(13) Baggesgaard-Rasmussen, H., Hahn, C., and Ilver, K., *ibid.*, 19, 41(1945).
(14) Baggesgaard-Rasmussen, H., and Ilver, K., *ibid.*, 19, 71(1945).
(15) Baggesgaard-Rasmussen, H., and Lanng, O., *ibid.*, 22, 201(1948).
(16) Baggesgaard-Rasmussen, H., *Ann. Pharm. Franc.*, 10, 639(1952).
(17) Baggesgaard-Rasmussen, H., *Bull. Federation Intern. Pharm.*, 21, 233(1947).
(18) Gottlieb, S., This Journal, 36, 379(1947).
(19) *Ibid.*, 37, 147(1948).
(20) Goodyear, J. M., *et al.*, *ibid.*, 43, 605(1954).
(21) Banes, D., *J. Assoc. Offic. Agr. Chemists*, 44, 323 (1961).
(22) Matsumoto, K., *J. Pharm. Soc. Japan*, 72, 1393 (1952).
(23) Novotry, B., *Cesk. Farm.*, 3, 85(1954).
(24) Jindra, A., Jungr, V., and Zyka, J., *ibid.*, 1, 177 (1952).
(25) Heusghem, C., *Bull. Soc. Chem. Biol.*, 31, 114(1949).
(26) Heusghem, C., *J. Pharm. Belg.*, 5, 3(1950).
(27) Hugelshofer, P., Kalvoda, J., and Schaffner, K., *Helv. Chim. Acta*, 43, 1322(1960).

Technical Articles

Stabilization of Fluoride Solutions in Glass Containers by Aluminum

By J. R. MELLBERG

The addition of sodium fluoride to liquid multivitamin preparations at acid pH in glass containers results in a reaction with glass. The addition of equimolar amounts of aluminum chloride and ethylenediaminetetraacetic acid to an ascorbic acid solution has shown no fluoride–glass reaction over a period of 5 months. Ascorbic acid stability remained good, and *in vivo* absorption of fluoride from a similarly stabilized multivitamin solution was approximately 80 per cent.

The addition of fluoride to liquid vitamin preparations has become widely accepted, especially for pediatric use. It has been shown, however, that when sodium fluoride is added to multivitamin solutions in glass containers at the most acceptable pH range of 2.5 to 4.5, a precipitate, which can be identified as sodium fluorosilicate (Na_2SiF_6) (1), occurs within 24 hr. Because of this, most vitamin–fluoride preparations are dispensed now in plastic bottles. Since glass has certain advantages, such as clarity and lack of oxygen transmission, which are superior to plastic, it was desired to find a way to stabilize fluoride in such containers. Of the usual inorganic elements known to complex fluoride (2), aluminum has the widest use in human medication and is known for its low toxicity. The addition of metals, like aluminum to vitamin solutions, however, is likely to increase the rate of decomposition of ascorbic acid. Therefore, an agent to complex aluminum was needed also. A combination of aluminum ion and ethylenediaminetetraacetic acid (EDTA) was found to provide both ascorbic acid and fluoride stability. Other fluoride or aluminum complexing agents having stability constants of the proper magnitude could be used also with the obvious limitations of low toxicity and solution compatibility.

The physiological availability of the aluminum complexed fluoride was determined *in vivo* by analysis of urinary fluoride after ingestion of known quantities of the vitamin–fluoride preparation.

Table I.—Effect of Various Concentrations of $AlCl_3$ and EDTA on Ascorbic Acid Stability in an Ascorbic Acid Preparation at pH 4.3

mmoles/L. F	mmoles/L. Al	mmoles/L. EDTA	Ascorbic Acid Concn., mg./ml. 4 Days	2 Wk.	1 Mo.	5 Mo.
Room Temperature						
43.9	0	0	50.2	48.6	49.5	43.3
43.9	0	97.8	51.8	50.3	48.1	43.3
43.9	43.9	43.9	52.4	48.6	47.2	43.7
43.9	43.9	0	51.4	49.1	47.2	40.6
43.9	97.8	0	46.2	44.8	43.7	36.6
43.9	195.6	0	38.3	32.9	32.3	28.3
43.9	195.6	195.6	51.0	49.1	48.5	44.2
43.9	195.6	97.8	45.8	41.4	40.1	36.2
50°C.						
43.9	0	0	47.1	39.7	32.7	8.4
43.9	0	97.8	48.4	41.9	33.2	8.9
43.9	43.9	43.9	...	40.7	31.4	9.3
43.9	43.9	0	44.0	35.0	22.7	4.9
43.9	97.8	0	40.9	29.9	15.7	2.8
43.9	195.6	0	33.4	25.2	12.6	4.0
43.9	195.6	195.6	...	42.3	31.4	9.7
43.9	195.6	97.8	...	33.3	20.9	4.4

EXPERIMENTAL

Determination of Ascorbic Acid and Fluoride Stability.—An ascorbic acid solution was prepared by dissolving 35 Gm. of ascorbic acid in 175 ml. of glycerol, 105 ml. of propylene glycol, 52.5 ml. of polysorbate 80,[1] and 200 ml. of distilled water

Received November 5, 1964, from the Research Center, Kendall Co., Barrington, Ill.
Accepted for publication December 10, 1964.
The author acknowledges the technical assistance of Mr. C. Nicholson in the analysis of fluoride.

[1] Marketed as Tween 80 by Atlas Chemical Industries, Wilmington, Del.

TABLE II.—TOTAL 24-HR. URINARY FLUORIDE AFTER INGESTION OF 5 mg. OF FLUORIDE IN LIQUID VITAMIN PREPARATIONS[a]

	Control, mg.	Al-EDTA Stabilized Fluoride–Vitamin, mg.	Aqueous NaF, mg.
	0.558	2.184	3.062
	0.744	2.930	2.827
	1.307	2.309	2.806
Av.	0.870	2.475	2.898
Ingested fluoride recovered, %	...	32.1	40.5
Fluoride absorbed, %	...	79.2	100

[a] Each value represents the average daily elimination of five subjects.

containing 1.29 Gm. of NaF. To 20-ml. aliquots of this solution were added various quantities of aluminum chloride hexahydrate and disodium EDTA in molar quantities proportional to the fluoride. The pH was adjusted to 4.3 with HCl and the volume made up to 25 ml. with distilled water. One-ounce clear glass bottles[2] were filled and stored at room temperature and 50°. After various times, a fresh unopened bottle was analyzed for ascorbic acid by titration with iodine–KI solution. The results of these analyses are listed in Table I.

The effect of aluminum chloride and EDTA on fluoride and ascorbic acid stability in a multivitamin solution was determined in a similar manner. Amounts of aluminum chloride and EDTA equimolar to the fluoride were added to the following solution at pH 4.3:

Methylparaben	0.95	Gm.
Propylparaben	0.11	Gm.
Sodium fluoride	0.995	Gm.
Niacinamide	9.9	Gm.
Riboflavin-5-phosphate	2.32	Gm.
Ascorbic acid	81.0	Gm.
Sodium hydroxide	8.0	Gm.
Thiamine · HCl	1.9	Gm.
Pyridoxine · HCl	1.09	Gm.
Sodium saccharin	2.71	Gm.
Vapad[3]	3.6	Gm.
Glycerol	135	ml.
Polysorbate 80	30.5	ml.
Propylene glycol	81	ml.
Water *q.s.*	540	ml.

The fluoride–glass reaction was noted visually by the presence of inorganic sediment and by etching on the glass surface.

Determination of Fluoride Absorption.—Five subjects each ingested 6.0 ml. of multivitamin preparation containing 5 mg. of fluoride ion with equal molar quantities of aluminum chloride and EDTA. Urine was collected for 24 hr. and analyzed for fluoride according to the method of Megregian (3). After 3 to 4 days, another 5 mg. of fluoride (as aqueous NaF) was ingested by the same subjects, and urine was collected again for 24 hr. and analyzed for fluoride. Normal urine fluoride levels also were obtained from 24-hr. urine collections. Urinary fluoride levels were obtained in triplicate on all subjects for both fluoride preparations and controls. A total of 15 values were therefore obtained for absorption of fluoride from each preparation. The amount of fluoride absorbed was calculated by subtracting the normal 24-hr. fluoride level from the test level. Assuming 100% absorption for sodium fluoride (4) and equal bone retention of fluoride for both preparations, the absorption of the aluminum–EDTA stabilized preparations can be determined. Results of the analysis and calculations are shown in Table II.

[2] Flint Glass; Owens-Illinois Glass Co., Chicago, Ill.

[3] A mixture of vitamin A palmitate and vitamin D_2, S. B. Penick Co., Chicago, Ill.

RESULTS AND CONCLUSIONS

Fluoride Stability.—From visual observation, it was shown that an equal molar amount of aluminum to fluoride in both the ascorbic acid and the multivitamin preparations prevented formation of inorganic sediment and glass etching by the fluoride. Higher molar quantities were not needed, and lower amounts were usually only partially effective.

Ascorbic Acid Stability.—Table I shows that an increase in the quantity of aluminum resulted in a greater loss of ascorbic acid. However, this was prevented by the addition of an equimolar amount of EDTA. An amount of EDTA less than equimolar did not appear to be sufficient, while an excess of that needed to chelate aluminum did not increase ascorbic acid stability and therefore was considered unnecessary.

After 2 months of aging at room temperature, a decrease in ascorbic acid content was found in the multivitamin solution containing no aluminum or EDTA from 150 to 113 mg./ml., while the same solution containing the stabilizers decreased to 118 mg./ml., again indicating no deleterious effect of EDTA chelated aluminum on ascorbic acid stability.

Fluoride Absorption.—Table II clearly shows that about 80% of the ingested fluoride was absorbed by the body; the 20% not absorbed is likely due to the presence of aluminum ion, since this has been known to reduce urine fluoride levels (5–7) by preventing absorption. However, to prevent absorption completely, larger amounts would be required.

The known caries potentiating effects of EDTA (8, 9) thought to be caused by chelation of tooth enamel calcium should not be observed with the aluminum EDTA chelate, since its log stability constant is 16.1 (10), while that of calcium EDTA is only 10.6 (10). In addition, aluminum is chelated strongly at the acid pH's used for vitamin solutions, while calcium requires a neutral or optimal pH 10 to 11 environment for strong chelation.

REFERENCES

(1) Tingstad, J. E., MacDonald, L. H., and Meister, P. D., THIS JOURNAL, **52**, 343(1963).
(2) Fritz, J. S., Garralda, B. B., and Karraker, S. K., *Anal. Chem.*, **33**, 882(1961).
(3) Megregian, S., *ibid.*, **26**, 1161(1954).
(4) Largent, E. J., "Fluorosis," The Ohio State University Press, Columbus, Ohio, 1961, p. 23.
(5) Venkataramanan, K., and Krushnaswamy, N., *Ind. J. Med. Res.*, **37**, 277(1949).
(6) Hodge, H. C., *J. Am. Med. Assoc.*, **177**, 109(1961).
(7) Becker, D. E., *et al.*, *J. Animal Sci.*, **9**, 647(1950).
(8) Wandelt, S., *Deut. Zahnaerztl. Z.*, **14**, 1255(1959).
(9) Zipkin, I., *J. Am. Med. Assoc.*, **177**, 310(1961).
(10) Welcher, F. J., "The Analytical Uses of Ethylenediaminetetraacetic Acid," D. Van Nostrand Co., Princeton, N. J., 1957, p. 6.