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EXPERIMENTAL STUDIES ON THE INGESTION OF LEAD COMPOUNDS*

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THREE general modes of study have been employed by experimental and hygienic workers in the determination of the degree and significance of human exposure to lead compounds. First, the results of animal experimentation have been applied more or less directly, and, in some instances, much too literally, to man. Second, the extent of an existing lead exposure, expressed on the basis of the measured lead content of food materials or drinking water, or, in industry, in terms of the quantities of particulate lead compounds in the atmosphere of work-rooms, has been set against clinical observations on exposed persons. Third, the relative magnitude of human lead absorption, as revealed by the determination of the lead content of certain tissues and excretions of exposed persons, has been correlated with clinical data.

For many reasons the third method of approach appeared to us to offer the best means for the practical solution of a problem which presented itself a number of years ago. Accordingly the rates of urinary lead excretion of groups of workmen in a variety of lead trades, determined under conditions of exposure and after discontinuance of exposure, were correlated with the presence or absence of symptoms and signs of lead intoxication so as to furnish a practical basis of differentiation between safe and dangerous occupational conditions (1, 2). The expression "practical basis of differentiation," is used advisedly here, in that our initial definition of degree of hazard in terms of lead excretion (1) was tentative and frankly pragmatic, as are all other available definitions of lead hazard. The method of study,

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however, based on the functional responses of exposed persons, gave promise of revealing the broader picture of lead metabolism in the human organism and thereby providing wholly valid and permanent criteria for clinical and hygienic practice. In any final sense, safety in relation to lead exposure, whether occupational or otherwise, must depend on a comprehensive knowledge of the physiological behavior of lead, and the manifest need for precise quantitative standards based on such knowledge has prompted us to increase our efforts in both intensity and scope.

When it became evident that men in general were absorbing and excreting appreciable quantities of lead (3, 4, 5, 6, 7, 8), that this was not a recent or localized phenomenon, and that lead in some small quantity entered into the normal physiological processes of the human organism, it followed that some level of lead absorption must be regarded as wholly devoid of any injurious effect upon man. As the logical outgrowth of this concept we sought to obtain fairly comprehensive information as to the natural sources of the lead which occurs in food, as well as the incidental and accidental means of contamination of foods and beverages (3, 7, 9), and to determine the quantities of lead regularly met with in the American dietary (4, 5, 6, 7). The ultimate disposition of the lead present in food and drink was also studied in a series of experiments in which the dietary lead intake as well as the lead output in the feces of normal human subjects was followed over a period of months (4, 5, 7). The facts obtained from these studies have been amplified and confirmed by further studies, implemented by more precise means for the collection of samples for analysis, and by recently developed analytical methods of great sensitivity and accuracy (10, 11, 12, 13, 14). Certain of the data of these later investigations are shown below, in illustration of the magnitude and variability of the lead content of the present freely chosen diet of healthy North American adults. Our chief attention, however, will be given to the presentation of some of the more obvious results of experiments designed primarily to determine the relationships obtaining between lead intake and output at various levels of ingestion over long periods of time and ultimately to establish the threshold of intoxication for lead when ingested by man.

It may be advisable to recall the fact that lead is taken in by the industrial worker largely through inhalation of particulate lead compounds, and that severity of exposure is expressed commonly in terms either of the measured lead content of the air, or of the estimated lead intake per day by inhalation. Neither of these expressions has any meaning when applied to lead taken orally, and attempts to identify the toxic level of daily ingestion with that of daily inhalation are ill conceived. The studies described here are concerned chiefly and most immediately, therefore, with the safe limits of lead ingestion in food and beverages on the part of the general population. On the other hand, the ingestion of lead by industrial workers is not without hygienic significance, however slight may be its relative importance. From the viewpoint of a physiological ap-

proach to the problem of lead intoxication, moreover, the rate of lead absorption as compared with the rate of lead excretion is of primary importance regardless of the avenue of entrance of lead into the body.

The Lead Content of the Present American Diet

Table 1 gives examples, from recent observations, of the range of variability of the daily intake of lead in the food of three normal healthy young adults who provided duplicate samples of all food and beverages consumed. The daily alimentary lead output for the corresponding period is also shown for each subject, and may be seen to be of the same order of magnitude as the intake, in conformity with our previous observations on other subjects (4, 5, 6). (The one appreciable discrepancy arose from the wide variability of the results on the feces of Subject M. R. and from the occurrence of an unusual number of high values within a short period of time, as shown by the large probable error and the Standard Deviation in relation to the mean value.) The similarity of the results for food and feces implies that the lead in food is not effectively absorbed by the alimentary tract, and this is verified by the significant positive correlation between the individual daily food samples and the corresponding individual fecal samples, with respect to their lead content. In the case of Subject H. D., for example, the comparison of 121 daily food samples with 121 twenty-four-hour samples of the feces for the following day, yielded a correlation coefficient of 0.31 ± 0.06; when all the samples were combined to represent forty-eight-hour periods,

TABLE 1

Daily Occurrence of Lead in the Food and the Corresponding Feces of Three Normal American Adults

Lead in mg. per 24 hrs.	Frequencies of occurrence of quantities of lead indicated					
	In food*			In feces		
	M. R.	E. B.	H. D.	M. R.	E. B.	H. D.
0–0.099			4	7	3	8
0.10	12	11	59	1	11	37
0.20	7	13	42	6	12	42
0.30	8	13	10	5	13	25
0.40	1	12	3	2	6	4
0.50	1	4	1	4	6	1
0.60 and over	2	3	2	6	5	4
Totals	31	56	121	31	56	121
Mean	0.270	0.352	0.220	0.377	0.338	0.247
P.E.	±0.017	±0.016	±0.007	±0.037	±0.016	±0.007
S.D.	±0.137	±0.181	±0.110	±0.301	±0.172	±0.121

* Lead in food includes that in all beverages for subject E. B. but excludes that in water for subjects M. R. and H. D. The latter amounts to ±0.02 mg. daily.

thereby reducing the effect of variation in alimentary emptying time, their correlation coefficient was 0.63 ± 0.05.

TABLE 2

LEAD INTAKE AND OUTPUT IN A NORMAL HUMAN SUBJECT (H. D.)*

SUCCESSIVE WEEKLY PERIODS	LEAD IN FOOD AND DRINK† IN MG.	LEAD EXCRETED IN MG.		
		Total	In feces	In urine
April				
1st	2.03	2.87	2.67‡	0.20
2nd	1.52	1.78	1.68	0.10
3rd	2.08	1.84	1.67	0.17
4th	1.50	1.75	1.61	0.14
December				
35th	1.35	1.67	1.35	0.32
36th	1.24	1.50	1.28	0.22
37th	1.17	1.44	1.28	0.16
38th	1.78	2.62	2.37	0.25
April				
51st	1.65	2.43	2.16	0.27
52nd	1.38	1.65	1.46	0.19
53rd	2.24	2.24	2.07	0.17
54th	1.42	1.79	1.57	0.22
August				
68th	1.63	1.88	1.67	0.21
69th	1.77	2.07	1.74	0.33
70th	1.41	1.85	1.58	0.27
71st	1.59	1.76	1.43	0.33
16	25.76	31.14	27.59	3.55

* Lead taken in by inhalation and excreted in perspiration ignored.

† Duplicate water samples were analyzed daily during last 4 periods. To all other weekly totals were added 0.14 mg.

‡ An aberrant result of 4.00 mg. obtained on one day of this week and not represented in the duplicate food sample was reduced to the mean value of 0.22 before the summation.

Representative data on the relationship between gross daily intake of lead (exclusive of that inhaled) and gross daily output of lead (exclusive of that lost through perspiration, expectoration, insensible desquamation of the skin and loss of hair and nails), are given in tables 2 and 3. It is apparent that there is an approximate balance, and considering the quantities of lead involved, as well as the obvious smallness of the factors on both sides which have been disregarded through present technical necessity, the balance is so nearly exact as to be quite convincing. In this connection Calvery's (15) comments on our previous and similar data may not be ignored. This investigator failed to note our reference to the lead content of the drinking water of our experimental subjects and suggested erroneously that this factor had been ignored. He also by implication gave undue weight to the factor of lead inhalation and suggested that our

TABLE 3

LEAD INTAKE AND OUTPUT IN A NORMAL HUMAN SUBJECT (E. B.)*

SUCCESSIVE WEEKLY PERIODS	LEAD IN FOOD AND DRINK IN MG.	LEAD EXCRETED IN MG.		
		Total	In feces	In urine
January				
1st	1.24	2.11	1.85	0.26
2nd	3.36	2.33	2.04	0.29
3rd	2.12	1.58	1.32	0.26
4th	1.90	2.73	2.47	0.26
February				
5th	2.73	2.50	2.26	0.24
6th	2.53	2.35	2.03	0.32
7th	2.69	3.37	3.14	0.23
8th	2.94	3.60	3.41	0.19
	19.51	20.58	18.52	2.06

* Lead taken in by inhalation and excreted in perspiration ignored. The latter at minimum during this period.

failure to provide a constant environment in this respect tended to vitiate our conclusions. There are adequate reasons for disregarding the factor of normal lead inhalation in experiments of this type at the present time,—first, because this factor is not unpredictable in its minute general magnitude under the conditions of our observations,* and second, because it is not feasible to carry out prolonged observations on the normal metabolism of human beings under artificial experimental conditions which impose undue restraints on normal life and behavior.

The lead content of food and beverages is clearly the predominant source of normal human lead intake, and the foregoing data partially illustrate the opportunities presented to 3 experimental subjects for lead ingestion from this source. Table 4 extends the picture by giving the results of similar observations on 12 subjects whose diets were somewhat variable both in quantity and quality. The whole range of variability and the frequencies of occurrence of various quantities of lead in the daily diet are here portrayed. These data relate to the commonly available food in Cincinnati and its environs, but they represent the facts as to the daily lead intake of men generally in the United States of America, since they are in agreement with our observations on persons in all parts of the country. Table 5 gives the results of some recent observations made in 10 cities in the United States.

TABLE 4

DAILY OCCURRENCE OF LEAD IN THE FOOD AND FECES OF TWELVE NORMAL AMERICAN ADULTS WITHOUT CORRESPONDENCE AS TO TIME

LEAD IN MG. PER 24 HOURS	FREQUENCIES OF OCCURRENCE OF QUANTITIES OF LEAD INDICATED	
	In food	In feces
0–0.199	259	248
0.20	583	414
0.40	143	95
0.60	43	30
0.80	12	5
1.00	9	2
1.20	2	3
1.40	3	1
1.80	1	
2.00 and over	8*	5*
Totals.....	1063	803
Mean and probable error ...	0.315 ±0.004	0.288 ±0.004
Standard deviation.....	±0.199	±0.180

* Omitted from calculations.

The variation among individuals with respect to the mean lead content of their daily diet, as illustrated in table 1, has been found to be due chiefly to differences in the quality of their food, (i.e. to their choice of food, or to differences in the preparation of food in various households and restaurants), rather than to gross differences

* Our subjects spent their working day in a laboratory building from which lead-bearing materials have been excluded, so far as possible, and in which such lead compounds as are required are segregated and handled with rigid precautions against their dissemination. Atmospheric dusts are eliminated to a considerable degree by filtration of in-coming air. The subjects lived in suburban Cincinnati. The data of Bloomfield and Isbell (16) give some direct basis for evaluating the extent of general atmospheric lead content, and our unpublished results on the correlation between certain low concentrations of lead in the air of industrial plants and the rate of urinary lead excretion indicate that the inhalation of lead by normal persons under our experimental conditions is less than 0.1 mg. per day, of which only some unpredictable but doubtless small proportion is available for absorption, the remainder escaping with the expired air.

in the quantities of food consumed. Nevertheless it seems profitable to examine the facts as to the gross quantities of food likely to be consumed daily by adults, and to note the actual concentration of lead in such food as a whole. We have assembled our data on the food of 12 healthy subjects in table 6, in which attention is called to the theoretically permissible limit of lead concentration in terms of the present tolerance established by federal authority in the United States. We do not imply that the regular occurrence of 7.62 mg. of lead in the daily adult diet is believed by government authorities to be safe, nor that there is a twenty-fold factor of safety in the present situation as we have presented it. We merely call attention to the facts as they now exist and as they would be found to exist if all foods were as high in lead as certain items are now permitted to be. Obviously the avoidance of any general increase in the lead content of a large number of food materials with a present minimal lead concentration is desirable.

Our observations over the past 15 years have given us no reason for believing that any appreciable or progressive change in the lead content of American food, or in the magnitude of the lead intake of the average American citizen has occurred during that period. It seems probable, however, that human ingestion of lead has decreased rather than increased in recent generations. Such evidence as we have, although inadequate to prove the point, also indicates that the general level of lead content of the Amer-

TABLE 5

LEAD CONTENT OF RANDOM SAMPLES OF FECES OF NORMAL PERSONS FROM TEN WIDELY SCATTERED AMERICAN CITIES

LEAD IN MG. PER SAMPLE OF FECES	FREQUENCIES OF OCCURRENCE OF QUANTITIES OF LEAD INDICATED
0-0.199	26
0.20	43
0.40	17
0.60	7
0.80	2
1.00	4
1.20	2
2.00 and over	1
Total	102
Mean and probable error	0.398 ±0.021
Standard deviation	±0.310

TABLE 6

WEIGHT OF FOOD CONSUMED DAILY BY NORMAL HEALTHY AMERICAN MEN ENGAGED IN PHYSICAL LABOR

(Including all beverages except water and large volumes of beer and wine)

(Combined data on 12 men varying from 120 to 240 pounds weight)

WEIGHT OF FOOD IN GM.	FREQUENCIES OF OCCURRENCE OF WEIGHTS OF FOOD INDICATED
800–999	11
1200	92
1600	271
2000	469
2400	232
2800	35
3200	4
3600	1
Total	1115
Mean and probable error	2135.16 ±7.87
Standard deviation	±389.53

Permissible mean lead content = 7.62 mg. = 3.57 p.p.m.

Actual mean lead content = 0.32 mg. = 0.15 p.p.m.

ican dietary is somewhat less than that current in France or England. There is thus little occasion for anxiety over the lead content of American food materials as they are now handled, processed, and distributed. From time to time, however, there are examples of foods contaminated with lead to an unusual and even dangerous degree, and greater care can be exercised in avoiding the introduction of lead into food at little or no cost or inconvenience except that associated with the recognition of numerous unnecessary contacts of foodstuffs with lead-bearing materials. For these reasons there is need of responsible and effective public and private agencies for the detection of such occurrences and for the correction of the conditions which bring them about.

Lead Ingestion and Lead Excretion at Abnormal Levels

The results of studies carried out over a period of years in industrial plants, the details of which have been reported in part elsewhere (1, 2), have indicated that an approximate balance exists between the lead absorption and the lead excretion of men who are subject to occupational lead exposure within certain limits. Such a balanced metabolism is apparently differentiated from that of the normal individual only in that it is maintained at a higher level after a period of absorption of lead into the tissues to a point determined largely by the magnitude of the regular lead intake. Expressed somewhat differently, these observations have shown that if the lead exposure of an individual be increased within certain limits above the normal or commonly observed levels, the increasing quantity of lead absorbed into the tissues results in a progressively increasing rate of elimination until eventually excretion approximates absorption. The determination of the limits within which such a balance can be achieved without injury or illness would, obviously, be of the utmost practical usefulness not only in the field of industrial hygiene but also in relation to the health and safety of the public. We, therefore, entered upon a series of experiments in which lead in known quantities was administered regularly to normal subjects over extended periods of time during which their daily lead intake in food and beverages and their daily output in the feces and urine were followed. (The intake and output of calcium and phosphorus have been determined simultaneously over the greater portion of the period of study.) In order to maintain the induced lead exposure of our subjects within safe limits, the dosage of lead was kept below that previously found to be toxic for experimental animals, and also below that necessary to produce concentrations of lead in the blood and urine which, from industrial experience, could be regarded as dangerous.

After a preliminary period of 28 days of clinical and experimental study which demonstrated the existence of a satisfactory state of health and physiologic reactivity, one young man was started on 1.00 mg. of lead acetate per day administered in solution in a dose of 0.333 mg. with each of three meals. This experiment has been continued over a period of almost 3½ years up to the present (May 1940). A second subject began taking a daily

dosage of 2.00 mg. in a similar manner at the end of February 1939, after 56 days of preliminary clinical and metabolic study, and has continued since on that dosage. At weekly intervals, each subject was given a physical examination, a complete microscopic examination of his blood, and an analysis of his blood for its lead content. Blood smears have been made daily for counts of stippled erythrocytes and reticulocytes. Aside from certain inconveniences associated with residence in or near the laboratory, and the necessity of collecting duplicate samples of all food and beverages as well as all excreta, these subjects have led normal and adequately active lives, and despite a somewhat detailed supervision of their daily activities, they have maintained a satisfactory attitude of mind, going about their daily duties with regularity and without apprehension or undue introspection.

During the course of the observations up to the present time, no clinical evidence of the slightest harmful effect has been recognized by either subject or has been detected by our examinations. No loss of appetite or weight, and no decrease in general health or in the sense of well-being has occurred. Both subjects have had intercurrent respiratory infections of minor severity which ran their normal course. One (M. R.) had a typical attack of food poisoning with diarrhea and vomiting, with recovery in 3 days. He was not too ill to collect all the materials evacuated by diarrhea and emesis, so that experimental observations were not interrupted. No appreciable changes in the lead metabolism occurred at this time. The same subject developed a faint but definitely punctate blue deposit at the margin of a central incisor in a localized area overlying a small pocket of purulent material. This was seen first on February 1st, 1938 and was allowed to remain untreated until October 24, 1939. During this interval the deposit varied somewhat in intensity, almost but not quite disappearing on two occasions when the gum seemed on the way to spontaneous recovery. When the lesion was evacuated and treated the deposit disappeared promptly and entirely, remaining absent during the healing process, which was complete, and failing to return. The other subject (E. B.) has not developed any gingival

TABLE 7

OCCURRENCE OF STIPPLED ERYTHROCYTES IN THE BLOOD OF NORMAL HUMAN SUBJECTS AT VARYING LEVELS OF LEAD INGESTION

STIPPLED ERYTHROCYTES PER 50 FIELDS	FREQUENCIES OF OCCURRENCE OF STIPPLED ERYTHROCYTES IN NUMBERS INDICATED			
	Normal subjects (control)		Normal subjects (test)	
	H. D.	C. H.	M. R.	E. B.
0–1.9	363	97	690	183
2	42	25	162	93
4	7	15	57	51
6	2	7	24	24
8	1	4	15	15
10		6	7	9
12		3	3	3
14		3	3	3
16		1		1
18–19.9			1	1
Totals	415	161	962	383
Mean	1.01	3.10	2.02	3.23
Probable error	±0.04	±0.19	±0.05	±0.11
Standard deviation	±1.10	±3.54	±2.14	±3.07

discoloration despite the presence of a slight gingivitis which has varied in degree in accord with his persistence in the care of his teeth.

No microscopic blood changes indicative of the effects of lead absorption have been observed in either subject. Table 7 groups the data of the daily observations on the stippling of the erythrocytes of the two subjects, in comparison with results obtained over comparable periods of time on 2 normal healthy young men who were not ingesting or absorbing abnormal quantities of lead during the period of the observations and who had no history of abnormal exposure to lead in any prior period. No evidence of any effect of the lead absorption is seen in comparisons of the mean values or the distributions of the frequencies under the different rubrics. It is of considerable importance to note that the variability in all the subjects as indicated by the Standard Deviations, is of the same degree. The absorption of lead evidently failed to induce either a constant or an intermittent stimulatory effect upon hematopoesis. Certain other factors, however, had a well defined influence upon the number of stippled erythrocytes. Attention is called to the wide fluctuations which occur on a seasonal basis, roughly, in the two subjects M. R. and E. B., as shown in figure 1. These fluctuations occurred at approximately corresponding times and to a similar extent in the control subjects C. H. and H. D., taking place in the one (H. D.) at a low level of this hematopoetic response, and in the other (C. H.) at a relatively higher level, as indicated by the averages for a year.

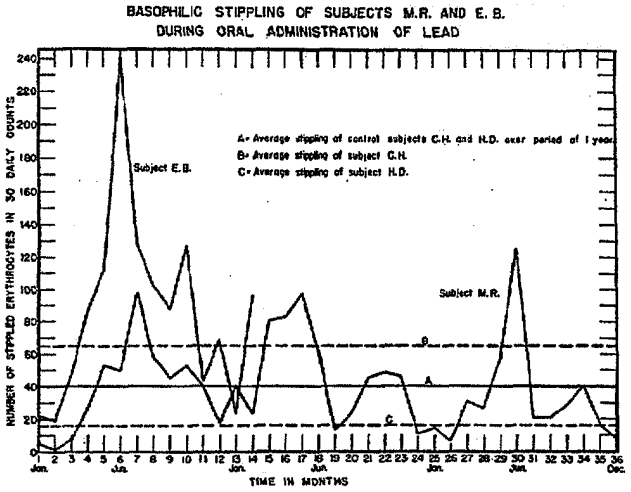

FIG. 1. Seasonal variation in occurrence of stippled erythrocytes in blood of human adults. The points on the curve represent the summation of the numbers of stippled erythrocytes found during the indicated periods in the examination of 50 microscopic fields (approx. 12500 erythrocytes) on a number of days, divided by the number of days on which observations were made, and multiplied by 30 so as to express them in terms of months.

(There is some question as to the most valid and illustrative means of expressing the results of blood examinations of this type. It is not clear whether the frequency of occurrence of stippled erythrocytes in the blood is a measure of intensity of this hematopoetic reaction or whether the actual number of such forms found is the better criterion. It seems probable that both the frequency and the numerical extent of the occurrence are important. We have chosen to express our results on a monthly basis, by adding the total number of stippled erythrocytes found on the several days of the month on which observations were made, dividing by the number of days represented, and multiplying by 30. This mode of expression, when combined with the data on the frequencies in table 7 would seem to portray all the facts faithfully.) The results on Subject M. R. by months vary above and below the average yearly values obtained in control subjects C. H. and H. D., remaining within the normal limits as defined by these subjects. The average of the results on Subject E. B. for the year is considerably higher than that of Subject M. R. and somewhat above that for control subject C. H., but the difference cannot be attributed with any degree of confidence to the factor of lead absorption in view of the comparison of the distribution of the frequencies and the mean values as shown in table 7.

TABLE 8

LEAD INTAKE AND OUTPUT OF NORMAL SUBJECT (M. R.) DURING ORAL LEAD ADMINISTRATION

SUCCESSIVE PERIODS OF 12 WEEKS	LEAD IN MG. INGESTED IN FOOD AND BEVERAGES AND EXPERIMENTALLY ADMINISTERED	LEAD IN MG. EXCRETED			LEAD IN MG. RETAINED
		Total	In feces	In urine	
1st	112.84	101.48	98.32	3.16	11.36
2nd	118.74	108.95	103.74	5.21	9.79
3rd	107.22	105.51	100.15	5.36	1.71
4th	118.79	111.57	105.57	6.00	7.22
5th	110.18	111.22	104.66	6.56	−1.04
6th	116.22	121.59	114.15	7.44	−5.37
7th	109.44	93.41	86.57	6.84	16.03
8th	125.45	118.86	112.47	6.39	6.59
9th	117.11	114.81	108.97	5.84	2.30
10th	105.48	98.86	91.22	7.64	6.62
11th	107.57	109.19	102.25	6.94	−1.62
12th	114.24	102.06	94.03	8.03	12.18
13th	113.14	100.68	94.02	6.66	12.46
	1476.42*	1398.19	1316.12	82.07	78.23

* Approximately 22.00 mg. in drinking water, 372.32 mg. in food and other beverages, and 1082.10 administered in solution.

Reticulocytes in the peripheral blood of both subjects varied within narrow limits without any definite sea-

sonal or other trends except some slight tendency toward increase in September, October and November. No parallelism between stippled erythrocytes and reticulocytes was apparent.

The facts of greatest and most immediate significance as derived from the study of analytical results on the two subjects are illustrated in a series of tables and charts. Table 8 gives a greatly condensed summary of the results obtained on Subject M. R. over 39 lunar months, while table 9 shows the corresponding data on Subject E. B. in a more extended form made possible by the much shorter duration of the observations on this subject. Attention is called in these tables to the large and prompt loss of the greater portion of the ingested lead in the fecal evacuations. In both cases approximately 95% of the ingested lead was eliminated during the period of the observations, and about 94% of the eliminated lead was found in the feces. The output of lead in the feces increased greatly and abruptly above the preliminary level for these subjects (cf. tables 1 and 3), as soon as the administration of lead was initiated. (The fecal lead was greatly elevated on the first day after the beginning of the administration, and reached almost its maximal level within a few days, these facts pointing clearly to the passage of most of the ingested lead through the alimentary tract without absorption.) The urinary output, on the contrary, increased slowly and gradually in response to the administration of 1.00 mg. daily (Subject M. R.), and some-

TABLE 9

LEAD INTAKE AND OUTPUT OF NORMAL SUBJECT (E. B.) DURING ORAL LEAD ADMINISTRATION

SUCCESSIVE PERIODS OF 4 WEEKS	LEAD IN MG. INGESTED IN FOOD AND BEVERAGES AND EXPERIMENTALLY ADMINISTERED	LEAD IN MG. EXCRETED			LEAD IN MG. RETAINED
		Total	In feces	In urine	
1st	60.94	55.55	53.20	2.35	5.39
2nd	59.76	54.85	52.27	2.58	4.91
3rd	65.34	57.16	54.32	2.84	8.18
4th	63.01	62.87	60.14	2.73	0.14
5th	63.51	61.79	58.71	3.08	1.72
6th	64.77	63.52	60.45	3.07	1.25
7th	64.46	64.93	61.67	3.26	−0.47
8th	63.81	60.34	56.78	3.56	3.47
9th	63.34	55.40	51.96	3.44	7.94
10th	62.87	56.11	52.43	3.68	6.76
11th	62.09	55.65	52.36	3.29	6.44
12th	58.89	56.62	53.31	3.31	2.27
13th	57.30	53.87	51.14	2.73	3.43
	810.09*	758.66	718.74	39.92	51.43

* Approximately 7.00 mg. in drinking water, 75.76 mg. in food and beverages, and 727.33 administered in solution.

what more rapidly in the case of Subject E. B. on the higher dosage. Thus, the urinary lead output of Subject M. R. practically doubled in about 12 weeks, but continued to increase for three times that length of time, while that of E. B. had doubled in less than 2 weeks, arriving at a substantially maximal level in about 10 weeks. Apparently, an important point of difference in the reaction to these two dosages consisted in the speed with which the rate of urinary lead excretion increased to a practically maximal level. Figure 2 provides a graphic illustration of the foregoing differences in terms of the mean values of the urinary lead concentration of the two subjects as calculated for each period of 28 days. The initial slope of the curve for E. B. is both steeper and shorter than that for M. R.

In figure 2 are also plotted the blood concentrations as averaged for periods of 4 weeks from the results of the weekly analyses. One notes here what we have found regularly in studying the results of occupational lead exposure, that the lead concentration in the blood shows less proportional change for a given increase in lead absorption than does that in the urine.

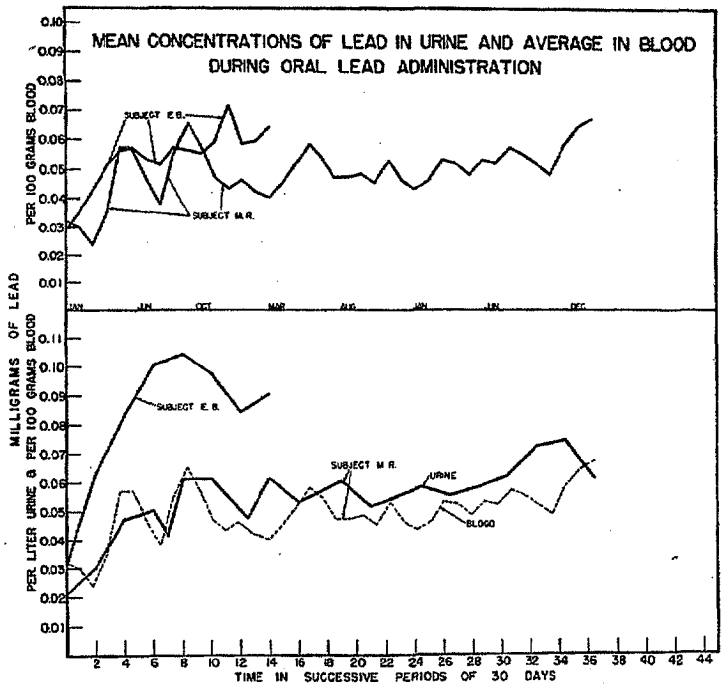

FIG. 2. The mean urinary lead concentrations were calculated from 60 daily analytical results, and the average lead concentrations in the blood were calculated from 4 weekly pairs of duplicate analyses. The dotted line in the lower section is the longer line in the upper section, superimposed upon the curve of urinary concentrations, for purposes of comparison.

Two significant features of the ef-

fects of prolonged lead absorption on the lead concentration of the urine and blood are revealed by the consideration of the facts in tables 8 and 9, and figure 2. First, it is apparent that the levels of lead concentration in the urine and blood in Subject E. B. are significantly higher than the corresponding levels in Subject M. R. in accordance with the higher dosage of administered lead. After making every allowance for individual variation in the metabolic responses of the two subjects, the conclusion is inescapable that the *rate* of daily lead absorption, regardless of the ultimate disposition of the lead in the body, is a significant factor in determining the concentration of lead in the urine and the blood.

The second point of importance relates to the length of time required for the development of changes in the lead concentration of the urine and blood comparable with the increases in the intake of lead. The lag here may be explained in part, no doubt, by the smallness of the quantities of lead absorbed from the alimentary tract, so that time was required to produce an appreciable effect. On the other hand, considering the fact that the ingestion and presumably the general rate of alimentary lead absorption over any considerable period of time were substantially constant for each subject during the entire period of the observations, the explanation of the gradual increase in these concentrations would appear to lie in a gradually increasing quantity of lead in the tissues of the subject. Obviously, as indicated above, the quantity of lead distributed in the tissues was not the only factor responsible for the elevation of the lead concentration in the urine and blood of the subjects. If it had been, the position of the subjects with respect to these concentrations should have been reversed at the point of termination of the urinary and blood curves of figure 2, since at this time, M. R. had retained more lead in his tissues as the result of the experimental regime than had E. B. (cf. tables 8 and 9). That retained lead was one factor, however, seems reasonably assured, and because of this assurance, coupled with the facts demonstrated by the study of the elimination of lead by persons whose lead exposure had been discontinued, the prediction is justified that when the administration of lead to these subjects is brought to an end, the lead concentration in their urine and blood will decrease rapidly to levels determined by the lead distributed in the tissues of the entire body, after which they will continue at lower and slowly decreasing levels until equilibrium between intake and output has been restored. This prediction, in due time, will be put to experimental test.

For present purposes, we shall omit a detailed examination and discussion of a number of factors which have influenced the absorption and excretion of lead on the part of these subjects. The practical significance of one matter, however, necessitates brief consideration. The daily variations in the concentration and output of lead in the urine were considerably greater than might be expected from the condensed results in tables 8 and 9 and from the curves in figure 2. The most cursory examination of data of daily observations revealed the importance of a factor which we have demonstrated elsewhere (4, 6), i.e. that the volume of water available for excre-

tion by the kidneys had a highly significant relation not only to the concentration of lead in the urine but also to the quantity of lead eliminated daily. The effects of other factors were not so obvious. It seemed advisable, therefore, to study the blood and urine at frequent intervals during the day to determine, if possible, the relationships obtaining between lead concentrations in the blood and urine, and the changes which occurred in both in response to the three doses of stage.) The urinary lead concentration, on the contrary, varied over a wide range during each 24 hour period, and the plotted curves of these concentrations are almost exactly opposite, throughout their course, to the corresponding curves of urinary volume. It is apparent, especially in the first group of observations, that no factor other than the volume of available water exerted any important role in the induction of variations in urinary lead concentration during these 24

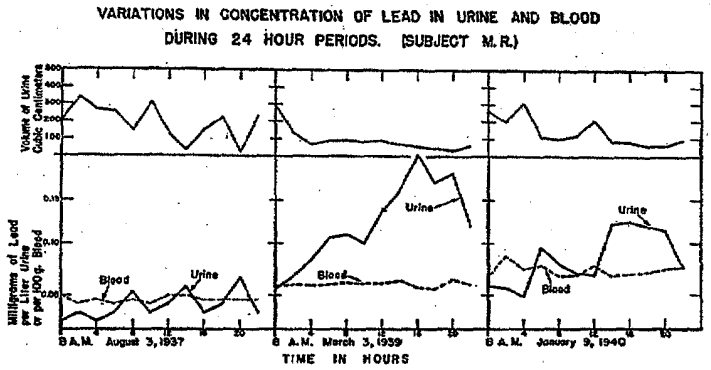

FIG. 3. The analytical results appearing as points on the lower section were obtained on the urine voided at 2 hour intervals, and on blood samples taken in duplicate at 2 hour intervals.

lead with the meals. Figure 3 shows the results of three sets of observations of this type on Subject M. R. at various stages of the experiment. The blood showed little variation in its lead concentration throughout any one 24 hour period, no change beyond the limits of analytical variation being demonstrable. Lead concentration of the blood increased progressively, however, from stage to stage of the experiment. (There was no appreciable change in the partition of lead between plasma and formed elements at any hour periods. The absorption of lead from the alimentary tract was too slight in its periodicity to cause demonstrable variations in the concentration of lead in either blood or urine.

Figures 4 and 5 give graphic representations of the periodic and total quantities of the lead taken orally and excreted in the feces and urine of the two subjects over the entire period of the observations reported herein. The abruptness of the gross increase in the alimentary output of lead, the gradual increase in the urinary output,

and the minuteness of the discrepancies between intake and output, are brought into their proper perspective. There was a surprisingly small retention of lead in the tissues of these subjects from the beginning of the observation. Indeed, in the case of Subject M. R., the intake and output as measured, with the exception of three periods, one at the beginning, another about midway, and the other near the end of the record, were so nearly equivalent as to be only doubtfully beyond the limits of the combined opportunities for error in the collection and analysis of the samples. Moreover, the excretory loss of lead from the body by way of the perspiration does not enter into the recorded total output. Recent observations, which are in process of extension and verification, have indicated that the concentration of lead in the perspiration of a normal individual corresponds closely to that in the urine. On this basis, our subjects probably excreted as much lead in this way during warm weather as in the urine, and the inclusion of such quantities on the elimination side would result in a virtually balanced metabolic picture. It seems probable, therefore, that the actual retention of lead on the part of these subjects was considerably less than that indicated by our figures, and that, in fact, it was of almost insignificant proportions.

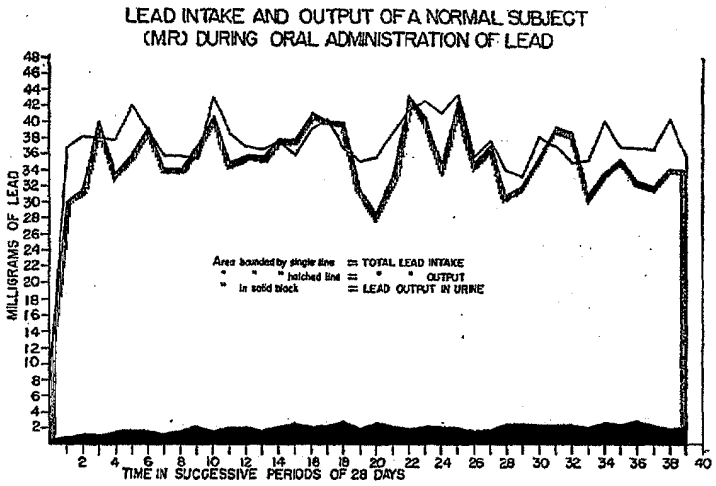

FIG. 4. Lead intake in food and drink and lead output in feces and urine of a human adult on a dosage of 1 mg. Pb daily as a solution of lead acetate, in addition to that occurring in the diet.

Despite the practical conclusion drawn above, critical interpretation of certain facts necessitates the opinion that some degree of lead retention occurred in both subjects. First, there is the fact, in accord with expectations, that the discrepancy between lead intake and output of Subject E. B. as measured, was greater for comparable periods of time than that in the case of M. R. Second, the concentra-

tion of lead in the blood and also the urine of M. R. showed a slight but definitely progressive increase at various stages in the observations (fig. 3), with some tendency, perhaps temporary, toward increase in the most recent period (fig. 2). Third, an examination of the curves in figure 6, in which the cumulative totals of the differences between intake and output of lead as measured for the two subjects were plotted against the successive periods of time, reveals a remarkable fact. The cumulative retention of lead was slight in both cases, and also apparently irregular, but if the attempt is made to fit a curve to the plotted points, the result is a straight line. It would be presumptuous at this time to draw a definite conclusion from this fact. The magnitude of the lead excretion in the perspiration must be accounted for, and certainly further study in the case of Subject E. B. is required. Doubtless other factors will have to be fitted into their proper place for the elucidation of the complete picture. Until such time, the evidence for or against the existence of a completely balanced lead metabolism under these conditions is not wholly convincing.

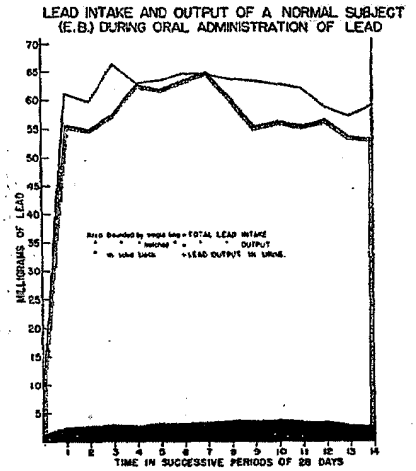

FIG. 5. Lead intake in food and drink, and lead output in feces and urine of a human adult on a dosage of 2 mg. Pb daily as a solution of lead acetate, in addition to that occurring in the diet.

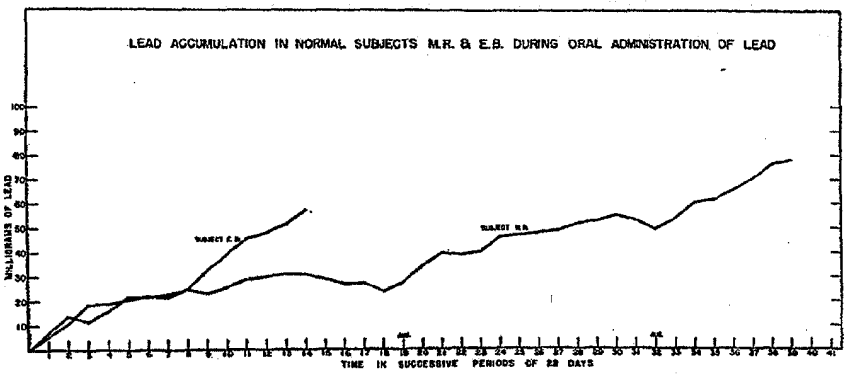

FIG. 6. Points on the curves represent the cumulative total of the differences between the lead in duplicate samples of food and drink, plus that administered, and the lead output in the feces and urine.

The emphasis placed by the foregoing remarks on lead retention under the conditions of our experiment seems

likely to give undue prominence to this point. However great may be the physiologic significance of the concept of a balanced lead metabolism under conditions of normal or moderately increased lead absorption, certain practical aspects of these experimental observations merit the greater present consideration. It is apparent, for all practical purposes, that the healthy human organism responds to such limited increases in lead ingestion by a greatly increased and substantially equivalent elimination. It is also clear that the largest proportion of ingested lead, even when taken in the form of a solution most favorable for absorption, is not absorbed in the alimentary tract, but is carried through the body and eliminated in the feces.

Discussion

The facts briefly outlined in the foregoing pages have a direct and important bearing upon the hygienic problem of lead as a contaminant of food materials. They do not justify the conclusion that the lead content of the food of the community may, with safety, be permitted to increase four-fold or more, but they do signify, we believe, that there is a reasonable public security against the dangers of insidious intoxication and gradual physical degeneration, which many have feared from the use of foods, especially fruits, generally available in this country. Such present security should not be permitted to lull us into an attitude of indifference toward potential hazards in food production and processing, but should promote a somewhat greater intellectual poise in the investigation of the facts with respect to such hazards. It should also alleviate a somewhat hypochondriacal tendency on the part of a segment of our population, and allow attention to be focused on the more serious problem of community health in relation to lead compounds, namely, the elimination of dangerous and disabling occupational lead exposure.

Perhaps an additional step toward the solution of the latter problem has been made in our attempt, as yet incomplete, to define the limits of safety for one type of lead exposure in physiological terms. The extension of similar studies to controlled conditions of respiratory lead exposure may go far toward the ultimate goal.

Summary and Conclusions

The mean values for the daily intake of lead in the food and drink of three normal American adults, as determined by recent and precise analytical methods, were 0.24, 0.29 and 0.35 mg., as compared with values of 0.25, 0.34, and 0.38 mg., respectively, for their daily fecal lead output during the same period.

The total quantities of lead ingested with their freely chosen food and beverages, by two American adults during periods of 16 weeks and 8 weeks, respectively, were 25.76 mg. and 19.51 mg.; the corresponding quantities of the combined output of lead in the feces and urine of the two were 31.14 mg. and 20.58 mg., respectively.

A highly significant degree of correlation existed between the lead content of the food for each daily period and that of the feces for the day following.

In observations which extended

over a period of a year, 12 normal American adults, engaged in occupations involving a moderate degree of physical effort, consumed a mean daily weight of 2135 gm. of food and beverages per person (excluding occasional large volumes of beer and wine), the mean lead content of which was 0.32 mg., equivalent to 0.15 p.p.m.

Experimental studies were carried out on 2 healthy adults to whom 1 mg. and 2 mg. of lead, respectively, as a solution of lead acetate, were administered daily in three doses taken with the meals, for periods of 39 lunar months, in the former case, and 13 lunar months in the latter. Clinical data were collected at daily and weekly intervals, while the daily oral intake and the daily alimentary and urinary output of lead were determined over the entire period of the study. The results may be summarized as follows: (1) no definite or suggestive evidence of lead intoxication developed in either subject; (2) no microscopic evidence of any hematopoetic response to lead absorption was found in daily examinations of the blood; (3) fecal output of lead in each case increased abruptly with the initiation of the experiment, promptly reached a maximal level, and accounted for the evacuation of about 90% of the ingested lead from the body; (4) urinary lead concentration and output increased gradually in each case, (more rapidly and to a higher level in the subject on the higher dosage), reached a substantially constant level in somewhat less than a year in the subject on the lower dosage, and in about 10 weeks in the other subject, and resulted in both cases in the elimination of about 5% of the ingested lead; (5) lead concentration in the blood of the two subjects increased in general correspondence with the increase in urinary concentration, but to a somewhat lesser degree; (6) in the case of the subject to whom 1 mg. of lead was administered daily, the mean level of lead concentration in the urine during the last 4 months of the observations was 0.068 mg. per liter, and the average concentration in the blood for this period was 0.059 mg. per 100 gm.; (7) the subject on 2 mg. daily, had corresponding values of 0.087 mg. per liter of urine and 0.063 mg. per 100 gm. of blood during the same period; (8) lead concentration in the blood of the subjects was subject to insignificant variation during the course of a day, and to only minor variations from day to day, while the urinary lead concentration, as well as output per unit of time, varied widely in accordance with the volume of water excreted by the kidneys; (9) the total amount of lead retained in the tissues of both subjects over the period of study was quite small, and with due regard for an additional and undertermined loss by way of the perspiration, as well as the effects of certain factors which could not be controlled entirely, it was almost negligible; (10) the discrepancy between total oral lead intake and output in the feces and urine of the subject on 1 mg. daily of administered lead for a period of 39 lunar months was 78.23 mg. of lead, while the corresponding discrepancy in the case of subject on the 2 mg. dosage for 13 lunar months was 51.43 mg., each of these quantities representing about 5% of the lead ingested during the respective periods; (11)

the combined evidence indicates that some actual retention of lead occurred in both subjects, and that the rate of retention was substantially constant for each subject over the period of the observations, being greater in the subject to whom the larger dosage of lead was administered.

The following conclusions are justified by these results:

1. The largest proportion of the lead which is ingested by normal persons, with food or otherwise, whether in soluble or insoluble form, is eliminated in the feces without absorption;

2. Daily ingestion of lead in quantities somewhat in excess of 1 mg. by healthy adults, over long periods of time ($3\frac{1}{2}$ years) is not only compatible with the maintenance of normal health and well-being, but is also associated with such an increase in the elimination of lead as to result only in a slight and almost negligible retention of lead in the body;

3. Daily ingestion of slightly more than 2 mg. of lead, for a period of more than 1 year, failed to result in any demonstrable effect upon the health or well-being of a healthy adult, but induced a level of lead elimination higher than that caused by the ingestion of 1 mg. daily, and a slightly greater rate of lead retention in the tissues;

4. Maintained concentrations of 0.06 to 0.07 mg. of lead per 100 gm. of blood over periods of months are entirely compatible with normal health and well-being of human adults;

5. Concentrations of lead in the urine which vary from 0.05 to 0.15 mg. per liter and slightly more, with mean concentrations as high as 0.09 mg. per liter over long periods of time, correspond with lead concentrations in the blood of 0.06 to 0.07 mg. per 100 gm., and likewise, are compatible with normal health and well being of human adults;

6. The quantities of lead ingested in food and beverages by adult citizens in various parts of the United States vary from somewhat less than 0.10 mg. to somewhat more than 2.00 mg. with a mean value of approximately 0.32 mg., per day;

7. From the results of the foregoing studies, it would appear that there is a sufficient factor of safety in relation to the lead content of the general food materials in the United States.

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