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TECHNICAL NOTE

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Determination of Fluoride by Specific Ion Electrode and Report of a Fatal Case of Fluoride Poisoning

Postmortem fluoride levels in fluoride poisoning cases can be measured conveniently with the specific ion or ion-sensitive electrode. Earlier techniques commonly used complexing agents and depended on colorimetric quantitation [*1–6*]. Separation of fluoride from the postmortem specimens was achieved by several means including microdiffusion [*3–6*]. The specific ion electrode measurement of fluoride has been employed directly in urine and other fluids and after separation of the fluoride from biological specimens [*6–10*]. The subjects of this report are a method combining microdiffusion as a separation procedure with specific ion electrode quantitation of fluoride and the results obtained from a case in which this method was used.

Materials

1. 4% NaOH in methanol (w/v)
2. H_2SO_4, concentrated
3. 4*M* potassium acetate pH 5 buffer prepared by diluting 100 ml of 6*M* acetic acid with 50 ml of distilled water, placing the diluted acid in a cool water bath, and adding 50% KOH (w/v) slowly with stirring to pH 5
4. Fluoride reference solutions: A standard solution of 100 mg F^-/100 ml water was prepared by dissolving 221 mg of sodium fluoride, NaF, in 100 ml of deionized water. One to 20 ml of this solution were added to blood, urine, deionized water, and a homogenate of liver and brought to a final volume of 100 ml. The liver homogenate was prepared by adding 75 ml of deionized water to every 25 g of liver in a tissue blender
5. Petri dishes: 35 by 10 mm polystyrene culture dishes (Falcon Plastics)
6. Orion Ionalyzer Model 401 pH/millivolt meter with expanded scale, fluoride ion electrode, and single junction reference electrode.

Method

We found, as Sunshine and Finkle [*5*] had indicated, that, instead of dishes with the classic microdiffusion configuration, plastic petri dishes could be employed for microdiffusion. The great efficiency of sodium hydroxide as a trapping agent for hydrofluoric acid permits substitution of the petri dish lid for the central trapping well of the microdiffusion dish. The lids of the petri dishes were lined up on a tray, 1 ml of 4% sodium

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hydroxide in methanol solution was dropped into each lid, and the sodium hydroxide was dried over a 100°C (212°F) steam table. The standards were run in triplicate. One ml of sample or standard was added to each petri dish and a ml of concentrated sulfuric acid was placed in the same dish so that the two solutions were not yet in contact with each other. Each dish was then placed on a tray and covered with one of the treated lids. The tray was agitated gently to cause the sulfuric acid to mix with the samples and standards, thus releasing the volatile hydrofluoric acid. The petri dishes were then kept on the 100°C table for one hour. At the end of this period the contents of each lid were analyzed, one dish at a time. The lid was removed so that 3 ml of 4*M* potassium acetate pH 5 buffer could be added to it. The residue readily dissolved in the buffer, and the resulting solution was poured into an inverted #9 plastic stopper. A 3 by 9.5-mm Teflon®-coated stirring magnet was added to the solution, and the specific ion electrode and reference electrode were lowered into the inverted stopper over a magnetic stirring base. After 5 min the mV deflection was measured on the pH/mV meter. The electrodes and stirring magnet were then rinsed with deionized water and blotted dry. In this way each microdiffusate was analyzed in turn.

For comparison, direct determinations were also made. These were much easier to perform. Five millilitres of homogenate or fluid were added to a 100-ml plastic beaker. To this 10 ml of 4*M* pH 5 potassium acetate buffer was added. A small magnet and the electrodes were placed in the beaker over the magnetic stirring base. After 5 min the mV deflection was measured on the pH/mV meter. Between determinations the electrodes and magnet were rinsed with deionized water and blotted dry.

Results

The results of measurements made directly and after microdiffusion of the standards are presented in Figs. 1–4. Water and urine give essentially similar curves, whether measured directly or after microdiffusion. Blood and liver standards after microdiffusion produce fluoride curves like those of water and urine. Direct measurements indicate smaller millivolt deflections for the same fluoride concentrations. This may be due to mechanical blocking by tissue debris. It is also consistent with complexing by the fluoride ion with tissue constituents. Cernik et al [*8*] and Taves [*11*] have reported that a portion of fluoride in individuals drinking 1 ppm fluoride is present in urine [*8*] and serum [*11*] in non-ionic or bound form. These authors found no significant binding effect in urine after microdiffusion. The effect we have observed is greatest for low fluoride concentrations and gradually decreases as the fluoride concentrations approach 10 mg/100 ml (Fig. 5). By contrast, after microdiffusion all of the standards produce similar slopes (Fig. 6).

Report of a Fatal Case of Fluoride Poisoning

We employed microdiffusion followed by specific ion electrode measurement in a forensic case. A 57-year-old man was found dead after he ingested rat poison. The time of ingestion is not known. He had vomited a blue-colored material prior to death. His gastric content had a concentration of 26 mg of fluoride ion per 100 ml; his blood, 1.6 mg/100 ml; and his urine, 32 mg/100 ml (Fig. 4).

The values in fluoride cases cited by Hodge and Smith [*12*] (0.4 to 1.0 mg/100 ml for blood, 22 mg/100 ml for urine, and up to 350 mg/100 ml for gastric content) are consistent with the values reported here. Those reported by Morano [*13*] (0.3 mg/100 ml for blood) and those reviewed by Curry [*6*] (0.2 to 0.3 mg/100 ml of blood) are lower than the values reported here.

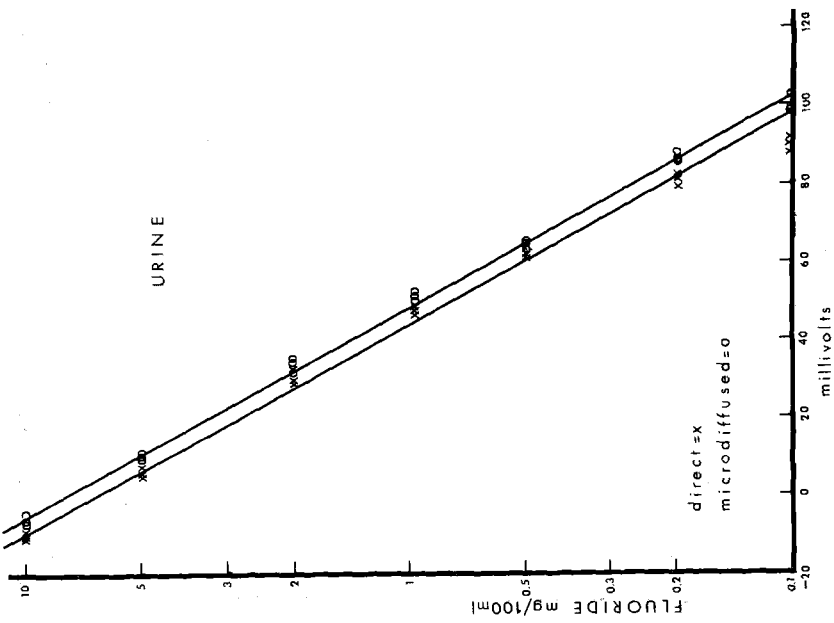

FIG. 2—*Plot of millivolt values for standard concentrations of fluoride in urine.*

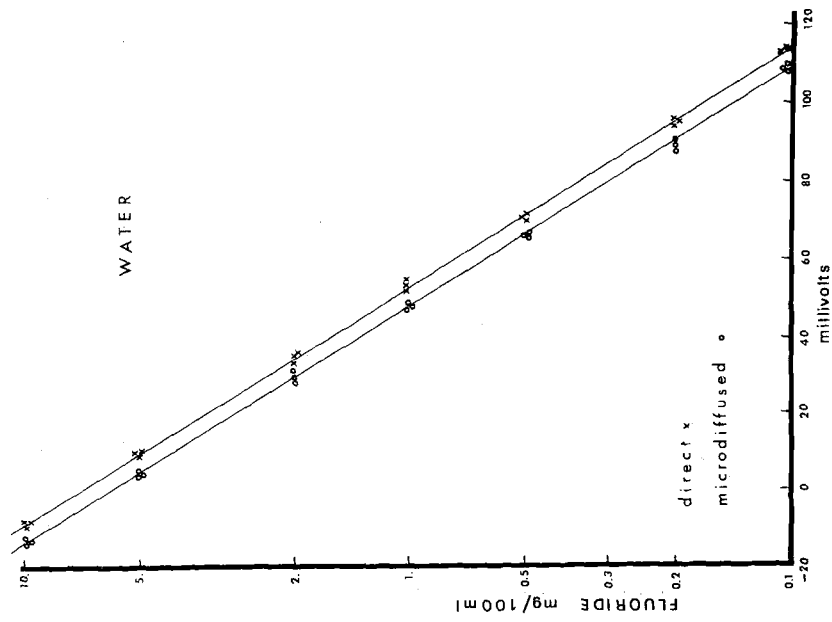

FIG. 1—*Plot of millivolt values for standard concentrations of fluoride in water.*

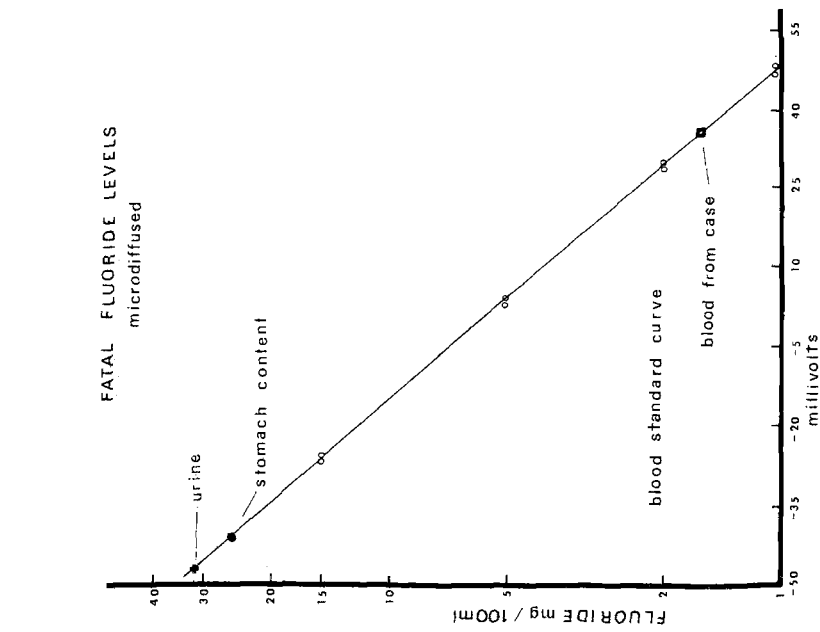

FIG. 4—*Plot of millivolt values for standard concentrations of fluoride in blood and fatal fluoride values in blood, urine, and stomach content.*

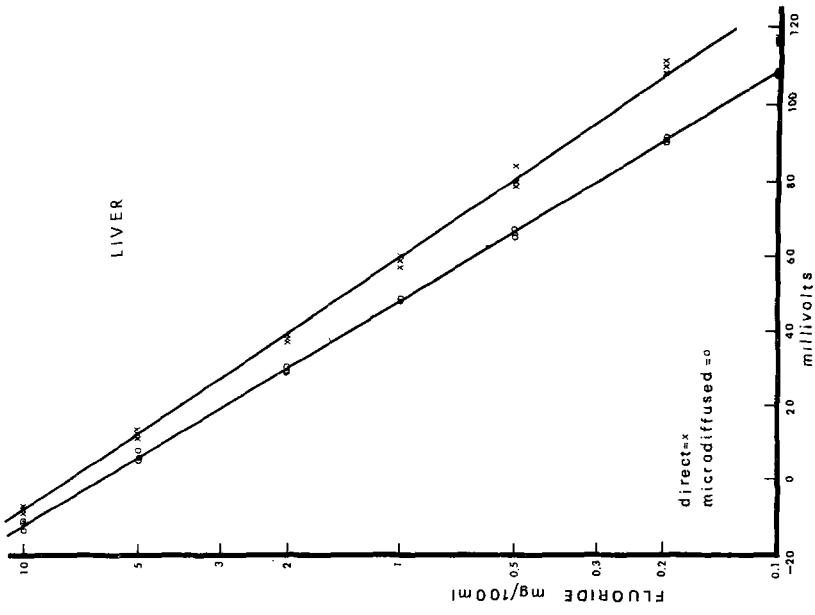

FIG. 3—*Plot of millivolt values for standard concentrations of fluoride in liver.*

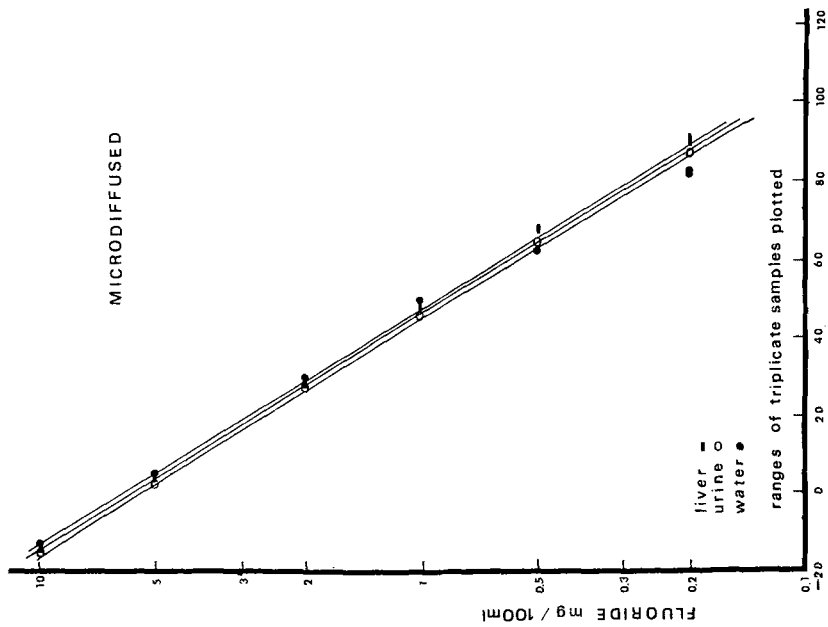

FIG. 6—*Plot of millivolt values for standard concentrations of fluoride in liver, urine, and water after microdiffusion.*

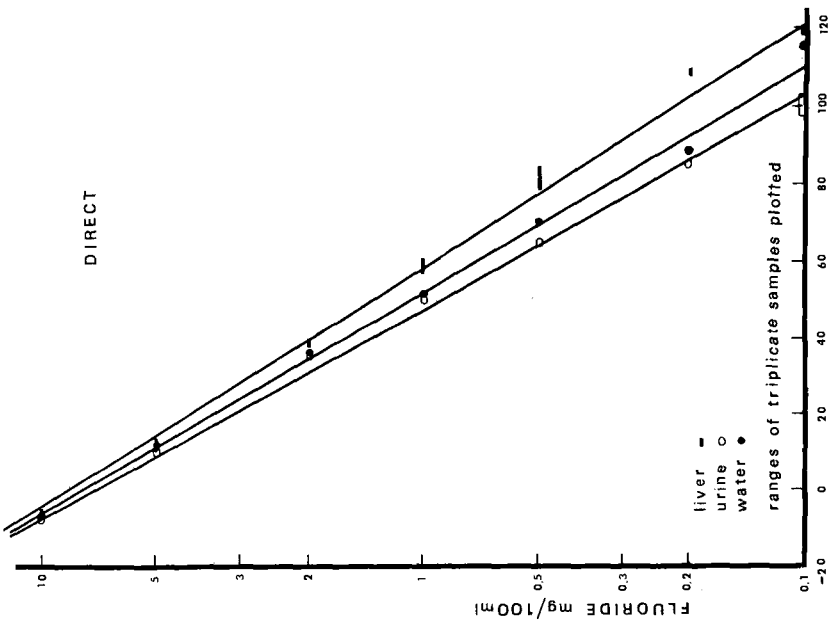

FIG. 5—*Plot of millivolt values for standard concentrations of fluoride in liver, urine, and water by direct immersion.*

When high concentrations are found in urine and stomach content the extra effort of separation by microdiffusion is clearly an unnecessary step. However, the smaller millivolt deflection found by direct measurement in blood and other tissues indicates that prior separation by microdiffusion is more efficient for low fluoride concentrations in tissues.

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