Surveys in two areas contribute additional evidence that no hazard of cumulative toxic fluorosis is associated with the use of fluoridated water containing 1 p.p.m. fluoride. Within 1 week, the concentration of fluoride in the urine equaled that in the drinking water for adults. For children, the period of adjustment was considerably longer.

Urinary Fluoride Levels Associated With Use of Fluoridated Waters

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PREVIOUS studies have demonstrated a close correlation between the concentration of fluoride in the urine and the fluoride occurring naturally in drinking water (*1, 2*). Only a limited quantity of data is available, however, concerning the urinary excretion of fluoride added to a municipal water supply (*2–4*). Moreover, it was not determined in any of these studies how soon after beginning the regular ingestion of a fluoride water a stabilized equilibrium is attained, nor was any attempt made to investigate the rate of fluoride excretion in individuals of various ages.

The present study was undertaken, therefore, to provide the following information with respect to fluoridated drinking waters: (*a*) the relation between the level of fluoride in the drinking water and the concentration of fluoride in the urine, (*b*) the time required after fluoridation for this relation to become stabilized, and (*c*) the effect of the age of the individual on the time required for establishment of this equilibrium.

The advent of water fluoridation in Montgomery County, Md., in December 1951 made it possible to obtain such information. Additional, preliminary information was available from Grand Rapids, Mich., where the urinary excretion of fluoride had been under observation since the start of water fluoridation in January 1945. For comparison with the data from these two areas, the results of a survey in 1948 in Aurora, Ill., are presented. The drinking wa-

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John D. Rust and Frederick A. Bullock, formerly with the laboratory, assisted in collecting the specimens in Montgomery County, Md. Virginia L. DuBois, with the Epidemiology and Biometry Branch of the National Institute of Dental Research, assisted in arrangements for collection of the urine specimens in Grand Rapids, Mich. Dr. Albert L. Russell, chief of that branch, made the statistical analysis of portions of the data.

ter in Aurora has contained approximately 1.0 p.p.m. fluoride from natural sources since 1895 (*2*).

Spot Specimens vs. 24-Hour Samples

The difficulties in obtaining 24-hour urine specimens in a study of this extent are obvious. Accordingly, spot specimens from a number of individuals were pooled in equal volumes to provide a sample for fluoride analysis. Early in the study, information was obtained regarding the relation of the fluoride content of spot specimens to that of the 24-hour total volume.

During a 24-hour collection period, spot specimens were taken between 9 and 12 a. m., 2 and 5 p. m., and 8 and 11 p. m., from 9 adult males who were drinking water containing 1.0 p.p.m. fluoride provided as sodium fluosilicate. These samples, as were all samples in the study, were alkalized with calcium oxide prior to evaporation and ashing and analyzed for fluoride according to standard procedures (*5*, *6*). The results are shown in table 1.

The fluoride concentration in a single spot specimen may vary considerably from that in the 24-hour sample. However, according to the *t* test, the mean concentrations for the 3 spot specimens from the 9 men (0.9 p.p.m., 1.0 p.p.m., and 1.1 p.p.m.) did not vary significantly from the mean fluoride concentration of 0.9 p.p.m. for the 24-hour samples.

Table 1. Fluoride concentration (p.p.m.) in spot and 24-hour urine specimens, Montgomery County, Md.

Subject	Spot specimens			Mean of spot specimens	24-hour specimen [1]
	9–12 a. m.	2–5 p. m.	8–11 p. m.		
A	0.3	0.5	0.5	0.4	0.6
B	.4	.5	.5	.5	.5
C	1.5	1.1	1.3	1.3	1.1
D	1.2	1.3	.8	1.1	1.0
E	.8	.9	.8	.8	.8
F	.6	1.5	2.0	1.4	1.2
G	1.5	.8	1.5	1.2	1.2
H	.8	.9	.9	.9	.6
I	1.1	1.5	1.4	1.3	.9
Mean	.9	1.0	1.1	1.0	.9

[1] Corrected for spot specimen analysis.

These results apply to exposure to 1.0 p.p.m. fluoride in the drinking water. They agree very well with the data obtained by Largent and Ferneau for other levels of fluoride (*7*). It appears, therefore, that the fluoride concentration of pooled urine specimens for a group of individuals reflects reasonably well the average fluoride concentration in 24-hour specimens.

Fluoridation of the Drinking Water

Beginning December 28, 1951, the drinking water of Montgomery County, Md., has been fluoridated to approximately 1.0 p.p.m. fluoride by the addition of sodium fluosilicate. The water supplying this area is obtained from the northwest branch of the Anacostia River (*8*). Water samples taken at widely scattered points in the study area for 10-day periods at the time of the urine collections showed approximately 1.0 p.p.m. fluoride after an initial adjustment period lasting 2 to 4 weeks.

The communal water of Grand Rapids, Mich., which is obtained from Lake Michigan, has been fluoridated for more than 10 years with sodium fluoride. The fluoride content has been maintained at a relatively constant level of 1.0–1.1 p.p.m. (*9*).

Sampling Procedures

The subjects studied in Montgomery County were male school children and male adults who had had no exposure to a fluoride-containing drinking water before the start of fluoridation and who were in continuous residence thereafter. Nine age categories, as shown in table 2, were represented. At the start of the study, the subjects in each age category were divided into four groups according to the time of day the specimen was obtained. The same subjects were followed throughout the course of the study. For the age category 5–14 years, the 4 collection periods were 8:30 to 10:30 a. m.; 10:30 a. m. to 1 p. m.; 1 to 2 p. m.; and 2 to 3 p. m. For the age categories 30–34 and 35–39, the first 2 collection periods were the same as for the younger age groups, and the second 2 were: 1 to 3 p. m. and 3 to 5 p. m.

Eleven collections of urine were made during a period of about 3½ years. For the first

10 collections, the subjects remained in their particular group relative to age and time of day. Approximately 10 specimens, 1 from each subject, were obtained for each of the 36 age-time groups. Equal volumes from these 10 specimens were pooled to provide the sample for analysis. No consistent variations in fluoride content with the time of the day the specimen was taken were observed; therefore, only mean fluoride values for each age category are given in this report. In the 11th collection, the specimens were obtained without regard for the time of day and were pooled for each age category to furnish four samples for analysis.

In the Grand Rapids survey, spot urine specimens were obtained from male school children aged 6 through 17 years. Approximately 15 spot specimens comprised each pool, and from 1 to 4 pools were analyzed for each age group. Continuous residents furnished the specimens for the last three collection periods. This survey, which was begun before the Montgomery County survey, did not parallel it in all details.

Fluoride Concentrations in Urine

The data on urinary fluoride concentrations for the Montgomery County residents are shown in table 2. Prior to fluoridation, the fluoride content of the specimens varied from 0.2 to 0.3 p.p.m. As early as 1 week after fluoridation, it was evident that the children were responding quite differently from the adults. At the end of 1 week, the adults' specimens contained 0.7 to 0.8 p.p.m. fluoride. At the end of 6 weeks, they contained the expected 1.0 p.p.m. fluoride (*10*). For these same periods, the fluoride concentration in the urine specimens from the children averaged about half these concentrations, and, although the children varied in age from 5 to 14 years, the fluoride content was essentially uniform for all. As shown by the data for succeeding sampling periods, about 3 years elapsed before the specimens of the children reached 0.9 to 1.1 p.p.m. fluoride.

In table 3, the ratio of urinary fluoride concentration to water fluoride concentration in Montgomery County for each collection period following fluoridation is shown. These ratios illustrate the particularly striking difference between the children and the adults during the first 2 years ($t=8.82$, $P < 0.01$). Equilibrium was reached by the adults after 1 week of fluoridation, whereas approximately 3 years was required for equilibrium to be reached by the children.

The data on urinary excretion of fluoride for the Grand Rapids residents are shown in table 4. The prefluoridation value of approximately 0.2 p.p.m. fluoride in the urine is consistent with the use of the original Grand Rapids water, which contained 0.1–0.2 p.p.m. fluoride. Approximately 2 months after fluoridation was started, the fluoride content of the urine had

Table 2. Fluoride concentration (p.p.m.) in urine specimens, Montgomery County, Md.[1]

Time after fluoridation	Date	Age, in years										
		5	6	7	8	12	13	14	Mean for 5–14	30–34	35–39	Mean for 30–39
0 week	Dec. 15–29, 1951	0.3	0.2	0.3	0.3	0.3	0.2	0.2	0.3	0.3	0.3	0.3
1 week	Jan. 3–5, 1952	.3	.3	.5	.4	.2	.3	.4	.3	.8	.7	.8
2 weeks	Jan. 9–11, 1952	.5	.5	.4	.5	.6	.6	.5	.5	.9	.9	.9
4 weeks	Jan. 23–25, 1952	.4	.4	.5	.6	.6	.6	.6	.5	.9	.9	.9
6 weeks	Feb. 7–11, 1952	.5	.5	.6	.6	.5	.5	.6	.5	1.0	1.0	1.0
10 weeks	Mar. 6–13, 1952	.5	.5	.6	.5	.7	.6	.5	.6	1.0	1.0	1.0
20 weeks	May 14–23, 1952	.7	.6	.5	.6	.6	.6	.7	.6	1.0	.8	.9
1 year, 17 weeks	Apr. 23–28, 1953	.8	.8	.8	.8	.8	.8	1.0	.8	1.2	1.2	1.2
1 year, 49 weeks	Dec. 2–4, 1953	.7	.7	.8	.7	.8	.8	.8	.8	1.1	1.1	1.1
2 years, 48 weeks	Nov. 29–30, 1954	1.1	1.0	1.0	1.1	.9	1.0	1.0	1.0	1.2	.9	1.1
3 years, 24 weeks	May 13–20, 1955	.9	.8	.9	.7	.9	1.0	1.0	.9	1.1	.9	1.0

[1] Fluoridation was begun Dec. 28, 1951.

Table 3. Ratio of urinary fluoride to water fluoride, Montgomery County, Md.

Time after fluoridation	Fluoride in water at tap (p.p.m.)	Urinary fluoride/water fluoride ratio, by age group	
		5–14	30–39
1 week	0.7	0.4	1.1
2 weeks	.8	.6	1.1
4 weeks	1.0	.5	.9
6 weeks	.9	.6	1.1
10 weeks	1.0	.6	1.0
20 weeks	1.0	.6	.9
1 year, 17 weeks	1.0	.8	1.2
1 year, 49 weeks	1.0	.8	1.1
2 years, 48 weeks	1.2	.8	.9
3 years, 24 weeks	.9	1.0	1.1

increased to 0.6 p.p.m. Approximately 5 years after the start of fluoridation, some of the school pupils were excreting 1.0 p.p.m. fluoride (ages 6, 7, and 17). In the final three collections in Grand Rapids (1952, 1954, and 1955), the excretion of fluoride averaged 1.0 p.p.m. for all age groups. The ratios of urinary fluoride to water fluoride would be the same as the mean urinary fluoride values, since the fluoride content of the water has been kept at a practically constant level of 1 p.p.m.

The 1948 survey in Aurora, Ill., provides the most recent data for that city. The fluoride content of urine specimens from boys aged 8 through 17 years varied from 0.8 to 1.0 p.p.m. and averaged 0.9 p.p.m. All age groups showed approximately the same fluoride concentrations. It remains speculative as to when these subjects arrived at this apparently stabilized relation between fluoride in the urine and fluoride occurring naturally in the drinking water. Similar data pertaining to the urinary excretion of fluoride in Aurora residents are presented elsewhere (*10*).

Discussion

Efficient elimination of fluoride by urinary excretion has been demonstrated in several clinical and experimental studies (*1*, *3*, *4*, *10–12*), and is now considered one of the main deterrents to the accumulation of a toxic amount of fluoride in the body tissues. It has been shown also that fluoride is eliminated with equal facility by persons drinking naturally fluoridated water (*1*, *10–12*) and by those drinking artificially fluoridated water (*3*, *4*).

The surveys in Montgomery County and Grand Rapids demonstrated a similar high level of efficiency in the urinary elimination of fluoride added to the drinking water as sodium fluosilicate or sodium fluoride. The findings confirm the results of previous studies, which showed that approximately 1.0 p.p.m. fluoride is found in the urine when fluoride is ingested in drinking water containing 1.0 p.p.m. from natural sources (*10*) or added as sodium fluoride (*3*, *4*). The findings are regarded as evidence that no hazard of cumulative toxic fluorosis is associated with the use of fluoridated water containing 1.0 p.p.m. fluoride.

An important consideration pertaining to water fluoridation is the time required for a stabilized equilibrium between water fluoride and urinary fluoride to be reached. That the period of adjustment can be remarkably short

Table 4. Fluoride concentration (p.p.m.) in urine specimens, Grand Rapids, Mich.[1]

Time after fluoridation	Date	Age, in years												
		6	7	8	9	10	11	12	13	14	15	16	17	Mean
0 weeks	Jan. 17–18, 1945	----	----	----	----	----	----	0.1	0.1	0.2	0.1	0.2	0.3	0.2
7 weeks	Mar. 6–8, 1945	0.6	0.6	0.5	0.7	0.8	0.6	.7	.7	.6	.6	.7	.7	.6
3 years, 18 weeks	May 20–21, 1948	.7	.8	.6	.7	.6	.7	.6	.7	.7	.8	.8	.7	.7
3 years, 43 weeks	Nov. 15–17, 1948	----	----	.7	----	.8	----	.8	----	.8	----	.7	----	.8
4 years, 43 weeks	Nov. 15–17, 1949	1.0	1.0	.6	.6	.8	----	.8	----	.6	.6	.6	1.0	.8
7 years, 37 weeks	Sept. 30–Oct. 7, 1952	.8	1.0	.8	.9	.8	1.0	1.3	.9	.9	1.1	.9	1.1	1.0
9 years, 17 weeks	May 13–14, 1954	1.0	.8	.8	1.0	1.0	1.0	.8	.7	1.0	1.0	.8	.9	.9
10 years, 19 weeks	May 25–26, 1955	.9	.9	.9	.8	1.0	1.0	1.1	1.1	.9	.9	.9	1.0	1.0

[1] Fluoridation was begun Jan. 25, 1945.

is clearly shown by the data for the adults in Montgomery County. As early as 1 week after fluoridation, the fluoride concentration of the urine was approximately that of the drinking water.

There is some indication that the period of adjustment was somewhat shorter for the children of Montgomery County than for those of Grand Rapids. The suggested difference in the two areas cannot reasonably be ascribed to a difference in the fluoridating agents, as it has been shown in studies with the white rat that sodium fluoride and sodium fluosilicate have similar metabolic effects (*13*, *14*). It is possible that the variation in sampling procedures was at least partly responsible for the seeming difference. It is also possible that the difference was due partly to the quantity of water ingested. Water consumption, and thus total fluoride ingested, may have been higher in Montgomery County, owing to the consistently higher mean monthly temperature in that area than in Grand Rapids. The temperature difference averages about 10 degrees throughout the year, according to information from the United States Weather Bureau. It is significant that in both Montgomery County and Grand Rapids the children reached the same state of apparently stabilized equilibrium between urinary fluoride concentration and water fluoride concentration as the adults in Montgomery County, even though they required a somewhat longer period of time.

Speculation as to the reasons for the apparently different response of the children and the adults must turn largely to the possibility of basic differences in fluoride metabolism reflected in the skeletal retention of fluoride. The rapid attainment of equilibrium in adults suggests certain limitations in the capacity of mature bone to deposit fluoride. In this connection it may be noted that the mature rat, without prior exposure to fluoride, stored considerably less fluoride in bones and teeth than did the young growing rat, although they ingested equal amounts of fluoride in their drinking water (*15*). Decreasing amounts of fluoride were stored by rats as the rate of growth diminished, and at maturity a relatively constant level of skeletal fluoride was attained. On the basis of these experimental data, it would seem reasonable for adults to excrete more fluoride than children and hence reach an apparent equilibrium between the concentration of urinary fluoride and water fluoride at an earlier date. The data of the present study thus lend support to the proposition that the retention of fluoride by bone is influenced by its state of maturity.

The length of time required for the children to reach a relatively constant concentration of urinary fluoride suggests some elevation in the fluoride deposited in the skeletal tissues during this adjustment period. However, this increase in retained fluoride would seem to be very small. The urine during the adjustment period contained 0.5–0.6 p.p.m. fluoride, providing for the elimination of substantial quantities of the waterborne fluoride. It has been shown that no adverse effects on the growth and development of the carpal bones accompany the continuous use of natural fluoride drinking water containing approximately 3.0 p.p.m. or more of fluoride (*16*). In addition, examinations of children after 10 years of use of a water supply containing 1.2 p.p.m. fluoride added as sodium fluoride failed to reveal anything unusual with respect to the blood, urine, height, or weight; and roentgenographic studies on the hand, wrist, knees, and lumbar spine of these children showed normal development (*17*). These observations support the conclusion that whatever this increase in the retention of skeletal fluoride by these younger age groups may have been, it was not a health hazard.

Summary

The concentration of fluoride in the urine was determined in individuals of different ages who were drinking water fluoridated to approximately 1.0 p.p.m. fluoride with sodium fluosilicate (Montgomery County, Md.) or with sodium fluoride (Grand Rapids, Mich.).

In adults (Montgomery County), the water fluoride and urinary fluoride concentrations became approximately equal within 1 week after the introduction of the fluoridated water. In school children aged 5 through 14 years (Montgomery County) and 6 through 17 years (Grand Rapids), a considerably longer period of time (approximately 3 and 5 years respectively) elapsed before the concentration of

fluoride in the urine reached that in the drinking water.

The difference in the response of adults and children during the initial period of exposure to a fluoridated drinking water suggests that the maturity of human skeletal tissue influences its capacity to retain fluoride.

The results of the study do not suggest any essential difference in urinary elimination of fluoride ingested in naturally fluoridated drinking water and the elimination of fluoride ingested in drinking water fluoridated with either sodium fluoride or sodium fluosilicate.

The findings are regarded as evidence that no hazard of cumulative toxic fluorosis is associated with the use of a drinking water fluoridated to contain 1 p.p.m. fluoride.

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