Investigation of Aluminum Blood Levels in Man After Oral Administration of an Aluminum-Containing Complex, Potassium Glucaldrate

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The effect of oral administration of a water-soluble aluminum complex, potassium glucaldrate, on the blood levels of the metal in humans was investigated. Aluminum blood levels in humans after the oral administration of an insoluble aluminum compound, aluminum hydroxide, were studied also. A fluorometric procedure was utilized for analysis of aluminum in the biological material. No significant increase in aluminum blood level was detected after the oral ingestion of single or multiple doses of either aluminum compound in normal subjects or in subjects diagnosed as requiring antacid therapy.

POTASSIUM GLUCALDRATE is a water-soluble aluminum complex prepared from potassium aluminate and glucono δ lactone. This metal complex exhibits desirable buffer antacid properties in a variety of *in vitro* tests. The evaluation of the *in vitro* antacid properties of a closely related substance, sodium gluconatodihydroxoaluminate(III), has been reported recently by Grossmith (1). The present investigation is concerned with the effect of oral administration of a water-soluble aluminum complex of this type on the aluminum blood levels of humans.

Previous investigations on the aluminum content of biological samples generally have utilized colorimetric and spectrographic methods of analysis. Underhill and Peterman (2) reported a colorimetric method for the analysis of aluminum, based on the formation of an alizarin complex, in various biological materials including blood. Wolff (3) employed a spectrographic technique to determine the aluminum levels in human blood and reported the average aluminum blood level to be 0.54 mcg./ml., with a range of 0.21 to 0.94 mcg./ml. Kehoe *et al.* (4) also used a spectrographic procedure for the determination of aluminum in numerous biological samples, including blood plasma.

The colorimetric analysis of aluminum based on the formation of a complex between aluminum and 8-hydroxyquinoline was described by Gentry and Sherrington (5). Goon *et al.* (6) extended the sensitivity of this method by utilizing fluorescence of the complex in a chloroform solution as the analytical measurement. Rubins and Hagstrom (7) also employed the fluorescence of a chloroform solution of the aluminum–8-hydroxyquinoline complex for determination of small amounts of the metal in plant tissue. A modification of the Rubins and Hagstrom procedure was used in the present investigation for the determination of aluminum in blood. Although the method described is not useful for the determination of aluminum at the levels normally present in blood, it is capable of detecting increases in the blood level of the metal of the order of 1 mcg./ml. and was satisfactory for the objectives of the study.

The purpose of this investigation was to study the effect of an orally administered aluminum compound on aluminum blood levels in humans, specifically to determine if any increase or accumulation of aluminum in blood occurs after oral administration of aluminum-containing compounds. A water-soluble aluminum complex, potassium glucaldrate, and a water-insoluble aluminum compound, aluminum hydroxide, were employed in the study. Aluminum determinations in blood were made by a fluorometric procedure after single and multiple dose regimens in normal subjects and subjects diagnosed as requiring antacid therapy.

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EXPERIMENTAL

The determination of aluminum in blood was made by a modification of the fluorometric procedure described by Rubins and Hagstrom (7) for the determination of aluminum in plant tissues. Several blood samples also were analyzed for aluminum by a spectrographic technique.

Reagents

Concentrated Nitric Acid.—Concentrated nitric acid, reagent grade.

Perchloric Acid.—Perchloric acid, 70–72% reagent grade.

Buffer Solution A.—Dissolve 200 Gm. of ammonium acetate and 100 ml. of glacial acetic acid in distilled water. The final volume is made up to 1000 ml. with distilled water.

Hydroxylamine Solution.—This solution is a 10% aqueous hydroxylamine hydrochloride solution.

Bathophenanthrolein Solution.—Dissolve 250 mg. of bathophenanthrolein (G. Frederick Smith Chemical Co., Columbus, Ohio) in 500 ml. of 95% ethyl alcohol; store the solution in a refrigerator.

Isoamyl Alcohol.—Isoamyl alcohol, reagent grade.

Buffer Solution B.—Dissolve 200 Gm. of ammonium acetate and 70 ml. of concentrated ammonium hydroxide in distilled water. The final volume is made up to 1000 ml. with distilled water.

8-Hydroxyquinoline Solution.—Dissolve 20 Gm. of 8-hydroxyquinoline in 60 ml. of glacial acetic acid and sufficient distilled water to make the final volume 1000 ml.

Chloroform.—Chloroform, reagent grade, is used.

Fluorometric Procedure

Ten milliliters of concentrated nitric acid was added to a 10-ml. sample of blood and heated gently on an electric heater until the first reaction subsided. Two milliliters of 70–72% perchloric acid was added, and the digestion was continued while the heat was increased gradually until fumes of perchloric acid were given off. Two milliliters of concentrated nitric acid was added and the digestion continued. Again,

nitric acid was added when perchloric acid fumes appeared; finally, the sample was fumed at full heat for several minutes. The flask was removed, cooled, and 50 ml. of distilled water was added and the mixture heated to boiling. The digested solution was transferred to a 100-ml. volumetric flask, cooled, and made up to volume with distilled water. A Thomas-Labenco high-temperature Kjeldahl digesting apparatus was utilized in the digestion procedure.

The digestate was filtered, and a 10-ml. aliquot was placed in a 60-ml. bottle fitted with a polyethylene cap and diluted to 20 ml. with distilled water. Two milliliters of buffer solution *A*, 2 ml. of 10% hydroxylamine hydrochloride solution, and 5 ml. of bathophenanthrolein solution were added. This mixture was allowed to stand at room temperature for 90 min. and extracted with 15 ml. of isoamyl alcohol by shaking for 10 min. The isoamyl alcohol was removed by aspiration, and the extraction was repeated with a second 15-ml. portion of isoamyl alcohol. The samples were centrifuged; the organic solvent was removed by aspiration. Five milliliters of buffer solution *B* was added to the aqueous layer remaining, and the pH was adjusted to 9, if necessary, with a 1:1 ammonium hydroxide solution. Two milliliters of 8-hydroxyquinoline solution was added, and the mixture was allowed to stand for 10 min. and extracted with 10 ml. of reagent grade chloroform by shaking for 10 min. The aqueous layer was removed by aspiration, and the chloroform layer was washed with 15 ml. of buffer solution *A*. The mixture was centrifuged; the aqueous layer was removed by aspiration. Two milliliters of the chloroform solution was diluted to 25 ml. with reagent grade chloroform, and the fluorescence of this solution was measured at 520 mμ after activation at 400 mμ. An Aminco-Bowman spectrophotofluorometer was used for all fluorescence measurements in this study.

Spectrographic Procedure

A 0.1-ml. sample of blood was added to the craters of spectroscopically pure graphite electrodes and dried in a hot air oven. The electrodes were burned in a 220 d.c. arc while being photographed through a large Bausch & Lomb Littrow spectrograph. The aluminum lines consisted of three pairs at wavelengths 2568, 2575, 2652, 2660, 3082, and 3092 Å. The line pair at 3082 and 3092 Å. was the most sensitive and the only one visible at the low aluminum concentrations present in the samples investigated.

Drug Administration

The effect of a single oral dose of an aluminum containing antacid on the aluminum blood level in normal humans was determined by the following procedure.

Sixteen subjects were divided into two groups of eight each. One group, subjects *1* through *8*, received 60 ml. of a potassium glucaldrate solution which contained 2.1 Gm. of aluminum. The second group, subjects *9* through *16*, were dosed with 100 ml. of an aluminum hydroxide gel which contained 2.1 Gm. of aluminum. Blood samples were drawn at 0, 1, 2, 4, and 8 hr. post administration from all subjects in the study. In addition, samples were obtained at 12 hr. post administration in subjects *1* through *4* and *9* through *12* and at 24 hr. after the dose in the remaining subjects. The zero-hour sample was used as a blank value, and the increase in aluminum content in the subsequent samples was determined by the fluorometric procedure described. In some instances, the spectrographic analysis also was utilized.

The effect of multiple-dose oral administration of aluminum containing antacid products on the aluminum blood levels of humans diagnosed as requiring antacid therapy was determined in the following manner.

Each subject in the study received 20 ml. of the antacid preparation under investigation four times a day at 10:00 a.m., 2:00 p.m., 4:00 p.m., and 8:00 p.m. Those subjects dosed with potassium glucaldrate received 0.70 Gm. of aluminum at each administration, while the subjects dosed with the aluminum hydroxide preparation received 0.42 Gm. of aluminum per dose. During the study, blood samples were drawn on various days late in the afternoon after the third dose, and the blood analyzed for aluminum by the fluorometric procedure. Blood samples were drawn 2 days and 1 day before medication was started to establish the blank value. The remaining samples were obtained on the first, third, seventh, fourteenth, twenty-first, and twenty-eighth days of administration.

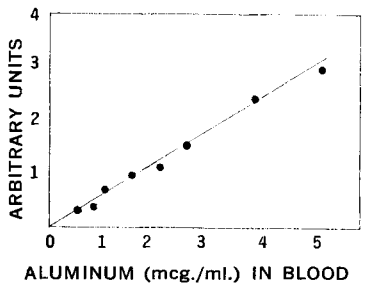

Fig. 1.—Standard curve for the determination of aluminum in human blood.

TABLE I.—INCREASE IN ALUMINUM BLOOD LEVELS (mcg./ml.) AFTER ORAL ADMINISTRATION OF POTASSIUM GLUCALDRATE SOLUTION IN NORMAL HUMANS

Subject	Time, hr. 1	2	4	8	12	24
1	0	0	<3	0	<3	..
2	0	0	0	0	<3	..
3	0	<3	0	0	7	..
4	13	<3	7	3	0	..
5	0	0	0	0	..	0
6	0	0	<3	3	..	6
7	0	<3	0	0	..	0
8	<3	<3	4	0	..	0

TABLE II.—INCREASE IN ALUMINUM BLOOD LEVELS (mcg./ml.) AFTER ORAL ADMINISTRATION OF ALUMINUM HYDROXIDE GEL IN NORMAL HUMANS

Subjects	Time, hr. 1	2	4	8	12	24
9	<3	0	<3	7	0	..
10	0	<3	4	3	<3	..
11	0	<3	0	0	0	..
12	0	0	0	8	6	..
13	<3	<3	0	0	..	<3
14	0	0	<3	<3	..	0
15	0	0	0	0	..	4
16	<3	<3	<3	0	..	0

TABLE III.—INCREASE IN ALUMINUM BLOOD LEVELS (mcg./ml.) AFTER ORAL ADMINISTRATION OF POTASSIUM GLUCALDRATE SOLUTION TO HUMANS REQUIRING ANTACID THERAPY

Subject	Time, Days 1	3	7	14	21	28
A	0	1	1.2	0	0	0
B	0	0	1.9	1.6	<1	0
C	1.1	1.6	1.7	2.1	0	0
D	1.7	<1	<1	0	0	0
E	0	0	3.2	0	0	0
F	1.1	1.2	0	0	3.2	4.4
G[a]	1.6	<1	<1	<1	<1	..
H	0	2.8	5.4	2.1	<1	0

[a] A blood sample from this subject was obtained on the thirty-fourth day of medication and showed no increase in the aluminum blood level.

TABLE IV.—INCREASE IN ALUMINUM BLOOD LEVELS (mcg./ml.) AFTER ORAL ADMINISTRATION OF ALUMINUM HYDROXIDE GEL TO HUMANS REQUIRING ANTACID THERAPY

Subject	Time, Days 1	3	7	14	21	28
A	0	0	0	0	1.1	2.9
B	0	0	0	0	0	0
C	0	0	3.8	4.0	0	1.0
D	0	<1	0	0	0	0
E	0	0	0	0	0	0
F	0	0	0	0	0	1.3
G	2.7	2.1	1.5	1.8	1.2	1.1

RESULTS AND DISCUSSION

The fluorometric procedure utilized in this investigation for the analysis of aluminum is not sufficiently sensitive to determine the levels of aluminum present in normal human blood. However, the method described is useful for the detection of increases in aluminum content in the blood of the order of 1 mcg./ml. For the purpose of the study, this sensitivity was considered satisfactory. The spectrographic technique described was also insensitive to concentrations of the metal below 1 mcg./ml. of blood.

Figure 1 illustrates a standard curve for the determination of aluminum in whole human blood. Both inorganic and organic aluminum compounds were utilized in preparing the aluminum standards. The amount of aluminum indicated on the graph is that added to the blood sample analyzed.

Tables I and II show the results of an investigation on the effect of a single oral dose of an aluminum containing antacid on the aluminum blood level in normal humans. Due to the use of smaller samples in these series of analyses, the limit of detection of an increase in aluminum content was of the order of 3 mcg./ml.

The data show that, in most instances, any increase noted in aluminum blood levels after oral administration of relatively large amounts of aluminum in the form of water-soluble or water-insoluble antacids are below the sensitivity of the analytical procedure employed. The isolated cases where the aluminum level is significantly greater than the blank value might be due to contamination since the earlier and/or later samples in the same subject show no consistently high levels of aluminum. It should be noted that subjects *2*, *3*, *4*, and *5* experienced nausea and vomiting 15 to 30 min. after administration of this high dose of potassium glucaldrate solution, and the amount of drug retained was unknown.

The blood samples obtained from subjects *6*, *7*, and *16* also were analyzed by means of the spectrographic procedure described; no increase in aluminum levels over those noted in the zero-time sample was observed.

Tables III and IV summarize the results of an investigation on the effect of multiple-dose oral administration of aluminum-containing antacid products on the aluminum blood levels in humans diagnosed as requiring antacid therapy.

The results are similar to those observed after single-dose administration because, in most instances, any increases in aluminum blood levels noted were below or near the limits of sensitivity of the assay procedure. The several samples where relatively high aluminum levels were noted were isolated in that no accumulation or continued high levels of the element were observed in previous or later samples.

This investigation was designed to determine the effect of oral administration of alaminum-containing antacids on aluminum blood levels. Specifically, the experiment was to determine if any elevation or accumulation of aluminum in blood was observed after large single oral doses or after multiple-dose administration of a soluble aluminum complex, potassium glucaldrate. A similar study utilizing an insoluble aluminum antacid, aluminum hydroxide gel, was conducted also. The analytical procedure described was adequate to show increases in aluminum blood levels of greater than 1 mcg./ml. From the data presented, there is no apparent increase or accumulation of aluminum in the blood of normal subjects or those diagnosed as requiring antacid therapy after oral administration of the soluble or insoluble antacids employed.

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