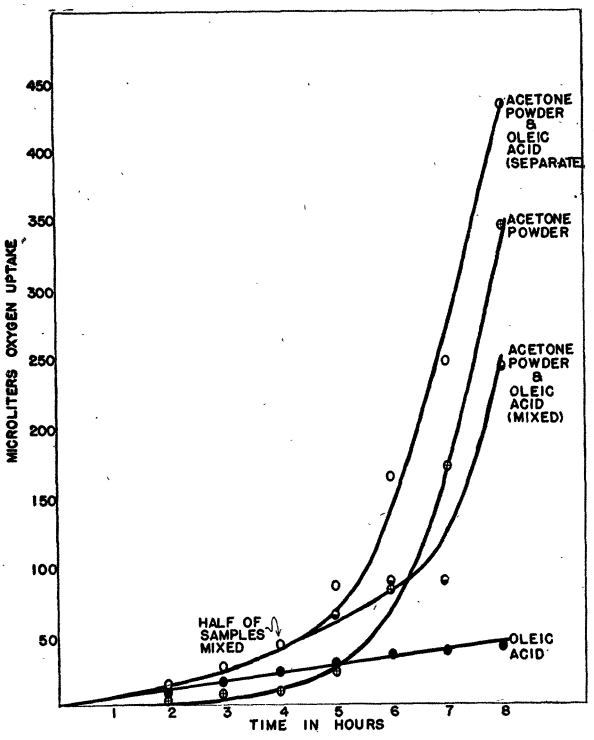

FIG. 2. Qualitative comparison by oxygen uptake of an acetone powder of chicken fat tissue and oleic acid.

as used for these experiments was the fat that melted free of the fat tissue at room temperature. Ten grams of each of the designated 5 substances was homogenized for 1 min with 30 ml of phosphate buffer of pH 7.6 in a Waring Blendor. All samples, except the chicken fat tissue, were boiled under nitrogen for 5 min in order to destroy any enzyme system that may have been present. Samples were then run on the Warburg respirometer, using the direct method, in an atmosphere of air at 37° C.

As can be seen from Fig. 1, the oxygen uptake for the fat tissue was greatly accelerated after 8 hr. Oleic acid, cod-liver oil, wheat-germ oil, and chicken fat all exhibited a progressive linear oxygen uptake. These results could not be accounted for by contamination with microorganisms.

The conclusion drawn from this was that the rapid oxygen uptake of the fat tissue system was dependent on the presence of tissue rather than fat. In order to further verify this conclusion, 45 g of finely chopped fresh chicken fat tissue was homogenized with cold acetone. The precipitate was washed 4 times in cold acetone, dried for 1 hr at 37° C, and resuspended in 270 ml of phosphate buffer of pH 7.6. A qualitative comparison of this suspension with a 2% homogenate of oleic acid was then made.

The acetone powder from the fat tissue took up oxygen very rapidly after an induction period of about 5 hr as is shown in Fig. 2. The primary difference between the oxygen-uptake curve of the chicken fat tissue (Fig. 1) and that of the acetone powder (Fig. 2) is that the fat tissue required a longer induction period before rapid oxidation started. This is possibly due to the presence of fat in the suspension. Additional evidence is that a homogenate of oleic acid lengthened the induction period of a suspension of the acetone powder. This is shown in Fig. 2. Boiling the acetone powder results in loss of activity.

The precise nature of this oxidative system is unknown. The authors do not believe that it can be explained as the autoxidation of a simple fat or fatty acid.

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Enhancement of Lead Excretion in Humans by Disodium Calcium Ethylenediamine Tetraacetate[1, 2]

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Ethylenediamine tetraacetic acid or Versene (EDTA), is a synthetic chelating agent used industrially on a large scale for the control of cations in solution. The compound forms strong, un-ionized, soluble chelate complexes with cations, especially those of the di- and trivalent type. In this way many characteristic effects of metal ions in solution may be controlled. When administered to animals and humans intravenously, the primary acute action of the compound in large doses over short periods was to lower the systemic calcium levels (*1, 2*). At extreme dosage hypocalcemic tetany was observed in animals. In contrast to this effect of EDTA, the administration of the preformed calcium chelate was without effect on calcium homeostasis (*3*). The preformed calcium EDTA was also singularly nontoxic by all routes (*4, 5*). The absence of toxicity of the compound may be attributed to its physiological inertness, since 99% of the material tagged in the methylene position with radioactive carbon could be recovered from the urine after intra-

[1] Trade name "Disodium Calcium Versenate"—Bersworth Chemical Co., Framingham, Mass.

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TABLE 1

EFFECT OF CALCIUM *EDTA* ON URINARY LEAD EXCRETION IN HUMANS

Day	Patient A			Patient B			Patient C[8]			Patient D		
	Treatment	mg Pb/l	Total mg Pb excreted	Treatment	mg Pb/l	Total mg Pb excreted	Treatment	mg Pb/l	Total mg Pb excreted	Treatment	mg Pb/l	Total mg Pb excreted
1	None	3.13	1.06	None	< 0.20	—	None	0.42	0.12	0.5 g	0.58	1.01
2	None	2.93	1.14	None	< 0.20	—	None	< 0.20	—	0.5 g	7.12	6.98
3	None	—	—	None	< 0.20	—	0.5 g	2.63	1.53	0.5 g	3.93	6.07
4	None	—	—	None	< 0.20	—	0.5 g	2.02	1.47	0.5 g	1.96	3.45
5	None	1.34	0.47	None	< 0.20	—				0.5 g	2.17	2.93
6	None	2.74	0.49	0.5 g	2.40	1.33				None	< 0.20	—
7	0.5 g	4.62	0.93	0.5 g	2.76	1.81				None	0.27	0.43
8	0.5 g	7.19	3.06	0.5 g	1.50	*				0.5 g	1.63	3.50
9	0.5 g	7.19	3.06	None	1.90	1.64				0.5 g	2.17	1.35
10	None	0.27	0.05	None	< 0.20	—				0.5 g	0.46	0.86
11	None	3.10	1.13	1.0	1.22	1.35				0.5 g	1.04	0.84
12	None	1.50	0.23	1.0 g	3.20	1.36				None	< 0.20	—
13	None	0.83	0.05							None	0.59	0.49
14	1.0 g	1.99	1.22							0.5 g	< 0.20	—
15	1.0 g	6.64	3.45							0.5 g	0.27	0.42

* Incomplete collection.

venous administration (*6*) to rats. The favorable pharmacological properties of the preformed calcium EDTA chelate, combined with the fact that calcium in this complex may be displaced by other metals, including lead (*7*), led us to examine its possibilities in metal poisoning. The detoxicant and antidote action of calcium EDTA in experimental nickel, copper, cadmium, lead, and cobalt poisoning was reported (*8*). Marked enhancement was demonstrated of the excretion of lead from rabbits given intravenous lead acetate (*8*) followed by intravenous calcium EDTA. Since this report, we have been able to study the effect of this compound on eleven cases of acute and chronic lead poisoning in humans. The preliminary clinical findings have been presented (*9, 10*) and a more extensive description of the medical observations is in preparation. The present report deals with the effect of the compound on lead excretion in humans.

Urine collections in the cases of young and comatose patients was made by indwelling catheter. The samples were collected in acid-washed Pyrex bottles. Occasional samples in these 24-hr collections were lost by accidental spillage. While the concentration of lead in these partial samples has been recorded, the total excretion has been omitted from the tabulation. Lead analysis was carried out on 25-ml aliquots of the total urine samples. The method of analysis was as follows: To the 25-ml aliquot of urine was added 1 ml of "low lead" sulfuric acid and 5 ml of conc. nitric acid. The sample was evaporated to about 3 ml on a hot plate. The nitric acid addition and evaporation was repeated until all readily oxidized material was gone. This usually required a total of three to four nitric acid evaporation repetitions. A final ashing was conducted after the addition of 1 ml of perchloric acid and 3 ml of nitric acid. The cooled residue of ashed sample was then analyzed for lead by the standard dithizone colorimetric procedure (*11*). With every unknown sample, simultaneous analysis of a reagent blank, recovery of a standard sample of lead, and recovery of an added known amount of lead in urine was conducted. The analytical method gave recoveries of 98–102% in the range of 0.2 to 12 mg/l of lead.

In Table 1 are recorded the urinary lead excretions of four patients treated with intravenous calcium EDTA. Patients A, B, and C[8] were children of ages 3–5. Patient D was a male adult. Patients A and D were started on the drug while in an acute phase of lead poisoning as was indicated by the clinical symptoms. Patient B had survived an acute episode of lead poisoning and was treated two weeks subsequent to this event. Patient C, suffering from chronic lead poisoning, was treated a week following exacerbation of the symptoms due to respiratory ailment. The data reported indicate that administration of calcium EDTA resulted in marked enhancement of the lead excretion in these patients.

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[8] This patient was treated by G. Ahrens, Babies Hospital, New York. We wish to thank Dr. Ahrens for the urine samples.