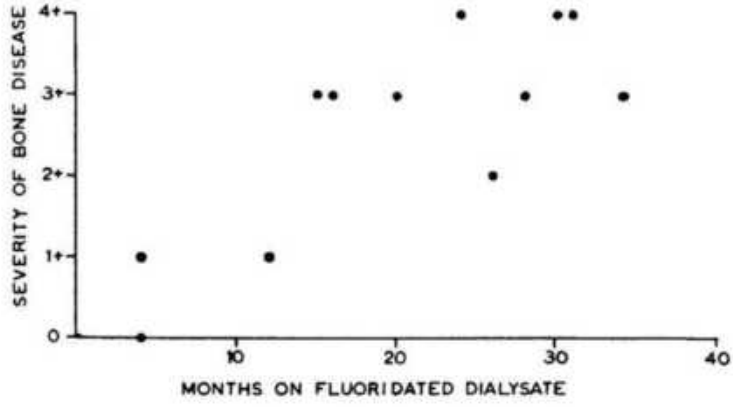
Localized Shoulder View of Patient D. C. in Sept. '67, Outer End of the Clavicle is Cupped and Irregularly Mineralized Due to Subperiosteal Bone Resorption



March '68: Following therapy, the mineralization of outer end of the clavicle has returned to normal.
June '68: Irregularity of mineralization again apparent, not as marked as in initial film.

Fig. 6

Severity of Bone Lesions After Maintenance on Fluoridated Hemodialysis

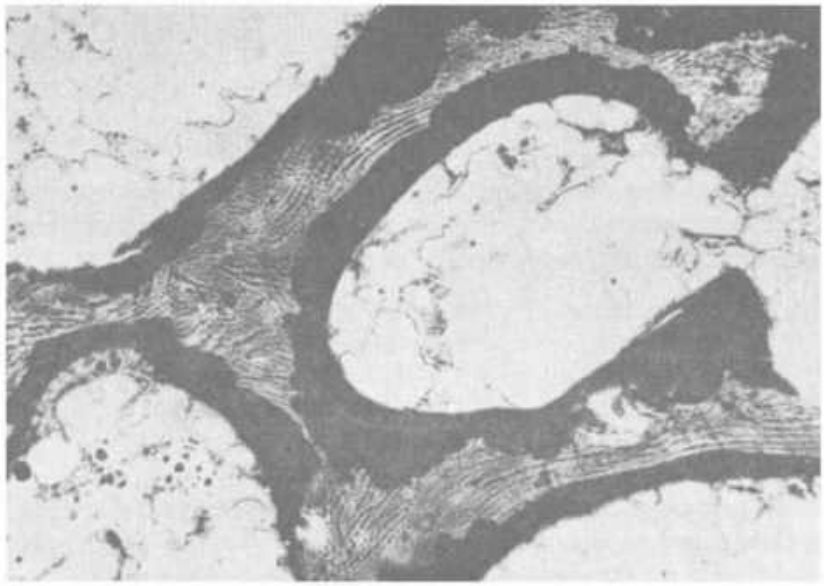


0 = No radiological or histological disease, no clinical symptoms.
1 = Mild radiological and histological disease, but no clinical symptoms.
2 = Moderate radiological and histological disease, with mild bone pains.
3 = Severe radiological and histological disease, with severe bone pains.
4 = Incapacitating disease, with multiple fractures.

The characteristic appearance of an undecalcified section of iliac crest sample is shown in Fig. 7. It was obtained from patient G. B. after six months of vitamin D in daily doses of 100,000 units. Large amounts of uncalcified osteoid and areas of bone resorption are representative of bone biopsies from our patients maintained on fluoridated hemodialysis, and the bones appeared most affected in those dialysed for the longest periods.

Fig. 7

Undecalcified Iliac Crest Bone Histology (Patient G. B.). After Six Months of Vitamin D Supplementation, 100,000 Units Daily



The pre-dialysis fluoride levels were elevated in all patients (Table 4), compared to the 1 μM found in humans not unduly exposed to fluoride (13). The levels rose as blood passed through the dialyser and, at the end of dialysis, the serum levels of arterial blood had increased markedly above their pre-dialysis levels. The estimated uptake of fluoride during a single dialysis ranged from 10 to 29 mg. Fig. 8 shows that the patients' serum fluoride levels increased as a function of time on fluoridated dialysis; the Rochester levels (5) are included for comparison.

The concentration of fluoride in the iliac crest biopsies of individual patients ranged from 800 to 22,700 ppm (Table 4), and were related to the length of time the patients had been maintained on fluoridated hemodialysis (Fig. 9). The two lowest values were found in patients just starting on the program; the lower value was in H. R. who, prior to treatment, had resided in a non-fluoridated community, whereas the higher value was obtained in A. D. who had resided in a fluoridated community for three years prior to treatment.

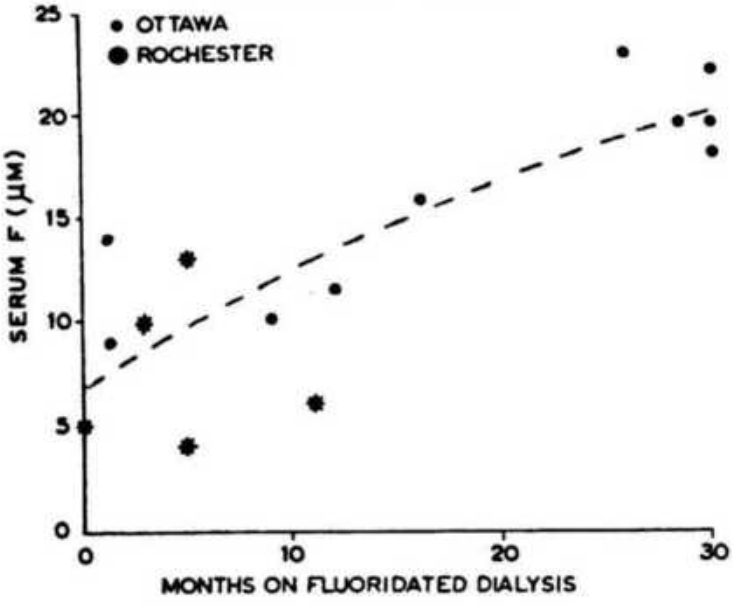
TABLE 4

Patient	Serum Fluoride (Micromoles/Litre) Pre-Dialysis A	Pre-Dialysis V	Post-Dialysis A	Post-Dialysis V	Fluoride Uptake* (mg)	Fluoride Iliac Crest ppm dry fat free
1. G.B.	19	33	32	37	17	22,700
2. C.W.	18	34	29	36	23	19,700
3. R.N.	22	32	29.5	33	10	-
4. L.C.	19.5	33	28	35	12	-
5. W.B.	23	36.5	30	37.5	14	-
6. Y.L.	16	24.5	28	30	12	13,300
7. R.V.	11.5	25.5	23	33	29	20,900
8. D.C.	10.5	20	22.5	28	17	15,900
9. H.R.	8.9	-	30	-	-	800
10. A.D.	14	27	30	34	16	9,500

A = Blood entering dialyzer; V = Blood leaving dialyzer
*on a "per dialysis" basis, and excluding fluoride uptake from foods and beverages, i.e., ingestion of 2 to 5 mg F per day (11,12).

Fig. 8

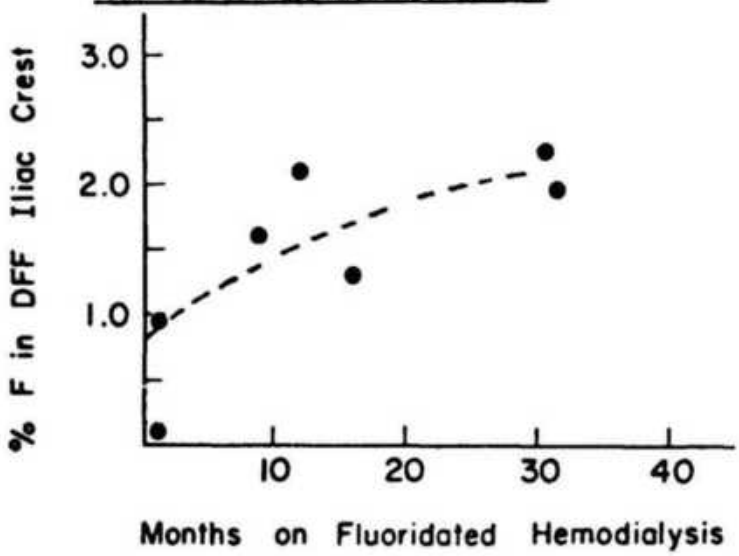
Serum fluoride (F^-) Levels as Related to Time on Fluoridated Dialysis



Each point represents a value in a different case. Dotted line indicates trend obtained by calculating a coefficient of variability (R^2). According to animal experiments pathological changes in bone appear when serum F^- approaches 15 μM (5,19).

Fig. 9

Dry Fat-Free Iliac Crest Fluoride Levels as Related to Time on Fluoridated Dialysis.



Each point represents a value in a different patient. Dotted line indicates trend obtained by calculating a coefficient of variability (R^2). A fluoride content of 1% is equivalent to 10,000 ppm.

The effect of fluoridated hemodialysis on the observed bone and serum fluoride levels in the Ottawa patients was statistically evaluated by calculating a coefficient of determination (R^2) for the data shown in Fig. 8 and 9. This calculation of "best fit" accounted for 65% of the variability in the bone fluoride data and 70% of that in serum fluoride results, and indicated that fluoridated hemodialysis has a comparable effect on bone and serum fluoride increments (Table 5).

TABLE 5

Months of Fluoridated Hemodialysis	Bone Fluoride ppm (estimated) dry fat-free	Bone Fluoride cumulative increment factor	Serum Fluoride µM (estimated)	Serum Fluoride cumulative increment factor
0	8,260	---	7.09	---
10	14,550	1.8	12.54	1.8
20	18,730	2.3	16.84	2.4
30	21,500	2.6	20.23	2.8

Comment

Clinically, radiologically, and histologically, the disease seen in these patients was indistinguishable from uremic osteodystrophy, although the manifestations of bone disease tended to appear sooner and in more severe form in our patients maintained on fluoridated dialysis. Uremic osteodystrophy is characterized by two well-recognized defects: the first is osteitis fibrosa (increased areas of bone resorption and marrow fibrosis) ascribed to secondary hyperparathyroidism; the second is osteomalacia (increased amounts of non-mineralized osteoid) ascribed to the acquired resistance to the action of vitamin D. These two features may be found in various amounts and combinations in individual cases. Stanbury (14, 15) states that when osteomalacia is predominant, uremic osteodystrophy should respond to vitamin D in the appropriate dose. Kay (1) has found histological osteodystrophy in practically all of his patients maintained on non-fluoridated hemodialysis, but prevented them from reaching the symptomatic stage by maintaining a calcium concentration of 3 meq per liter in the dialysis fluid, supplementary dietary calcium, proper usage of phosphate binders, and small doses of vitamin D. Our patients were treated in the same manner and given increasing doses of vitamin D as they became symptomatic, but without improvement.

The failure of our therapeutic efforts suggested that there may have been other factors complicating the disease. The possibility that fluoride was involved was raised by the observations of Taves et al. (4, 5) that the serum fluoride (i.e., F^-) levels in patients regularly hemodialysed with fluoridated water are elevated and comparable to those producing fluorotic bone disease in various mammalian species (6). Our study has confirmed these observations and, in addition, has demonstrated that the basal serum fluoride levels (i.e., arterial values at the beginning of each dialysis) are related to the duration of exposure to fluoridated hemodialysis. An increase in the basal serum levels would be expected as the more reactive bones become increasingly saturated with fluoride, and thus, less able to clear fluoride from the serum (5).

The "zero time" values estimated for bone and serum fluoride (Table 5) are much higher than those normally observed in adult humans who have not been unduly exposed to fluoride (6, 13). However, Taves et al. (5) have reported a serum F^- value of 5.1 uM in a patient not previously dialyzed, but residing in a community with fluoridated water. In the present study, one of the patients had a bone fluoride level of 9,500 ppm (dry fat-free) after only one month of dialysis; this patient also resided in a fluoridated community. The fact that these patients had little-or-no kidney function should be borne in mind, especially as Call et al. (16) have demonstrated that humans with certain types of bilateral kidney disease accumulate more bone fluoride than do humans who do not have these kidney ailments.

Our bone fluoride concentrations, however, must be interpreted with caution for three reasons. First, they are higher than any previously-reported values by as much as 50% (17). Second, the samples were too small to permit duplicate ashings, even though replicate analyses were done on the single distillates.

Third, subsequent analysis (with larger samples) on three of these patients at autopsy, and in patients biopsied four months after defluoridation of the dialysis water, showed only a fraction as much fluoride. (Note: This data will be reported in detail when the effects of defluoridated dialysis, introduced in our center in the fall of 1968, are known). The uptake (and presumably, the loss) of fluoride is less than 30 mg per dialysis (Table 4), so that only a small fraction of the skeleton could have changed as much as indicated in the present study. We have not, however, been able to find a reasonable technical basis to question the analytical procedures; therefore, we tentatively postulate that the fluoride content of these patients' bones may vary markedly with time and with sampling site in a particular bone (18).

Another point that needs to be resolved is why these patients showed no histological or radiographic evidence of increased bone production, as often seen in endemic fluorosis. The serum fluoride concentration may have been too high for this in some patients; but, presumably, earlier in the course of their dialysis, their serum fluoride content was in the range expected to produce osteosclerotic fluorosis (19). Several explanations may be advanced. First, the accumulation of fluoride may have been too rapid (6). Second, since these patients usually have histological osteodystrophy when accepted on the dialysis program, and since osteodystrophy is characterized by a very slow bone-turnover rate (20), their bones could not respond to fluoride levels stimulating bone formation. The third possible explanation is the synergistic effect noted in senile osteoporotics treated with both vitamin D and fluoride (21). In those patients, vitamin D was used to correct the fluoride-induced increase in osteoid seams; but, instead of helping, vitamin D appeared to contribute to the widening of osteoid seams. Our findings of worsening bone disease after combined vitamin D and fluoride exposure may thus be the same phenomenon noted in the study of senile osteoporosis.

Histologically and radiographically, these patients showed features of uremic osteodystrophy instead of the fluorosis characterized by exostoses and osteosclerosis. Nevertheless, the observed changes (osteomalacia, osteitis fibrosa and osteoporosis) were similar to those induced by high doses of fluoride in humans and experimental animals, in which widened osteoid seams have been observed (6, 22-27), and where increased areas of resorption due to secondary hyperparathyroidism may be seen (28). Therefore, it seems likely that fluoride was aggravating the underlying renal osteodystrophy in our patients, and that this effect was enhanced by concomitant administration of high doses of vitamin D.

Bibliography

1. Kay, M., Mangel, R. and Neubauer, E.: Studies in Calcium Metabolism in Patients on Chronic Hemodialysis. Proc. Europ. Dialysis Transplant Assoc. 3:17-23, 1966.
2. Pendras, J. P. and Erickson, R. V.: Hemodialysis: A Successful Therapy for Chronic Uremia. Ann. Internal Med. 64:293-311, 1966.

3. Hampers, C. L. and Schupak, E.: Long Term Hemodialysis. New York & London: Grune and Stratton, pp 110-114, 1967.
4. Taves, D. R., Terry, R., Smith, F. A. and Gardner, D. E.: Use of Fluoridated Water in Long Term Hemodialysis. Arch. Internal Med. 115:167-172, 1965.
5. Taves, D. R., Freeman, R. B., Kamm, D. E., Ramos, L. P. and Scribner, B. H.: Hemodialysis with Fluoridated Water. Trans. Amer. Soc. Artif. Internal Organs 14:412-414, 1968.
6. Johnson, L. C.: Histogenesis and Mechanisms in the Development of Osteofluorosis, in "Fluorine Chemistry, Vol. IV", J. H. Simons (ed.), New York: Academic Press, pp 424-441, 1965.
7. Taves, D. R.: Separation of Fluoride by Rapid Diffusion Using Hexamethyldisiloxane, Talanta 15:969-974, 1968.
8. Taves, D. R.: Determination of Submicromolar Concentrations of Fluoride in Biological Samples. Talanta 15:1015-1023, 1968.
9. Fry, B. W. and Taves, D. R.: Serum Fluoride Analysis with the Fluoride Electrode. J. Lab. Clin. Med. 75:1020-1025, 1970.
10. Bohatirchuk, F.: Calciolysis as the Initial Stage of Bone Resorption. Amer. J. Med. 41:836-846, 1966.
11. Marier, J. R. and Rose, D.: The Fluoride Content of Some Foods and Beverages — A Brief Survey Using a Modified Zr-SPADNS Method. J. Food Sci. 31:941-946, 1966.
12. Spencer, H., Lewin, I., Fowler, J. and Samachson, J.: Effect of Sodium Fluoride on Calcium Absorption and Balances in Man, Amer. J. Clin. Nutr. 22:381-390, 1969.
13. Taves, D. R.: Normal Human Serum Fluoride Concentration. Nature 211: 192-193, 1966.
14. Stanbury, S. W.: Bony Complications of Renal Disease, in "Renal Disease, Edition V", D. A. K. Black (ed.), Oxford: Blackwell Scientific Publications, pp 665-715, 1967.
15. Stanbury, S. W.: Bone Disease in Uremia. Amer. J. Med. 44:714-724, 1968.
16. Call, R. A., Greenwood, D. A., LeCheminant, W. H., Shupe, J. L., Nielsen, H. M., Olson, L. E., Lamborn, R. W., Mangelson, F. L. and Davis, R. V.: Histological and Chemical Studies in Man on Effects of Fluoride. Public Health Rep. 80:529-538, 1965.
17. Kilborn, L. G., Outerbridge, T. S. and Lei, H. P.: Fluorosis with Report of an Advanced Case. Canad. Med. Assoc. J. 62:135-141, 1950.
18. Soriano, M.: Periostitis Deformans: Un Nuevo Tipo de Fluorosis Osea en el Hombre, Revista Clinica Espanola 97:375-388, 1965.
19. Taves, D. R.: New Approach to the Treatment of Bone Disease with Fluoride. Fed. Proc. 29:1185-1187, 1970.
20. Sarnsethsiri, P., Jaworski, Z. F., Shimizu, A. G. and Frost, H. M.: New Bone Formation and Osteoid Thickness: Appositional Rate in Patients Undergoing Renal Dialysis. Arch. Path. 88:49-53, 1969.
21. Merz, W. A., Schenk, R. K. and Reutter, F. W.: Paradoxical Effects of Vitamin D in Fluoride-Treated Senile Osteoporosis. Cal. Tiss. Res. 4: (Suppl.): 49-50, 1970.

22. Comar, C.L., Visek, W.J., Lotz, W.E. and Rust, J.H.: Effects of Fluoride on Calcium Metabolism and Bone Growth in Pigs. Amer. J. Anat. 92: 361-389, 1953.
23. Belanger, L.F., Visek, W.J., Lotz, W.E. and Comar, C.L.: Rachitometric Effects of Fluoride Feeding on the Skeletal Tissues of Growing Pigs. Amer. J. Path. 34:25-35, 1958.
24. Adams, P.H. and Jowsey, J.: Sodium Fluoride in the Treatment of Osteoporosis and Other Bone Diseases. Ann. Internal Med. 63:1151-1155, 1965.
25. Hodge, H.C. and Smith, F.A.: "Fluorine Chemistry, Vol. IV", J.H. Simons (ed.), New York: Academic Press, p. 411, 1965.
26. Burkhart, J.M. and Jowsey, J.: Effect of Variations in Calcium Intake on the Skeleton of Fluoride-Fed Kittens. J. Lab. Clin. Med. 72:943-950, 1968.
27. Jowsey, J., Schenk, R.K. and Reutter, F.W.: Some Results of the Effect of Fluoride on Bone Tissue in Osteoporosis. J. Clin. Endocr. Metab. 28: 869-874, 1968.
28. Faccini, J.M.: Fluoride and Bone. Calc. Tiss. Res. 3:1-16, 1969.

Acknowledgements: We are indebted to Dr. R. A. Couture and Dr. A. Rashid for clinical assistance; to Dr. G.N. Tolnai, Dr. F.P. Bohatirchuk and T. Jeletski for histological preparations and interpretations; to Dr. D. Gray for reviewing the X-rays; to Dr. D. R. Taves, who performed the serum fluoride analyses; to Drs. D. Rose and L.F. Belanger for advice; to Mrs. J. Howe for technical assistance and Mrs. R. Yu and the nursing and technical staff of the Hemodialysis Unit of the Ottawa General Hospital for general help.