

## The Effects of Sodium Fluoride on Calcium Metabolism of Subjects with Metabolic Bone Diseases \*

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After the initial description of skeletal fluorosis (1), Roholm in 1937 (2) reported a study of 68 cryolite miners, of whom 50 had slight or moderate osteosclerosis. Seven others, who had advanced skeletal changes, also had calcification of the ligaments and tendinous insertions, particularly of the vertebral column. Minor gastrointestinal and joint symptoms were frequent, but except for restricted mobility of the spine of the most severely affected workers, physical examination and tests of urine and peripheral blood were unremarkable. These findings led Roholm to characterize osteosclerosis in this group of subjects as "fairly benign." It has since been amply confirmed that osteosclerosis can result from prolonged exposure to fluoride and that subjects with minimal or moderate osteosclerosis are usually asymptomatic and in good general health (3).

This leads to the consideration that salts of fluorine might be used as treatment of patients with osteoporosis to cause a degree of osteosclerosis that would strengthen the skeleton but not lead to other changes. This report describes the results of studies of the effect of fluoride on subjects with metabolic bone diseases. Although our main interest in undertaking an evaluation of this problem has been in the possible therapeutic effect of fluoride in osteoporosis, we have studied also its effect in subjects with Paget's disease, because of the possibility that fluoride would suppress the increased osteoclastic activity and thereby retard progression of this disease. We have

found that calcium retention is brought about in most subjects given 40 to 75 mg fluoride ion per day, but usually only after 2 to 3 months of treatment. These results extend our earlier, brief reports (4, 5) and are consistent with those recently described by Purves (6) and in a preliminary report by Bernstein and co-workers (7).

### Clinical Material and Methods

Thirteen balance studies of 3 to 14 weeks were carried out on six patients. Clinical summaries are given in the appendix. Patients were admitted to the metabolic ward of the Clinical Research Center, University Hospital, and placed upon a diet calculated to provide an almost constant daily intake of calories, protein, fat, calcium, and phosphorus. Food was purchased in large lots to reduce variation because of differences in composition from lot to lot. Two or three daily menus were repeated regularly throughout the hospital stay, and one duplicate of each menu was analyzed each week. All urine and stools were collected separately and refrigerated until analyzed. Charcoal or carmine was given orally at the start of each balance period and its appearance in stool used as an indication of the delay in transit of food through the gastrointestinal tract.

The first balance period was not started until after the patient had been 2 weeks on the diet. Four or five 1-week periods of balance measurement were made, and a number of clinical studies were carried out as a base line for comparison when repeated during treatment with sodium fluoride (Table I). Thereafter, patients were treated with 0.7 to 1.4 mg fluoride per kg body weight per day, given orally as sodium fluoride, either in solution or in enteric-coated tablets. In some studies, continuous balance measurements were made for the first 6 to 9 weeks of treatment. In other instances, patients were discharged as soon as treatment was started and, while continuously under treatment with fluoride, were readmitted for repeat balance and clinical studies.

Stool and diet collections were homogenized and weighed. Weighed samples were reduced to ash at 600° C in a muffle furnace, the ash was dissolved in HCl, water was added, and the pH was brought to 4.5. Ammonium oxalate was then added to cause a calcium oxalate precipitate, which was collected. Calcium oxalate precipitates were prepared also from samples of urine.

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TABLE I  
 Results of clinical chemical testing of patients

Diagnosis:	Patient:	Primary osteoporosis					
		M.H.			F.F.		
		Control	Treatment		Control	Treatment	
Study period:		1	2		1	2	
Weeks of treatment			1-7	32-36		30-34	72-75
Dose of fluoride	mg/day		50	50		50	40
Total dose of fluoride	g		2.5	13		11.9	24.8
Height	cm	156	153		167	167	
Weight	kg	49			55		
Test (average of all values)							
Plasma calcium	mg/100 ml	10.2	9.5	9.8	8.8	9.3	9.4
Plasma phosphorus	mg/100 ml	3.4	3.1	3.1	3.3	3.5	3.3
Serum alkaline phosphatase	King-Armstrong U	8.7	8.7	8.4	6.6	9.1	5.5
Serum acid phosphatase	U/100 ml	0.18	0.27	0.34	0.51	0.86	0.68
Packed cell volume	%	41	37	38	50	49	49
Leukocyte count	10 <sup>3</sup> /mm <sup>3</sup>	4.9	5.6	5.0	6.0	6.5	6.4
Urinalysis		†	†	†	†	†	†
Creatinine or (BUN)	mg/100 ml	(24)		(23)	0.8	1.0	0.9
Clearance: creatinine or (BUN)†	ml/min	(20)		(88)		83	133
P.S.P. excretion†	% in 1 hr	68		39		88	
Fasting blood sugar†	mg/100 ml	74			77	90	72
B.S.P. excretion†	% in 45 min	26		21	6	2	
Serum glutamine oxalate trans-aminase†	U/100 ml	43		17		10	
Absorption of dietary fat	% of intake	93	94	96	98	99	99
Urine hydroxyproline	mg/24 hr	11.3	14.4	15.2	53.2	59.5	59.7

\* BUN = blood urea nitrogen; P.S.P. = phenolsulfonphthalein; B.S.P. = bromosulfaphthalein.

† Obtained once or twice in each study period; other values were usually determined each week.

‡ Normal.

These precipitates were dissolved in HCl, and the calcium was measured by titration with Versene (ethylenediaminetetraacetic acid tetrasodium salt) as previously described (8), except that an automatic titrating and recording system was used. Addition of 100 mg of fluoride to the starting sample (1-day collection of urine or stool) or 200  $\mu$ g fluoride to the final sample (which contained 30 to 70  $\mu$ g calcium) did not affect this analysis. Calcium in plasma was analyzed directly by titration with Versene (8). Stool and urine nitrogen was measured by the micro-Kjeldahl method (9), phosphate by the method of Fiske and Subbarow (10), urine creatinine by the Jaffe reaction (9), hydroxyproline by the method of Prockop and Udenfriend (11), and stool fat by conversion to fatty acid and extraction and titration of the resulting acidity (12). Fluoride was measured after perchloric acid distillation of samples of diet, stools, or urine (13) by reaction with a zirconium lake (14).

Balance results and theoretical balance of phosphorus were calculated and charted according to the method of Reifstein, Albright, and Wells (15), with slight modification of the scales used for charting. The intake is charted down from the zero line, stool up from intake, and urine above stool, so that a positive balance is present when the sum of stool and urine excretion does not reach the zero line.

## Results

*Balance studies.* Figure 1 illustrates the results of studies of M.H., including changes in balance of calcium, phosphate, nitrogen, and fluoride. There was little change of calcium balance during the first 7 weeks of treatment, but after 32 weeks of treatment definite calcium retention was present. (As in all other studies, the patient had been on the metabolic diet for 2 weeks before the first balance measurement.) Phosphate balance reflected similar changes, while nitrogen balance was only slightly altered. There was a striking initial drop in the rate of excretion of calcium in urine, from control values of 112 mg per day to 17 mg per day during the second and third weeks of treatment, but 6 months later, when the maximal change of calcium balance was found, excretion of calcium in urine was the same as it had been before treatment. Fecal calcium was reduced during both periods of study during treatment.

TABLE I  
before and during treatment with sodium fluoride\*

		Primary osteoporosis				Steroid osteoporosis		Paget's disease	
A.S.		R.K., study 1		R.K., study 2		H.L.		E.M.	
Control	Treatment	Control	Treatment	Control	Treatment	Control	Treatment	Control	Treatment
	1-9		1-5		28-32		31-35		40-45
	50		80-100		50		75		60
	3.0		3.1		11.2		18.4		21.4
168	168	167		167		177	177	148	148
50		71				85		60	
10.6	9.4	10.5	10.6	10.1	10.0	10.2	9.6	10.2	9.4
3.5	2.9	3.7	3.4	2.6	2.2	2.9	2.3	3.2	3.7
4.5	5.4	5.7	7.4	5.7	7.4	4.9	5.7	30.2	29.2
0.63	0.20	0.22	0.28	0.22	0.28	0.36	0.30	0.54	0.17
45	43	45	39	50	51	48	45	40	41
10.8	10.6	8.6	7.3	7.1	8.2	9.5	7.4	7.9	7.0
§	§	‡	‡	‡	‡	‡	‡	‡	‡
1.1		(14)		(21)	(18)	(12)	(11)	(15)	(19)
(41)		(41)		60	(47)	118	120	(57)	(97)
65				67	52	83	59	37	73
105				92	87	94	66	69	72
12		14		18	9	8	9	6	4
		25		52		20	11	15	
96	95	98	96	96	95	89	88	96	96
24.2	24.1	23.3	33.8	40.0	41.5	37.0	40.3	176	134

§ Leukocytes, few granular casts.

|| Numbers in parentheses refer to BUN.

Figure 2 shows similar results in a second patient, F.F., studied twice during 1½ years of treatment. There was moderate calcium retention 30 and 72 weeks after starting treatment not associated, in this patient, with reduction of calcium in urine, but instead, with some decrease in the amount of calcium in stools. The apparently negative phosphorus balance was associated with nitrogen loss, as shown by the theoretical balance, which agreed with that found by analysis.

A third subject with primary osteoporosis, R.K., was studied during two courses of treatment of fluoride and showed no response (Figure 3). The first study was carried out while he was given an intake of 200 mg calcium per day. He had definitely increased excretion of fat and nitrogen in stool (Figure 3) accompanied by aching epigastric pain and diarrhea when he was given 1.4 mg fluoride per kg per day during the first 2 weeks of treatment. These symptoms and chemical evidence of absorptive difficulty disappeared when the dose was reduced. The rate of cal-

cium excretion in urine, low during the control period, diminished even further during treatment, but the over-all balance of calcium was unchanged. This result was not unexpected, because of the short duration of treatment. Similar changes in excretion of calcium in urine, but lack of effect upon calcium balance, were observed 8 months later when the patient was re-treated at a time when he had a normal calcium intake.

The balance results for all patients studied are summarized in Table II. In this table, the average balance for the entire 28 days (four periods) of the pretreatment study is compared to the last 28 days of each balance measurement made during treatment (except for the final study of F.F., where only three periods of balance study were measured). This shows the data already commented upon and results of the three other studies described below.

A.S., a fourth patient with primary osteoporosis, was treated with fluoride for only 9 weeks without change in calcium balance.

A subject with steroid osteoporosis (H.L.) was treated with fluoride for 31 weeks before the second balance study but had substantial increase of his dose of prednisone, from 20 mg per day before treatment to 30 mg per day during most of the period of treatment with fluoride. It is probable that this may have influenced the results, a speculation supported because of increased calcuria, which has not been seen in any other patient treated with fluoride but which is the usual result of glucocorticoid therapy.

Balance measurements were performed also on E.M., a patient with Paget's disease, for 5 weeks before and after 28 weeks of treatment with fluoride. There was slight reduction of calcium in urine and stools and slight, but significant, change of calcium balance from an average of 64-mg loss per day in the control period to reten-

tion of 45 mg per day. Phosphorus and nitrogen balances did not change.

The calcium balances recorded in Table II are shown by the closed circles in Figure 4. The open circles show the results from previous work calculated in the same way (4). This gives our total present experience in testing the effect of fluoride in the dose used in this study. Significant calcium retention was not observed during the first 8 to 10 weeks of treatment with fluoride. Of the eight subjects treated for 10 or more weeks, four had primary osteoporosis, two had steroid-induced osteoporosis, and two had Paget's disease. The results in these different states are as follows:

a) Primary osteoporosis. Three of four subjects retained more calcium during treatment than during the control period, the average positive change in these three being 138 mg per day. Two

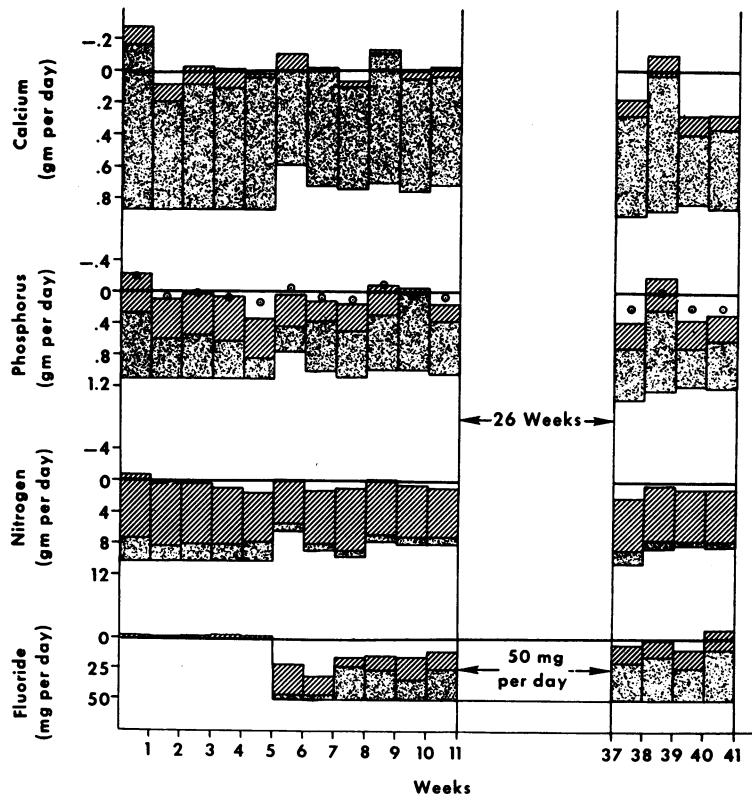


FIG. 1. RESULTS OF BALANCE STUDY OF M.H., AN 84-YEAR-OLD WOMAN WITH PRIMARY OSTEOPOROSIS. Treatment with fluoride was continued at the dose shown during the period between the first and second study. 10 ml aluminum hydroxide gel was given twice a day after the second week of treatment until the end of the final study. Theoretical balance of phosphorus is indicated by the circles.

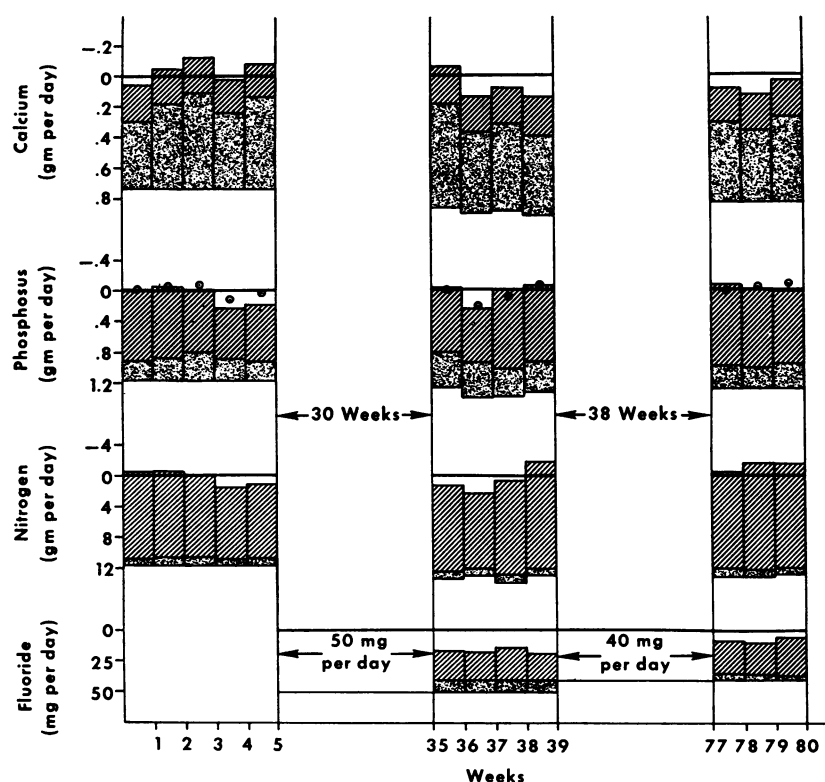


FIG. 2. RESULTS OF BALANCE STUDY OF F.F., A 62-YEAR-OLD MAN WITH PRIMARY OSTEOPOROSIS. Treatment with fluoride was started at the end of the first period of study and continued thereafter.

patients were elderly postmenopausal females and the other (F.F.) was a male with what classically is called idiopathic osteoporosis, a severe form of osteoporosis with early onset, said not to respond to any commonly used form of therapy (16).

b) Steroid osteoporosis. One subject retained 100 mg additional calcium per day during treatment with fluoride; the other (H.L.), whose dose of prednisone was increased during the period of fluoride administration and who had diminished fat absorption before and during treatment with fluoride, did not respond.

c) Paget's disease. Both subjects showed calcium retention (the average was 149 mg per day), and one (W.T.) experienced marked relief of symptoms and fall of alkaline phosphatase activity during treatment.

Except where the fluoride was poorly absorbed, as with M.H., whose fecal excretion increased when aluminum hydroxide gel was given (Figure 1), the amount of fluoride in stools re-

mained low. Excretion in urine, however, equal to less than half the amount given during the first few weeks of treatment, usually rose during continued treatment (Figures 2 and 3), so that the gross retention of fluoride diminished. In spite of this tendency, very substantial amounts of fluoride were retained by all but one of the subjects, even after 30 to 75 weeks of treatment. As shown in Table II, the average positive balance of the four subjects studied after about 30 weeks of treatment was 18 mg per day, equal to one-third of the dose given. The only subject studied after a much longer period of treatment (F.F.) retained half as much at 72 as he had at 30 weeks.

*Other studies.* The values for calcium, phosphorus, and alkaline and acid phosphatase of plasma, as well as tests of renal, hematologic, hepatic, and other systemic functions, obtained periodically throughout treatment of all patients, showed no significant alteration from control values. The averages of the results of these tests are shown in Table I. Fat balance was meas-

TABLE II  
Average of results of balance studies during the last four periods

Patient:	M.H.		F.F.		A.S.			
	Control	Treatment	Control	Treatment	Control	Treatment		
Study period:	1 2		1 2					
<b>Ca, mg/day</b>								
Intake	868	724	868	727	879	812	858	842
Stool	817	710	608	567	576	514	786	769
Urine	112	40	108	224	243	229	73	60
Balance	-61	-26	152	-64	60	69	-1	13
<b>P, mg/day</b>								
Intake	1,091	1,022	1,238	1,158	1,318	1,251	1,335	1,344
Stool	579	712	687	305	421	297	483	495
Urine	535	284	360	773	880	1,012	788	792
Balance	-23	26	191	80	17	-58	64	57
<b>N, g/day</b>								
Intake	9.16	8.33	8.57	11.50	13.10	12.96	10.50	10.58
Stool	1.27	0.88	0.82	0.90	0.86	.95	1.31	1.29
Urine	7.77	6.98	6.68	10.23	11.85	13.33	7.95	8.73
Balance	0.12	0.47	1.07	0.37	0.39	-1.32	1.24	0.56
<b>F, mg/day</b>								
Intake (less diet)		50.00	50.00		50.00	40.00		50.00
Stool	0.37	24.40	33.30	0.04	8.66	3.65	1.30	7.31
Urine	0.76	13.60	16.04	0.26	25.01	28.04	0.14	28.64
Balance		12.00	0.66		16.33	8.31		14.05

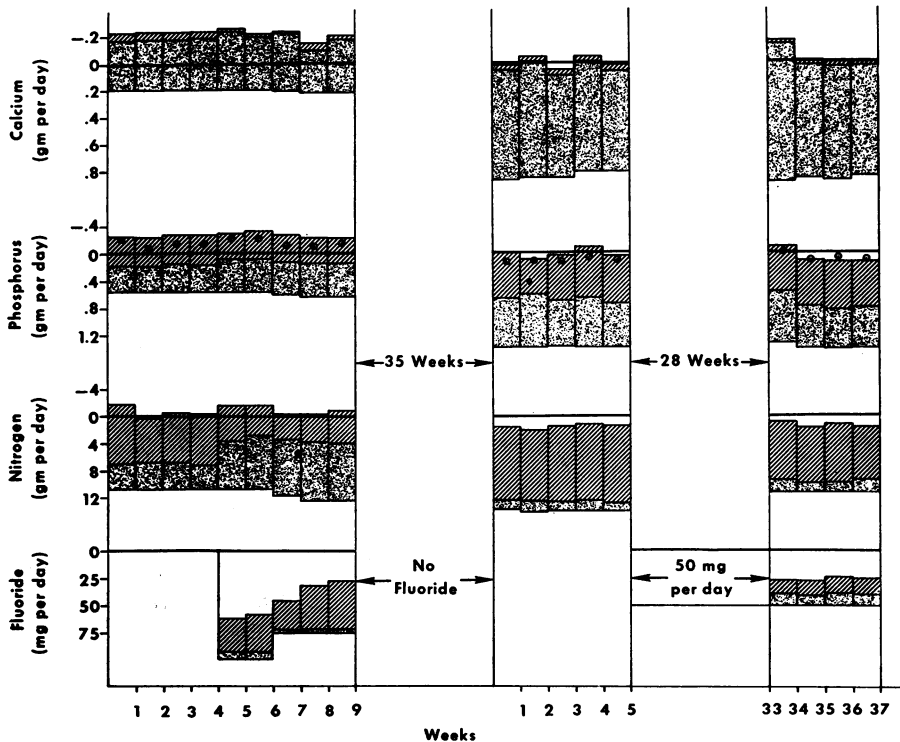


FIG. 3. RESULTS OF BALANCE STUDY OF R.K., A 70-YEAR-OLD MAN WITH PRIMARY OSTEOPOROSIS. Sodium fluoride was given for 5 weeks during the first study period and was then stopped for 40 weeks. It was started again at the end of the second control study and continued thereafter.

TABLE II  
(28 days) of control study and treatment with sodium fluoride

R.K., study 1		R.K., study 2		H.L.		E.M.	
Control	Treatment	Control	Treatment	Control	Treatment	Control	Treatment
187	206	827	859	802	944	851	840
371	386	794	871	761	797	720	676
56	15	40	24	301	454	196	119
-240	-195	-7	-36	-260	-307	-65	45
561	615	1,370	1,391	1,690	1,511	1,170	1,195
391	484	689	641	1,707	1,489	532	499
437	391	654	676	75	3	603	673
-267	-260	27	74	-92	19	35	23
10.67	11.88	14.09	11.58	16.56	16.19	11.04	10.92
3.78	4.29	1.50	1.96	2.16	1.82	1.13	1.40
7.12	8.39	10.97	8.26	14.31	13.41	9.29	8.64
-.23	-.80	1.62	1.36	0.09	0.96	0.62	0.88
0.16	85.00	0.17	50.00		75.00	0.15	60.00
0.05	7.26	0.07	9.52		25.67	0.22	19.73
	36.60		14.75		27.34		11.61
	41.14		25.73		21.99		28.66

ured as an indication of whether or not fluoride reduced gut absorptive ability, but was not consistently altered. No change in appearance of X-ray films of the skull, vertebrae, chest, pelvis, or long bones was found in any patient.

Only two symptoms, epigastric distress and joint pain, have been encountered in subjects given fluoride. Most patients who take more than 10 mg fluoride in solution at one time experience aching epigastric pain occurring within a minute and lasting about 30 minutes. It is decreased if fluoride is given after a meal and abolished if fluoride is given after aluminum hydroxide gel, calcium carbonate, or in enteric-coated tablets.

Three patients (R.K., H.L., and E.M.) experienced aching joint pain in the knees or ankles upon weight bearing. This symptom started gradually after several months of treatment, and persisted until fluoride was discontinued, after which it slowly disappeared. The pain was present only upon weight bearing, was moderately severe, and definitely reduced the subjects' physical activity. There was no effusion, restriction of motion, tenderness, or change in X-ray findings. Each of these patients gave a history of inter-

mittent aching joint pain for many years before treatment.

### Discussion

*Dose and toxicity.* Although many studies of fluoride effect have been carried out in lower mammals and man, relatively few describe the effect of doses such as we have used, or report data which allow an estimate of the lower limit of toxicity for humans (2, 3). Black, Kleiner, and Bolker found no evidence of toxicity in 60 human subjects treated for as long as 6 months with an average daily dose of 320 mg NaF, equal to 2.4 mg fluoride per kg per day for a 60-kg subject (17). Others, however, have considered this dose rate to be toxic when given for prolonged periods to humans or small mammals (18).

A second consideration in giving fluoride to patients is duration of treatment. Roholm found sclerosis only in subjects who had been exposed to fluoride for more than 5 years and found advanced changes only after 10 years of exposure (the average being 20 years). The suggestion that progressive changes occur with prolonged exposure is given weight by reports of crippling

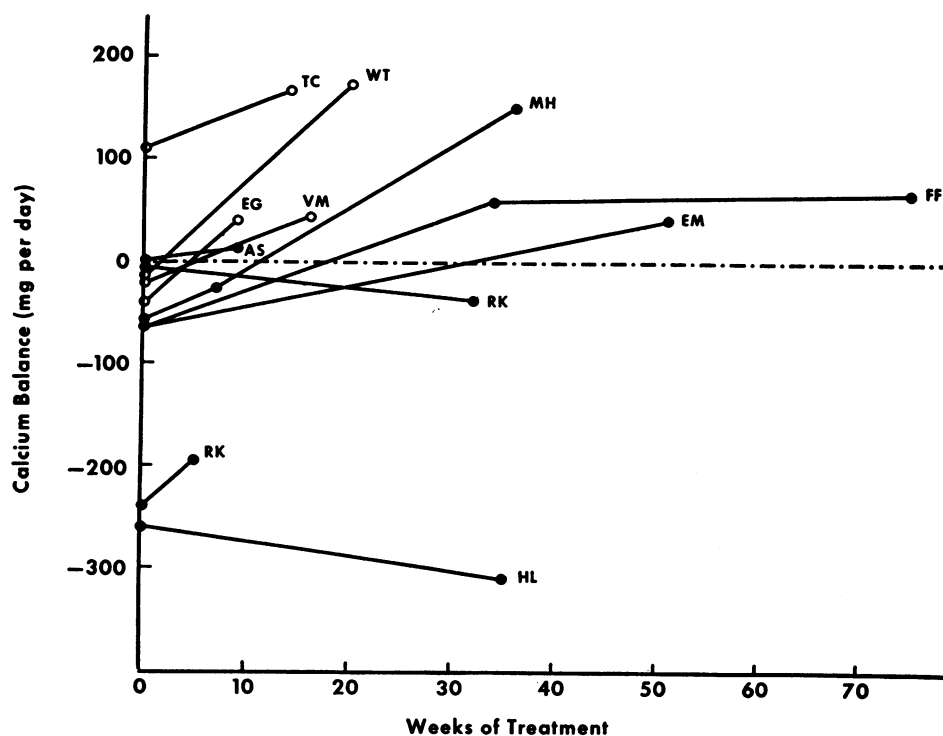


FIG. 4. SUMMARY OF MEASUREMENTS OF CALCIUM BALANCE OF ALL SUBJECTS SO FAR TREATED WITH ABOUT 1 MG FLUORIDE PER KG BODY WEIGHT PER DAY. Closed circles are for patients described in this study and open circles are for those reported previously (4). The abscissa gives weeks between the end of the control study and the end of any period of study during treatment. On the ordinate is shown the average calcium balance during the last 4 weeks of the control period and of each period of balance measurement during treatment.

fluorosis from several areas of Asia where life-long exposure to fluoride in drinking water and dust eaten with food occurs (19). Thus, if fluoride is to be used in a therapeutic trial, both the dose and duration of treatment must be controlled.

It was possible to calculate, from the amount of fluoride found by Roholm in the skeleton of two autopsied osteosclerotic patients (2), and because about half of an oral dose is retained (3), that his subjects ingested at least 0.3 mg fluoride per kg body weight per day. We treated our first patients for relatively short periods with this dose but found no effect on calcium metabolism (4) and have since given about 1.0 mg fluoride per kg per day. At this dose rate, we have found no evidence of nonskeletal effects (clinical, chemical, or radiographic), except for the two symptoms already described. The epigastric pain, which occurs immediately and is abolished by measures which reduce gastric acidity or by use of enteric-coated tablets, presumably is from a direct action

of fluoride on the gastric mucosa. One subject who took a higher dose, 1.4 mg per kg per day, given without either enteric coating or antacid, experienced symptoms and signs of reduced absorptive ability, probably indicating a toxic effect on the intestinal mucosa from this dose. It is probable that Albright, Reifenstein, and Forbes, who treated an osteoporotic subject with 30 to 90 mg fluoride per day (20) were dealing with a similar toxic effect on gastrointestinal absorption, inasmuch as they observed increased fecal excretion and a negative change of balance of nitrogen as well as of calcium and phosphorus.

The joint pain seemed definitely related to treatment and appeared clinically to be an exacerbation of previously symptomatic degenerative arthritis. Tracer studies show that fluoride is concentrated slightly in cartilage and tendon, compared to blood and other soft tissues (21, 22). Although direct evidence is lacking, the joint pain



may be related somehow to concentration of fluoride in structures around the joint.

Our present studies show that 1 mg fluoride per kg per day causes calcium retention, but do not give information as to the minimal effective dose. That half of this amount (the dose we are now using) may be effective is suggested by the work of Purves, who has reported metabolic effects from about 0.5 mg per kg per day in treatment of subjects with Paget's disease (6).

*Mechanism of fluoride effect.* The positive change in calcium balance occurred because of reduced excretion of calcium in both stools and urine. The urinary changes were not accounted for by altered glomerular filtration or reduction of total concentration of calcium in plasma, and the reason, particularly for the marked initial reduction of calcuria of some patients, is not clear. Decreased fecal calcium was seen in all subjects whose calcium balance showed a positive change during treatment, and, except for H.L., in none of those who did not. This may be interpreted as the result either of increased gastrointestinal absorption, as suggested by Bernstein and co-workers (7), or reduced excretion of endogenous calcium, or both. Although the balance data do not allow a conclusion as to which of these possibilities is more likely, the latter seems more in keeping with the frequent finding of a reduction of excretion of calcium in urine of patients treated with fluoride.

In spite of an enormous amount of toxicological study in animals (3), no reliable concept of the mechanism of fluoride action on bone can be formulated. It appears to be quite complex, combining physical and cytological effects, with different histologic abnormalities depending upon the dose of fluoride. Probably because of the structural similarity between fluorapatite and hydroxyapatite of bone, fluoride is concentrated in this tissue, presumably causing cells of bone to be exposed to a greater concentration of fluoride than cells of other tissues. Fluoride in large excess of the concentration found in body fluids is known to be a potent inhibitor of numerous enzyme activities, including phosphatases, lipases, and enolase (3, 23), but how it alters bone cell function is not clear. The observed changes in fluorotic human bone, of stimulated growth of architecturally abnormal periosteal, endosteal, and trabecular

bone (2, 3, 19), and probably also decreased osteoclastic activity (2), are clearly the result of a cytological effect. In addition, there is evidence to suggest that bone crystals coated with fluoride (as fluorapatite) have fewer defects and are less reactive than normally (24). Such physical changes could well depress bone resorption and thus cause an increased skeletal mass.

*Clinical application.* The major objective of this study was to show whether or not fluoride causes calcium retention when given to subjects with metabolic bone disease. If this occurs in the absence of extraskeletal calcification, it may be assumed that the rate of bone formation is accelerated or the rate of resorption reduced, or both, but not necessarily that the strength of the skeleton has been increased. Although data as to strength of fluorotic bone of humans appear to be lacking, and such data from animals given a comparable dose of fluoride are scanty, they suggest that bone strength is normal (25, 26).

The fact that fluoride administration results in retention of calcium by subjects with several types of osteoporosis, with or without concurrent testosterone therapy, as well as by patients with Paget's disease, suggests that it occurs because of a general effect on bone metabolism rather than from interference with the metabolic abnormality of any given disease process.

The positive change of calcium balance, which was usually about 150 mg per day, is significantly less than that often observed when normal subjects or osteoporotic patients are treated with estrogen (16, 27, 28) or by supplementation of the dietary intake of calcium (29-32). However, although it is unlikely that an agent would be useful as a treatment of osteoporosis if it did not cause calcium retention, it does not follow that the best agent causes the greatest retention. Not only does fluoride cause significant retention of calcium when given for 3 to 18 months, but it is the only agent suitable for prolonged administration to humans known to increase the density of the skeleton.

Purves, in a recently published study of effect of fluoride in Paget's disease (6), has noted symptomatic improvement, marked reduction of the abnormal uptake of  $Ca^{47}$  by the involved portions of the skeleton, and changes in calcium balance similar to those we have reported. These findings,

and the reduction of the abnormally high rate of excretion of hydroxyproline observed in one of our subjects, suggest the possibility that, in addition to a general effect on the skeleton which tends to cause calcium retention, fluoride may directly affect the metabolic abnormality of Paget's disease. A somewhat comparable reduction of activity of this disease has been brought about by cortisone (33) and aspirin (34), as indicated by symptomatic remissions, and changes of hydroxyproline excretion in urine and calcium balance that are comparable to those shown here.

The demonstration of calcium retention in osteoporotic subjects treated with fluoride brings up the question of whether or not a smaller dose of fluoride, given for years before substantial demineralization usually is seen, might reduce the incidence or severity of osteoporosis. This cannot be resolved by showing response to therapy of patients who already have the disease, but can best be examined by study of incidence of osteoporosis in groups of subjects with backgrounds that are similar except for different levels of fluoride exposure. Although there is work which suggests a possible relationship between level of fluoride intake and incidence of osteoporosis (35), it is inconclusive. It would seem of considerable importance that a study specifically designed to test this point be carried out.

### Summary

1) Detailed clinical and biochemical studies have been carried out on six subjects before and during treatment with about 1 mg fluoride per kg body weight per day (given by mouth as sodium fluoride) for 5 to 75 weeks.

2) External balance of calcium, phosphorus, nitrogen, and fluoride was measured in each case. In four subjects, all in negative calcium balance during the control period, these measurements were carried out under conditions that allowed assessment of the effect of fluoride on calcium balance. Of three patients with primary osteoporosis, two retained calcium during treatment, showing an average positive change of 173 mg per day. A third subject did not respond. The fourth subject, with Paget's disease, also retained calcium during treatment.

3) Urinary excretion of calcium in several subjects fell to very low values, and fecal excretion of

calcium was usually reduced during treatment with fluoride.

4) Three subjects, who had had repeated attacks of aching joint pain for many years, had an increased amount of this pain. There were no other symptoms and no biochemical evidence of toxic effect in any patient.

5) On the basis of this and other published work, it is concluded that further clinical trial of fluoride is warranted in patients with osteoporosis and Paget's disease.

### Appendix

*Primary osteoporosis.* M.H., an 84-year-old woman, had a compression fracture of the first lumbar vertebra at the age of 72. She was treated intermittently with diethylstilbestrol and testosterone propionate until 5 months before admission but has been given none since. Severe demineralization of the spine and several vertebral compression fractures were shown by X ray. She was studied during a control period and then for the first 7 and last 4 of 36 weeks of treatment with 1.0 mg fluoride per kg per day. After the first 2 weeks of treatment, she was given 20 ml aluminum hydroxide gel per day.

F.F., a 62-year-old male, had X-ray evidence of compression of the eighth thoracic vertebra when 44 years old. He was treated with testosterone propionate for 5 years, without improvement, and was given testosterone 17 $\beta$ -cyclopentanepropionate, 400 mg every month for 7 months before and throughout the study. Our X rays showed marked demineralization of pelvis and spine with compression fractures of the eighth and twelfth thoracic and first lumbar vertebrae. He was studied before and twice during 75 weeks of treatment with 1.1 mg fluoride per kg per day. He felt markedly improved, experiencing relief of back pain.

A.S., a 61-year-old male was found to have compression of the last two thoracic vertebrae when he was 48 years old. He was given 200 mg testosterone 17 $\beta$ -cyclopentanepropionate every 2 weeks for 2 years before and throughout the period of study. Our X rays showed vertebral demineralization with deformity of the end plates of several vertebrae. He was treated for 9 weeks with 1.0 mg fluoride per kg per day without symptomatic change.

R.K., a 70-year-old male with a history of back pain and episodic aching pain in the knees and ankles for 10 years, was found 6 months before admission to have compression deformities of the last thoracic and fourth lumbar vertebrae and a rib fracture. During the first test of fluoride effect he was given a low calcium intake of 200 mg per day. After discharge he was placed upon a normal diet and 200 mg testosterone 17 $\beta$ -cyclopentanepropionate every other week until completion of the final study. He was readmitted 7 months later and studied during a second control period and for the last 4 of 32 weeks of treatment with 0.7 mg of fluoride per kg per

day. During the last 3 months he had progressively increasing knee pain on weight bearing.

*Steroid osteoporosis.* H. L., a 52-year-old male gave a history of intermittent aching pain in the joints of the extremities and of increasingly severe and frequent asthma for 23 years. He had been treated with prednisone for 6 years and for the past year had taken 20 mg daily. Our X rays showed slight biconcave deformity of several lumbar vertebrae. The patient was studied before and at the end of 7 months of treatment with 0.9 mg fluoride per kg per day. During the interim his asthma had been severe, and he had usually taken 30 mg prednisone daily. He had increasing knee pain, especially on weight bearing during the last 2 months of treatment. Fat absorption was decreased when measured both before and during treatment.

*Page's disease.* E.M., a 61-year-old female, had a 10-year history of aching pain in the extremities and spine. Osteitis deformans had been recognized because of typical X-ray changes of the skull and pelvis 10 years before. The X-ray appearance of the spine was normal. She was studied before and at the end of 10 months of treatment with 1.0 mg fluoride per kg per day. She experienced pain in the ankle upon weight bearing during the last month of treatment.

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#### Addendum

#### Plasma Fluoride Concentrations of Patients Treated with Sodium Fluoride \*

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Plasma was harvested from heparinized blood drawn before breakfast from patients described in this paper. Specially washed syringes and glassware (1) were supplied to the Seattle workers to reduce the possibility of fluoride contamination, and the plasma was sent frozen to Minneapolis. The analyses for fluoride were carried

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out by the method of Singer and Armstrong (1). The table identifies the patients, briefly summarizes the fluoride treatments, and gives the results of the fluoride determinations.

With the exception of patients R.K. and M.H. no large increases in plasma fluoride concentration were noted, and in these subjects there was only a 4- and 5-fold change, in spite of a 50- to 100-fold increase in intake of fluoride. The remaining patients had only slight analytically significant elevations of plasma fluoride content.

These results were obtained with plasma samples drawn some hours after the last dose of fluoride, and they indicate the plasma fluoride contents at equilibrium conditions after the operation of the homeostatic mechanisms of renal excretion and of skeletal sequestration of this ion. Other work (2) has indicated that about 10% of an oral dose of fluoride is present in the entire plasma volume 1 hour after fluoride ingestion in water.

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#### Fluoride concentration of human plasma

Patient	Plasma fluoride content Control levels		Duration of treatment	Dose of fluoride	Plasma fluoride content during treatment		
	mg/L	mg/L				mg/day	mg/L
R. K.	0.21	0.17	3 days	100	0.79		
			10 days	100	0.85		
			Treatment withdrawn				
			35 weeks		0.30		
			38 weeks		0.24		
M.H.	0.26	0.38	3 days	50	0.65		
			14 days	50	1.77		
			33 weeks	50	0.45		
			34 weeks	50	0.48		
F.F.	0.14	0.19	5 days	50	0.23		
E.M.	0.21	0.19	1 day	60	0.34		
			2 days	60	0.30		
			12 weeks	60	0.25		
H.L.	0.21	0.27	2 days	75	0.27		
			16 weeks	75	0.32		
			24 weeks	75	0.28		

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