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THE ACUTE DISTRIBUTION OF INTRAVENOUSLY ADMINISTERED LEAD ACETATE IN NORMAL AND *BAL*-TREATED RABBITS

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Little information is available about the actions of 2:3-dimercaptopropanol (BAL) in poisoning by lead salts, and so far as is known nothing has been published about its effect on the distribution of lead in the body. This paper provides some new observations on the distribution of lead, indicated by the use of Pb^{212} (thorium B) as a tracer, and on changes in the distribution after the administration of BAL.

METHODS

Radioactive lead was obtained from the deposit on a charged button exposed to a thorium source. A button with Pb^{212} (intensity of about 1 millicurie) was heated gently in a slightly acidified 0.01 per cent (w/v) solution of lead acetate, so that the Pb^{212} deposit was exchanged for part of the lead in the solution. For administration to rabbits a solution was prepared such that the volume injected (2.0 ml.) contained 2.07 mg. of lead (as acetate) per kg. body weight (i.e. 0.01 mM./kg.) incorporating 100–300 microcuries of Pb^{212} dissolved in 4 per cent (w/v) dextrose. Young rabbits of both sexes and various breeds and of average weight 1.3 kg. were used. Injections were made into the marginal vein of one ear. After injection the rabbits were placed in metabolism cages and allowed access to water but not food. Their bladders were emptied by suprapubic pressure before injection and six, twelve, and eighteen hours afterwards, unless the rabbits were killed earlier. The animals were killed by a blow on the head one, six, or twenty-four hours after the injection of lead acetate. Solutions of BAL were freshly prepared in 66 per cent (v/v) aqueous propylene glycol and were injected into the paravertebral muscles as discussed below.

Immediately after death the thorax was opened, and blood was collected by bleeding from the great veins. Clotting was prevented with heparin, and the samples were centrifuged immediately. The organs were then dissected, starting with those in which a low content of lead was expected, and taking care to avoid contamination between different tissues. Complete organs were washed with water, dried of superficial moisture between filter papers, and weighed; they were then chopped finely and, in organs weighing less than 10 g., ashed entire. From other tissues samples weighing 2 to 10 g. were ashed. Liver samples were taken from well-mixed choppings of the entire organ. Muscle samples were taken from the outer part of the thigh. Bone epiphysis and diaphysis were obtained from the long bones of one hind limb and one fore limb. Marrow was completely removed from the central cavities of these bones and estimated separately. The samples of diaphysis were therefore practically marrow-free; the values for epiphysis, ribs, vertebrae, and skull vault were influenced by the amount of marrow contained in the cancellous tissue. The injected ear and the small piece of cotton-wool used to control bleeding were taken as an independent sample, and in the majority of experiments the uninjected ear was taken as a control. The amount of lead lost by leakage at the site of injection could therefore be assessed; the average amount was slightly more than 2 per cent of the dose. Duplicate samples were taken from the liver and one other organ; from these duplicates an estimate of the error of the lead determinations was made.

After being weighed the tissues were placed in long-necked flasks containing 40 ml. concentrated nitric acid and 20 ml. 60 per cent (w/w) perchloric acid, and allowed to stand for at least 30 minutes. The flasks were then heated cautiously until the contents were boiling, care being taken to prevent the formation of excessive amounts of foam. The tissues dissolved and the solutions were boiled gently until about 20 ml. remained, when a further 30 ml. of concentrated nitric acid was added. Boiling was continued until about 10 ml. of clear solution remained. In all, the solutions were boiled for 1½ to 3 hours. If charring occurred during this process the flasks were allowed to cool and a few drops of fuming

nitric acid or hydrogen peroxide were added until a clear solution was obtained.

The contents of the flasks were washed into beakers and the volume made up to about 100 ml. with water. The reaction of the solution was adjusted to neutrality with 40 per cent (w/v) sodium hydroxide (using B.D.H. Universal Indicator); 0.25 ml. of concentrated hydrochloric acid was then added, followed by 0.5 ml. of 1.12 per cent (w/v) lead acetate. The lead was then precipitated as lead sulphide by the addition of 1.0 ml. of 10 per cent (w/v) sodium sulphide. In order to avoid the precipitation of calcium phosphate from ashed bone samples on the addition of the 40 per cent sodium hydroxide, a slightly different procedure was adopted: the alkali was added until the phosphate began to be precipitated and then just enough concentrated hydrochloric acid to redissolve it was added with vigorous stirring; lead acetate and sodium sulphide were then added as described above.

The precipitates of lead sulphide were allowed to stand for at least one hour before filtering under moderate suction in small Hirsch funnels. The precipitates were collected on small circles of filter paper 2 cm. in diameter. They were washed once with distilled water and once with acetone to carry down the portion adhering to the sides of the funnel and to facilitate drying. The papers were removed when dry and the funnels were washed, first with a small quantity of concentrated hydrochloric acid and then with distilled water. The filtrates and funnel washings were then neutralized, hydrochloric acid, lead acetate, and sodium sulphide were added, and the fresh precipitates were filtered as described above.

The radioactivity of the precipitates so obtained on paper circles 2 cm. in diameter was estimated by means of a Geiger-Müller counter of the usual bell-shaped type with a thin, supported mica window, a paper screen being used to absorb the α particles. The counter was calibrated at each experiment with samples prepared from aliquots of the solution used for injection by precipitation with a lead sulphide carrier as described above. In preliminary experiments it was found that the total count per minute, corrected for the background, was proportional to the amount of radioactive solution used, over the wide range employed in the biological experiments, and that appreciable changes were not produced by doubling or halving the amount of lead carrier used.

From the counts obtained on aliquots of the injected solution and the counts on individual samples under identical conditions, the proportion of the dose per gramme of tissue, and hence the lead concentration per gramme of tissue, were calculated. The total amounts of lead in organs of known weight were calculated from the data, and approximate values were obtained for other tissues on the assumptions that the total blood volume was 70 ml./kg., the total weight of bone marrow 20 g./kg. (Nye, 1931), the total weight of bone without marrow 60 g./kg., the total weight of skin 120 g./kg., and the total weight of skeletal muscle 520 g./kg. (Levine, Mann, Hodge, Ariel, and Du Pont, 1941). With one exception the lead concentration in bone was taken as the value obtained from diaphysis, as being the sample freest from marrow.

By taking two precipitates the error of the estimates was considerably reduced, since a small proportion of the lead sulphide of the first precipitate, lost either because it slipped under the paper or because the particles were too fine to be retained, was recovered in the second precipitate. Also the ratio between the Geiger-Müller counts for the first and second precipitates (generally about 10) gave an indication of the reliability of a particular estimate. The standard error of the lead estimations, calculated from duplicate determinations made during these experiments, was ± 3 per cent. The mean recovery of lead from the entire animal, calculated as described above, was 90 per cent with a standard deviation of ± 10 per cent of the dose.

Results

Data are presented for the distribution of lead in fourteen rabbits, all of which received a single dose of lead acetate containing 2.07 mg. of lead per kg. of body weight and eight of which were treated with BAL. Of the rabbits which did not receive BAL, two were killed at one hour after injection, one at six hours, and three at twenty-four hours. Of the treated rabbits, two were given 50 mg./kg. BAL (i.e. 40 molecules per atom of lead) immediately after the lead acetate and were killed at one hour. One rabbit was given 50 mg./kg. one hour after the lead acetate and 12.5 mg./kg. four hours later and was killed at six hours. Two were treated in the same way but were not killed until twenty-four hours. Three more were treated with BAL 50 mg./kg. nineteen hours after the lead acetate and 12.5 mg./kg. at twenty-three hours and were killed at twenty-four hours. The doses of BAL used were very near the toxic range, as maximal effects were being sought: one rabbit of the last group (No. 93) died nearly twenty-three hours after receiving lead acetate, shortly before the small injection of BAL was due. Ill effects were not observed in any of the other rabbits.

The concentrations of lead in microgrammes per gramme of tissue and the percentages of the dose found in various organs and calculated for various tissues are shown in Tables I and II. Considerable variation occurred between different rabbits which received the same treatment, especially at twenty-four hours, but from the general trend of results it appeared that with a few exceptions the distribution of lead when BAL was not given did not alter greatly between one and twenty-four hours. The exceptions were the plasma and lungs in which the lead content showed a steady decline, and in the contents of the alimentary canal, in which the amount of lead increased in the longer experiments. Changes in other organs did not exceed the variation

TABLE I

THE EFFECT OF TREATMENT WITH BAL ON THE CONCENTRATION OF LEAD IN THE TISSUES OF RABBITS ONE HOUR, SIX HOURS, AND TWENTY-FOUR HOURS AFTER THE INTRAVENOUS ADMINISTRATION OF LEAD ACETATE (2.07 MG. Pb/KG.)

Time after giving lead:	μg. lead per g. fresh weight of tissue													
	1 hour				6 hours		24 hours							
Treatment with BAL (intramuscular injection):	None		50 mg./kg. at once after lead		None	50 mg./kg. at 1 hr. 12.5 mg./kg. at 5 hr.	None			50 mg./kg. 1 hr. 12.5 mg./kg. 5 hr. after lead		50 mg./kg. at 19 hr. and 12.5 mg./kg. at 23 hr. after lead		
Rabbit No.:	74 ♂	76 ♀*	75 ♀	79 ♂	83 ♀	82 ♀	86 ♀	90 ♂	91 ♂	92 ♂	95 ♀	93† ♂	94‡ ♀	97 ♀
Weight, kg.:	1.20	1.70	1.15	1.22	1.10	1.05	1.40	1.00	0.98	1.15	1.50	1.25	1.25	1.30
Plasma	0.50	0.82	2.67	3.17	<0.005	0.18	<0.02	<0.04	0.05	<0.01	<0.31	0.40	<0.15	0.11
Erythrocytes	1.49	1.71	0.88	1.78	1.99	0.46	1.65	1.38	1.94	0.29	1.61		<0.30	0.40
Spleen	43.0	30.0	4.74	0.98	32.5	37.6	29.3	2.98	—	10.5	15.5	34.1	27.2	22.8
Bone marrow	16.6	17.1	8.39	7.90	26.4	13.7	25.4	9.65	7.18	7.86	31.7	13.4	6.47	12.1
Liver	46.6	38.8	20.4	18.3	22.6	27.3	23.0	52.8	46.2	35.7	12.2	18.5	33.2	24.4
Bile	0.20	0.18	8.55	0.20	3.00	8.17	9.10	0.80	7.30	0.83	10.9	29.5	1.14	7.67
Stomach	0.15	0.18	0.93	0.53	0.20	0.35	0.26	0.09	—	1.14	0.87	0.49	1.71	1.26
,, contents	—	—	—	—	0.02	0.04	0.87	<0.01	—	1.29	1.84	0.49	<0.06	0.30
Small intestine	0.41	0.28	3.81	10.8	0.05	1.56	0.46	0.40	0.45	0.24	0.53	4.24	1.42	1.51
,, ,, contents					0.66	2.17	2.12	0.56	—	3.12	2.86	7.25	4.35	4.32
Colon	0.22	—	0.41	0.56	0.19	0.37	0.20	0.29	—	0.29	1.55	0.39	0.36	0.59
,, contents	0.02	0.03	0.39	0.37	0.37	10.5	0.29	1.49	—	3.93		2.35	1.73	—
Kidneys	5.31	1.84	12.0	10.0	2.43	3.00	2.22	1.63	4.10	1.52	2.95	2.95	3.15	2.23
Lungs	5.30	3.75	0.99	1.10	3.67	3.11	0.75	1.66	1.16	3.99	4.97	3.60	42.7‡	1.85
Heart	0.16	0.23	0.60	0.79	0.17	0.39	0.17	0.31	0.26	0.48	—	0.75	0.49	—
Skeletal muscle	0.05	0.03	0.25	0.41	0.03	0.24	0.03	0.08	0.01	0.15	<0.09	0.21	0.32	0.09
Diaphyses	2.57	1.66	0.64	0.80	3.71	2.76	0.41	1.48	—	1.79	4.66	5.05	2.25	3.45
Epiphyses	3.77	3.91	2.40	2.61	8.22	4.24	—	—	8.72	—	5.43	—	5.05	—
Ribs	4.15	4.44	2.98	0.86	—	—	—	1.14	—	—	4.97	—	5.44	2.58
Vertebrae	4.72	—	—	—	—	—	—	—	5.96	—	6.83	—	—	—
Skull vault	3.60	3.51	1.04	1.86	—	—	—	—	10.4	—	—	—	—	—
Brain	0.07	0.07	0.07	0.16	0.17	0.07	0.07	—	0.08	—	0.29	0.07	—	0.08
Skin	0.10	0.23	0.20	0.23	0.29	0.17	0.10	0.25	0.32	<0.01	—	0.44	0.31	—

* Pregnant. † Died 22 hr. 40 min. after injection of lead acetate. ‡ The injected solution contained some particulate matter, presumably basic lead acetate.

TABLE II

THE EFFECT OF TREATMENT WITH BAL ON THE DISTRIBUTION OF LEAD IN RABBITS ONE HOUR, SIX HOURS, AND TWENTY-FOUR HOURS AFTER THE INTRAVENOUS ADMINISTRATION OF LEAD ACETATE (2.07 MG. Pb/KG.)

Time after giving lead:	Percentage of dose in entire organ or tissue													
	1 hour				6 hours		24 hours							
Treatment with BAL (intramuscular injection):	None		50 mg./kg. at once after lead		None	50 mg./kg. at 1 hr. 12.5 mg./kg. at 5 hr.	None			50 mg./kg. 1 hr. 12.5 mg./kg. 5 hr. after lead		50 mg./kg. 19 hr. 12.5 mg./kg. 23 hr. after lead		
Rabbit No.:	74 ♂	76 ♀*	75 ♀	79 ♂	83 ♀	82 ♀	86 ♀	90 ♂	91 ♂	92 ♂	95 ♀	93§ ♂	94‖ ♀	97 ♀
Weight, kg.:	1.20	1.70	1.15	1.22	1.10	1.05	1.40	1.00	0.98	1.15	1.50	1.25	1.25	1.30
Plasma	1.25	1.45	4.48	6.22	<0.01	0.43	<0.03	<0.09	0.10	<0.005	<0.66	1.51	<0.36	0.21
Erythrocytes	1.69	2.77	1.48	2.54	3.32	0.54	1.83	1.67	2.42	0.37	2.03		<0.36	0.59
Spleen	2.60	0.43	0.26	0.05	1.14	0.87	0.81	0.16	—	0.31	0.25	0.66	0.42	0.28
Bone marrow	16.1	16.8	8.12	7.63	25.5	13.2	24.6	9.32	7.08	7.64	30.6	13.0	6.25	11.6
Liver	67.7	56.8	40.1	33.0	50.3	60.8	38.1	71.1	51.9	43.5	20.3	31.9	48.8	45.2
Bile	0.006	0.007	0.11	0.006	0.029	0.23	0.22	0.035	0.29	0.017	0.49	1.71	0.058	0.095
Stomach	0.09	0.10	0.85	0.36	0.14	0.30	0.17	0.06	—	0.72	0.51	0.31	0.62	1.00
,, contents	—	—	—	—	0.04	0.10	1.28	<0.01	—	1.29	4.10	1.26	<0.16	0.73
Small intestine	0.32	0.37	6.40	20.3	0.07	2.06	0.56	0.52	0.66	0.27	0.59	3.72	1.84	2.83
,, ,, contents					0.29	0.95	0.81	0.11	—	0.67	0.62	3.12	1.11	1.58
Colon	0.11	—	0.26	0.30	0.10	0.16	0.07	0.10	—	0.07	0.50	0.11	0.14	0.24
,, contents	0.013	0.006	0.10	0.029	0.17	0.77	0.50	0.21	—	0.61		0.28	0.51	—
Kidneys	1.50	0.52	4.02	4.00	0.65	0.92	0.75	0.49	1.27	0.38	0.74	0.82	1.00	0.60
Lungs	1.13	0.62	0.25	0.44	0.42	0.63	0.38	0.52	0.44	0.41	0.31	0.63	11.7‖	0.32
Heart	0.018	0.036	0.100	0.135	0.023	0.049	0.018	0.032	0.026	0.042	—	0.074	0.048	—
Skeletal muscle	1.25	1.65	6.60	10.3	0.84	6.19	0.78	1.97	0.26	3.69	<2.34	5.38	7.91	2.21
Bone†	7.44	4.80	1.83	2.32	11.8	7.99	1.18	4.28	12.90‡	5.20	13.3	14.6	6.54	10.0
Brain	0.018	0.014	0.020	0.050	0.050	0.023	0.018	—	0.031	—	0.092	0.019	—	0.030
Skin	0.59	1.35	1.11	1.41	1.68	0.94	0.61	1.44	1.92	<0.01	—	2.60	1.80	—
Injected ear	0.24	7.43	0.12	0.14	0.60	0.18	3.27	1.12	0.31	0.42	7.36	0.87	1.60	6.97
Excreted, urine	0.49	none	none	0.81	0.15	5.92	0.69	0.22	1.16	6.99	15.4	1.39	2.52	4.69
,, faeces	none	none	none	none	0.02	0.75	0.55	0.08	0.75	1.39	2.3	0.80	none	1.01
Total recovery	103%	95%	76%	90%	98%	104%	77%	93%	82%	74%	102%	85%	94%	90%

* Pregnant.
† Based on concentration in diaphyses. Excluding marrow.
‡ Based on half concentration in epiphyses. Excluding marrow.
§ Died 22 hr. 40 min. after injection of lead acetate.
‖ The injected solution contained some particulate matter, presumably basic lead acetate.

between different rabbits killed at the same time: 50–70 per cent of the entire dose was found in the liver, where, as in the bone marrow and the spleen, the highest concentrations of lead (10–50 μg./g.) occurred. Concentrations of 1–5 μg./g. occurred in the red cells, lungs, kidneys, and bone. All the other tissues regularly sampled contained about 0.05 to 0.5 μg. of lead per gramme. A small amount of lead was excreted in the urine (Fig. 1), less than 1 per cent in twenty-four hours: and to judge from the movement of lead in the gut contents, another 1 per cent or so was likely to be excreted in the faeces on the second and third days.

When BAL was administered several differences in the distribution of lead were observed. The content and concentration of lead in the liver, spleen, and bone marrow were consistently reduced, except in the liver at six hours. Data at six hours have been obtained for only one normal and one BAL-treated rabbit; the closely similar concentrations found in the livers do not seem to be an outstanding exception to the thesis that BAL reduces the amount of lead in the liver. The concentration of lead in red cells was also consistently lower after BAL, but this effect was offset by an increase in the amount in the plasma, so that the effect on total blood lead was variable. On the other hand, the excretion of lead in the urine was increased five- or tenfold (Fig. 1 and Table III). The increase was still apparent between twelve and twenty-four hours even when BAL had not been given after the fifth hour. In view of the usually transient effects of BAL on the excretion of other metals (Wexler, Eagle, Tatum, Magnuson, and Watson, 1946; Eagle, Magnuson, and Fleischman, 1946; Eagle, Germuth, Magnuson, and Fleischman, 1947) this finding was somewhat unexpected. In the rabbit which died at twenty-three hours (No. 93)

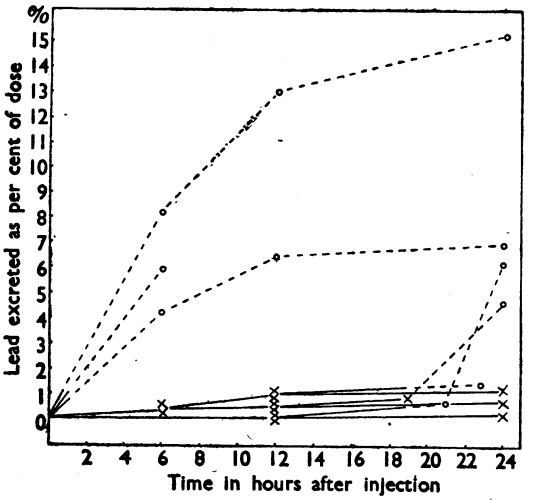

FIG. 1.—The effect of BAL on the cumulative excretion of lead in the urine of rabbits after the intravenous administration of lead acetate (2.07 mg. Pb/kg.).

Ordinates: amount of lead excreted as a percentage of the dose administered (1% = 20.7 μg. Pb/kg. body weight). Abscissae: time in hours after injection of lead. BAL given intramuscularly, either 50 mg./kg. at 1 hour and 12.5 mg./kg. at 5 hours, or 50 mg./kg. at 19 hours and 12.5 mg./kg. at 23 hours.

×——× No BAL or before BAL.
O- - - - - O After BAL.

TABLE III

THE URINARY EXCRETION OF LEAD AFTER INTRAVENOUS ADMINISTRATION OF LEAD ACETATE (2.07 MG. Pb/KG.) TO RABBITS, WITH AND WITHOUT SUBSEQUENT INTRAMUSCULAR INJECTIONS OF BAL

Rabbit No.	BAL 50 mg./kg. at (hrs. after lead)	BAL 12.5 mg./kg. at (hrs. after lead)	Urine volume ml. at hours after injection of lead 0–6	6–12	12–18	18–24	Urine lead concentration μg./ml. at hours after injection of lead 0–6	6–12	12–18	18–24
83	nil	nil	50	—	—	—	0.06	—	—	—
86	nil	nil	50	60	60		0.15	0.14	0.07	
90	nil	nil	0	50	50		—	0.04	0.05	
91	nil	nil	18	14	15		0.36	0.91	0.24	
93	19	—	24	18		50*	0.39	0.67		0.29*
94	19	23	25		30†	10‡	<0.03		0.70†	4.35‡
97	19	23	20	28	12	18	0.31	0.20	0.93	5.77
82	1	5	12	—	—	—	10.7	—	—	—
92	1	5	10	5	20		10.0	10.7	0.64	
95	1	5	20	24	36		12.8	6.4	1.90	

The heavy black lines in the body of the table separate values before injection of BAL from values after its injection.
* 18 hours–22 hours 40 minutes after injection of lead acetate.
† 12–21 hours after injection of lead acetate.
‡ 21–24 hours after injection of lead acetate.

the one dose of BAL failed to increase the excretion of lead in the urine. Excretion into the alimentary canal was also greatly increased, but the data are insufficient to show whether the increase was primarily due to biliary or to intestinal excretion. Precautions were not taken to prevent coprophagy, and it is possible that some or all of the lead in the stomach contents got there in that way. Increased concentrations of lead were regularly found in the skeletal and cardiac muscle and in the stomach wall. In the wall of the intestine increased amounts of lead were found in animals killed shortly after administration of BAL, but in the rabbits (Nos. 92 and 95) to which BAL was given at one and five hours and which were killed at twenty-four, any increase produced by BAL had disappeared. In other tissues changes were small or variable. For example, bone diaphysis and lungs showed a decrease in lead content at one hour and an increase at twenty-four hours. In rabbit No. 94 the injected solution was observed to be cloudy, and it appears likely that the exceptionally high concentration in the lungs was due to retention of some particulate matter, probably basic lead acetate, rather than to the action of BAL.

The various changes have been summarized in Table IV, in which tissues having common physiological functions and similar concentrations of lead, and in which the lead concentrations were similarly affected by BAL, have been grouped together. The oversimplifications involved are perhaps justified by the more immediate comprehensibility of the data so presented.

TABLE IV

Summary table of the distribution of lead after intravenous administration of lead acetate (2.07 mg. Pb/kg) to rabbits with and without subsequent intramuscular injections of BAL. The figures given are the mean percentages of the dose found in each group of tissues for all similarly treated rabbits. Details of treatment are as shown in Tables I, II, and III.

Distribution at:	1 hour		6 hours		24 hours		
Treatment:	No BAL	BAL	No BAL	BAL	No BAL	Early BAL	Late BAL
No. of Rabbits:	2	2	1	1		2	3
Liver, spleen, and bone marrow	80	45	76	75	68	52	54
Alimentary canal and contents, faeces, and bile	0.5	14	0.7	4.5	3	7	8
Urine	–	–	0.1	6	0.7	11	3
Blood	4	7	3	1	2	1	1
Skeletal muscle	1.5	8	1	6	1	3	5
Bone	6	2	12	8	7	9	10

Discussion

The extensive literature on the distribution of lead in animals contains few observations on the immediate fate of a single dose of a soluble inorganic salt administered intravenously. The most comprehensive data appear to be those of Weyrauch (1931) and of Kehoe and Thamann (1933) for rabbits and of Behrens (1925) for mice and cats. In general the present results are consistent with those already published (Table V) for the rabbit, at least in order of magnitude. However, certain discrepancies are larger than might be expected from differences in dosage and in experimental technique. Except for a single figure of Behrens's for cats (*loc. cit.*, p. 353), bone and marrow apparently have been taken together by previous authors, in unspecified proportion and usually from unspecified sites. In the experiments reported here, bone marrow has been found to contain up to eight times the amount or twenty times the concentration of lead in bone, particularly when relatively marrow-free diaphyses were sampled. It follows that estimates for bone and marrow taken together mean little, as small changes in the proportions of the two tissues in a sample must greatly influence the result. Differences in the lead content of different bones and different

TABLE V

Distribution of lead one hour after intravascular injection of lead salts (after various authors). The figures given have been deduced from the published data in accordance with assumptions used elsewhere in this paper. Kehoe and Thamann's data are taken from rabbits killed half or two hours after the injection of lead.

Author:	Per cent of dose in various tissues				
	Behrens (1925)			Kehoe and Thamann (1933)	Ginsburg and Weatherall (1948)
Species:	Mouse	Cat	Cat	Rabbit	Rabbit
Lead salt:	Chloride	Chloride	Chloride	Chloride	Acetate
Approx. dose: (mg. Pb/kg.)	15	4	0.04	7–17	2
Blood	20	35	25	2.5	3.5
Spleen	—	—	0.2	2.5	1.5
Liver	29	18	20	8.5	62
Intestine	11	3	6	5	0.5
Kidneys	9	1.5	4	1.5	1
Lungs	—	—	0.6	—	0.9
Heart	0.7	2*	0.4*	—	0.03
Muscle	—	—	5	6.5	1.5
Bone	10	20	20	6.5	6
Brain	—	—	—	1	0.02

*Lead given by intracardiac injection.

parts of the same bone in acute experiments are partly explicable on this basis. Behrens's one figure for the lead concentration in the bone marrow of a cat is considerably lower than that given for bone in the same animal. This finding is the opposite of all the present observations, and apart from the obvious possibility of a species difference no explanation is apparent. The present findings are also directly the opposite of those described in chronic poisoning in cats, and perhaps rabbits, by Aub, Fairhall, Minot, and Reznikoff (1925). In other tissues the present values for blood and for the alimentary canal are low when compared with the general trend of results. Weyrauch's data are given for dry weights of tissue: suitably amended they appear quite consistent with those here. Kehoe and Thamann's recoveries in their five experiments with lead chloride lasting less than twenty-four hours average 44.5 per cent, and while they have admittedly discarded their "remainder" it is difficult to account for up to (in one case) 88 per cent of the dose outside the liver, spleen, kidneys, washed intestinal tract, blood, muscle, bone, and central nervous system. It seems more probable that losses occurred during administration or estimation, and consequently that their values err towards the low side. Some figures of Brady (quoted by Aub *et al.*, 1925) for the lead content of the livers of anaesthetized rabbits with cannulated bile ducts after considerably larger doses of lead show, like the present data, about 50 per cent or more uptake of lead by that organ in the first few hours after intravenous administration. Behrens's figures indicate that in mice the uptake and excretion of lead by the kidney is much more rapid than in rabbits, and that excretion through the alimentary canal is also much greater: the concentration per unit dose remaining in the tissues is correspondingly less. Urinary excretion in the present experiments has been fairly steady, amounting to not more than 1 per cent of the total dose in twenty-four hours. The early disappearance of lead from the plasma agrees with the observations of Bambach, Kehoe, and Logan (1942), and with those of Mortensen and Kellogg (1944) in dogs and guinea-pigs.

It should be noted that the present work was done with young rabbits. Kasahara and Arimichi (1934) found higher concentrations of lead in the blood of rabbits weighing about 1.0 kg. than in rabbits of double this weight fed on the same relative amounts of lead. Their data can be interpreted as due to differences either in the rate of absorption or of deposition and excretion, but they indicate some difference in the metabolism of lead by young and old animals. It should also be noted that the amount of radioactive material used in the present experiments was large. The possibility exists that some injury was done to those tissues in which the material was most highly concentrated, and that the affinity of the tissues for lead was consequently altered, but it seems unlikely that such an effect would be produced in the short duration of these experiments.

There is at present very little indication how BAL produces the described changes in the distribution of lead. Some of the changes are probably due to the actions of BAL on the rabbit. In view of the cardiovascular effects of BAL (Chenoweth, 1946), it is not surprising to find an increased amount of lead in the splanchnic area after simultaneously administered BAL. But such actions are likely to be less important when lead has been fixed by the tissues, as appears largely to have happened within an hour. Changes observed after this time more probably depend on the relative dissociability of the compounds formed between lead and the tissues and between lead and BAL, on the behaviour of the undissociated lead-BAL compound or compounds, and on the amount of BAL present in a given tissue. The importance or otherwise of the last factor is illustrated by considering the data of Peters, Spray, Stocken, Collie, Grace, and Wheatley (1947), who showed that in a rat one hour after a dose of 100 mg./kg. of BAL containing S^{35}, 2.8 per cent of the dose was found in the liver and 0.17 per cent in the brain. If a similar distribution of BAL is assumed in rabbits poisoned with lead, the amount of BAL in the liver of the rabbits described here would not have greatly exceeded one molecule per atom of lead, even if all the S^{35} were present as unaltered BAL, whereas the amount of BAL in the brain would have been a two-hundredfold excess. In spite of this, BAL had a greater effect on the lead content of the liver than on that of the brain. Without data about the distribution of BAL in the conditions of the present experiments, detailed consideration on these lines is not profitable.

Weatherall (1948) has shown that, after a single dose of lead acetate by stomach tube in rabbits, BAL diminishes the subsequent anaemia, increases the coproporphyrin excretion, and does not greatly affect the mortality. The present observations are consistent with Weatherall's hypothesis that BAL acts by preventing the uptake of lead ions by red cells, and they indicate moreover that BAL can probably remove lead from cells which have already taken it up. On the other hand, no light is thrown on the increased coproporphyrin excretion, nor on the increased mortality produced by BAL in subacute lead poisoned rabbits (Braun, Lusky, and

Calvery, 1946), nor on the prevention of the renal effects of lead in rats (Chiodi and Sammartino, 1947). Further and more chronic experiments are necessary to provide evidence on these points.

Summary

1. The distribution of lead in the tissues of rabbits after intravenous injection of lead acetate (2.07 mg. Pb/kg.) has been studied by the use of the short-lived isotope Pb^{212} (thorium B).

2. The distribution has been similar in most respects at 1, 6, and 24 hours. High concentrations were found in the liver, spleen, and bone marrow: moderate concentrations in red cells, lungs, kidney, and bone: and small amounts in all other tissues: 50–70 per cent of the administered lead was found in the liver, 15–25 per cent in bone marrow, and 5–12 per cent in bone. Less than 1 per cent was excreted in the urine in the first 24 hours, and 1–2 per cent was found in the contents of the gut.

3. After intramuscular injection of BAL the urinary excretion of lead was increased to 3–15 per cent of the dose, even when BAL was not given until nineteen hours after the lead, and larger amounts of lead were found also in the contents of the gut.

4. Other changes when BAL was administered were a reduction of the concentration of lead in the liver, spleen, bone marrow, and red cells, and an increase of the concentration in cardiac and skeletal muscle. After BAL 20–48 per cent (in one case 60 per cent) of the lead was found in the liver, 7–13 per cent (in one case 30 per cent) in the bone marrow, 1–13 per cent in bone, and up to 10 per cent in skeletal muscle.

5. The significance of these changes is discussed.

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