Report of Progress

(as of July 31, 1966)

IN AN INVESTIGATION OF "HUMAN EXPOSURE TO PARTICULATE LEAD COMPOUNDS IN THE AIR UNDER EXPERIMENTAL CONDITIONS"

Project No. LH-75 - of International Lead Zinc Research Organization

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(entitled) "The Fate of Inhaled Particulate Lead Compounds"

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INTRODUCTION

In the early presentation of proposals for the initiation of this work, a brief description was given of a systematic, experimental program which had been in progress for some twenty-five years, with the objective of elucidating the metabolism of lead in man, within the limits of intake (dosage) that are believed surely compatible with the avoidance of any known or demonstrable toxic response. No full account of the results obtained in the course of this experimental program, such as would include the detailed presentation of all of the hematological, biochemical, clinical and analytical observations believed to be useful in establishing normal baselines and certain threshold values, has been assembled. Many of the data have merely been scanned for significance, and, in their apparent acceptability, laid aside for later detailed presentation. The principal findings, i.e., those related most directly to the main objectives of the investigation, have been published, in somewhat less than extended form, from time to time. In 1960, a series of three lectures were given, in an effort to present the high points of these investigations in a running, but fairly well illustrated and documented account. These were published under the title of "The Metabolism of Lead in Man in Health and Disease," as The Harben Lectures, 1960, of the Royal Institute of Public Health and Hygiene, London, England, in the J. Roy. Inst. Public Health. 24:81; 101-129; 177, 1961. Other papers (prior publications of the Kettering Laboratory) concerned with this series of investigations were the following: (a) Experimental Studies on the Ingestion of Lead Compounds. J. Industr. Hyg. and Toxicol. 22:381, 1940. (b) Experimental Studies on Lead Absorption and Excretion, and their Relation to the Diagnosis and Treatment of Lead Poisoning. J. Industr. Hyg. & Toxicol. 25:71, 1943. (c) Exposure to Lead. Occup. Med. 3:156, 1947. (d) Experimental Studies on the Inhalation of Lead by Human Subjects. Pure Appl. Chem. 3:129, 1961. (e) Criteria For Human Safety From The Contamination of The Ambient Atmosphere With Lead. Prepared for publication in the Proceedings of the XV International Congress on Occupational Health, Vienna, Austria, September, 1966. All of the foregoing

publications, with the exception of the last, have been supplied to representatives of the sponsors of the experimental program. The information to be made available in the paper referred to last, is supplied in this report, in somewhat more extended form.

The physiological, toxicological and clinical literature relative to lead and lead poisoning, which has appeared during and for some years prior to the initiation of this ~~program of~~ investigation, has been voluminous, and could scarcely even be referred to, much less abstracted and summarized, within a short period of time or in a report or publication of less than momumental proportions. (The bibliography of pertinent contributions during 1965 alone, abstracted, translated (when necessary), and assembled by the staff of the Kettering Laboratory, runs to 236 single-spaced, typewritten pages.) No other prolonged experimental observations of the type of "balance experiments" on human subjects have been described, however, so that there are no strictly pertinent references to be made than those (a-e) given above.

EXPERIMENTAL RATIONALE

As indicated in the several proposals or reports to various sponsors, the experiment under way at that time, and projected for continuation, involved 2 men in parallel observations, in which time (duration of exposure per unit of time over an extensive period of time) was to be the sole significant variable.

The rationale of this experiment lay in certain previously observed facts, as follows: - When ~~experimental subjects~~ selected persons had been subjected to the oral administration, daily, of a standard dosage of lead in solution along with their meals, over periods of months and years, thus providing for a slightly variable but continuous rate of absorption from the alimentary tract during every successive period of 24 hours, the rate of the excretion of lead in the urine and feces, and the rate of accumulation of lead in the body, with some seasonal (and other) variability, continued at a substantially uniform yearly rate, without any sign of tapering off during the entire period of the

administration of lead, in one instance, for 4.5 years. When, however, other experimental subjects were confined in respiratory chambers within which they inhaled lead dispersed in air (under standardized and controlled conditions) for 7.5 hours per day on 5 days per week (in simulation of the usual occupational regimen), the rate of the excretion of lead in the urine, and the concentration of lead in the blood increased steadily to a nearly constant level (in about 16 weeks), after which it continued essentially unchanged so long as the experimental conditions remained constant.

It is obvious, therefore, that continuous exposure to an absorption of lead, above a certain mean level (at which day-to-day variability in intake does not counteract the continuity of continuous absorption), results in progressive accumulation of lead in the body. It may be presumed, for the lack of evidence to the contrary, that this state of affairs persists during the lifetime of the individual, (or, during the working lifetime) and that this will lead, at a sufficiently high rate of intake, to the accumulation of a dangerous "body burden" of lead. Under the prevailing environmental conditions in this country, with respect to the absorption of lead by persons in the general population, no such accumulation, or only a negligible degree of accumulation, occurs during a lifetime. This phenomenon can be accounted for, on the basis of the relative insignificance of the quantity of absorbed lead that is retained in the body from day to day - a quantity too small to be detected in our "balance experiments," or in the analytical findings at necropsy, in the face of the variability of the terminal findings. Barth (1), Tompsett (2), and Schroeder (3) have submitted evidence, which, in their judgment, was to the contrary, but none of these investigators limited the sources of human specimens for analysis to those definitely

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(1) Barth, E. Untersuchungen uber den Bleigehalt der Menschlichen Knochen - Virchows Arch. f. path Anat, 281:146, 1931.

(2) Tompsett, S.L. Determination and Distribution of Lead in Human Tissues and Excreta, The Analyst 81:330, 1956.

(3) Schroeder, H.A. et al Abnormal Trace Metals in Men: Lead. J. Chronic Dis. 14:408, 1961.

identified as lacking in any influence other than the current general environmental exposure to lead. This point is crucial. An additional feature of the environmental exposure to lead contributes further to the paucity of evidence indicative of the progressive accumulation of lead in the bodies of persons in the general population with advancing age. This factor is the variability in the rate of intake and in the consequent variability in the rate of absorption of lead, from person to person, in the population, because of variable selection and utilization of food, qualitatively and quantitatively, by individuals. Bodily size, occupation, climate, habits of life and of upbringing, social and economic status, and a variety of other factors influence the manner of eating and the quantity and variety of food consumed. These factors, in turn, determine the quantities of lead taken into the alimentary tract. Moreover, at different periods in a single lifetime, from infancy, through childhood and adolescence, adulthood and old age, the quantity and variety of the food (and its content of lead) consumed by an individual undergo considerable variation, with the result, akin but not equivalent to that of temporal intermittency of exposure, that there are times when lead is being lost from the body, and others when the reverse is true. In this connection, the balance experiments on "normal," healthy, young subjects have demonstrated once and for all time, if there has been any very serious doubt about the matter, that the lead which finds it way into the human skeleton is actively involved in the metabolism of the body. The osseous metabolism, as lead is involved therein, is slow, as compared to that of other tissues of the body, but it responds promptly, although slightly and gradually, to conditions that disturb the metabolic equilibrium, such as increase and decrease in the rate of absorption of lead into the body, or the increased loss of lead from the "soft tissues" of the body following chelation therapy. (The metabolism of lead is not influenced appreciably, under the ordinary conditions of life, or within the limits of cautious experimentation, so far as present evidence is concerned, by that of calcium or phosphorus or of any other mineral element, but is, rather, strikingly stabile and predictable in

its behavior in the body, as has been demonstrated clearly by these experiments. Moreover, the uniformity in the responses of a series of subjects to a variety of experimental conditions, has been little short of remarkable.)

Because of the facts recited above, it seemed that the demonstration of the maximum concentration of a reproducible dispersion of absorbable lead in the air that could be inhaled continuously, with complete safety, by normal healthy human subjects, would require direct observation of the metabolic effects of continuous exposure at levels above those now prevalent in the community. Since such direct experimental observation was not feasible, except, perhaps, through the expenditure of inordinate sums of money, through the actual creation of conditions of housing, subsistence and prolonged maintenance of an essentially normal conduct of life, for the experimental subjects, an alternative approach was thought to be that of establishing the gradient of the response of human subjects to incremental increases in the duration of intermittent but quantitatively constant exposure, from which to extrapolate to a near approximation to the metabolic effects of continuous exposure. The use of the expression, "approximation" arises from the likelihood that, as the experimental intermittency approaches continuity, the shortening periods of freedom from exposure and the lengthening periods of exposure will operate against the achievement of a balance between the intake and output of lead, so that the curve indicative of the progress of events will assume a trend bending upward from a straight line (c.f. Fig's 1 and 2). From the strictly practical point of view, this deviation should have little importance, especially when the concentration of lead in the respired air is low.

EXPERIMENTS CARRIED OUT SINCE 1962

Preliminary Experiment

The feasibility of the experimental approach described above was tested in an experiment involving detailed observations (including all of the procedures described in

prior balance experiments) of two human subjects over the period of 144 and 178 weeks, respectively. (The period of observation of one of the subjects, prior to the initiation of experimental exposure to lead, was unusually prolonged so as to enable the other, secured later, to undergo his initial period of experimental exposure to lead on the same date as the first, and so to continue parallel procedures.)

Subject [redacted] and [redacted] were engaged in daily preliminary (control) observations, (determinations of their daily intake of lead in food and beverages, and their output of lead in the feces and urine, in association with a battery of hematological, clinical, microscopic, and physical examinations, some at daily, and some at weekly, intervals), over periods of 56 to 86 weeks, respectively.

They entered the two correspondingly designed respiratory chambers into which air containing 150 micrograms of lead per cubic meter, as the sesquioxide, dispersed in particles ranging up to 0.18 micron in diameter, and having the mean diameter of 0.05 micron, was being introduced and evenly distributed, and from which it was being evacuated, stripped (by electrostatic precipitation) of its lead, and discharged into a stack. The design of the experiment called for exposure to these conditions on the following schedule: 16 weeks for 10.5 hours per week; 16 weeks for 21 hours per week; and so on, 16 weeks at a time, for 31.5 and 42 hours per week. (This schedule did not result in carrying the severity of the exposure beyond the level which has been established, by both occupational experience and physiological experimentation, as being well within the limits of safety.)

During each successive period of 16 weeks, as has been the case in every other experiment involving intermittent exposure to lead in simulation of occupational experience (i.e. in practice, 40 $\pm$ 4 hours per week), the level of the urinary output of lead per day, and the concentration of lead in the urine of the subjects, as well as the concentration of lead in their blood, rose up to a certain point and then continued at essentially constant levels, so long as the conditions of exposure remained unchanged;

each incremental increase in the weekly duration of the exposure at least resulted in a further graduated increase in the levels of the lead in the urine and blood; a line connecting the points representing the final (or nearly final, i.e. average) levels of the concentration of lead in the blood reached in the successive periods of 16 weeks became a straight line of uniform upward slope. The results of the extension of this line, at the same slope, to a point that corresponded, in the case of each subject, to continuous exposure (i.e. 168 hours per week) is shown in Figure 1, (Subject L.D.), and in Figure 2, (Subject [redacted]). Other analytical data of this experiment have been grouped and charted in Figures 3, 4 and 5 (Subject [redacted]), and in Figures 6, 7 and 8 (Subject [redacted]. Several features of these charts are noteworthy, but are generally similar to those observed and commented on in previous experiments, the results of which have been reported in the Harben Lectures. The unique feature is found in the comparison between the observed equilibrated responses to varying degrees of intermittent exposure, and the extrapolated response to continuous exposure. The gross difference between the end-points of these two situations, with respect to the absorption of lead, is a matter to ponder in relation to the lead content of the ambient atmosphere. It is evident that the only sure provision for human safety lies in the avoidance of a persistent level of combined alimentary and respiratory dosage that will result in a measurable accumulation of lead in the bodies of exposed persons in the course of a lifetime.

The results of this experiment achieve their greatest practical importance for present purposes, however, in indicating the feasibility of this method of investigation for determining the maximum concentration of lead in the ambient atmosphere (in association with the current intake of lead in food and beverages), that is compatible with human health and well-being. The promptness and the quantitative orderliness of the physiological response to a graduated shift toward continuity of exposure, as represented best by the incremental increase in the concentration of lead in the blood, demonstrates clearly the essential reliability of this criterion as an indicator of the

absorption of lead, and gives strong support to the experimental hypothesis which was being tested.

Critical Experiment

With the evidence of the foregoing experiment at hand, a further experiment was designed in an effort to determine the level of exposure to airborne lead, which, under previous experimental conditions with respect to the compound of lead and its dispersion into the air, would barely yield a physiological response, in the form of the least demonstrable increase in the urinary excretion of lead, without corresponding increase in the concentration of lead in the blood, as in the experiments involving the oral administration of lead. (It has been shown, previously, and by other means, that the earliest evidence of an increase in the rate of the absorption of lead by the intact (living) human organism is a measurable increase in the rate of the excretion of lead in the urine).

Two new subjects were obtained and investigated as to the patterns of their intake and output of lead under standard conditions in the laboratory, one, (Subject [redacted]), over the period of 29 weeks, and the other, (Subject [redacted]. secured somewhat later in the year), over the period of 19 weeks. Following these preliminary observations, which demonstrated the suitability of the subjects, the two embarked upon parallel schedules (parallel in type but not in the time of starting) of exposure to lead in the concentration of 10 micrograms of lead per cubic meter of air. The temporal schedule, as a rule, involved 16 weeks of exposure, at each of the prescribed series of weekly rates, (10.5, 21.0, 31.5, 42.0, 52.5, 63.0 and 73.5 hours per week), although for different reasons (the convenience of a subject - a short vacation from exposure, to visualize the effects, if any, of freedom from experimental exposure - or the desirability of extending a period of exposure so as to check on its adequacy in disclosing the peak of the physiological response), the schedule of each subject was altered slightly.

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The terminal results of this experiment are not available as yet, but the sequence of the more significant events during the total period of the exposure of one (Subject N.K.), and during all but the final period of the exposure of the other (Subject S.S.), are portrayed in Figures 9 and 10, and 11 and 12, respectively.

It appears that there was a slight increase in the output of lead in the urine and also, somewhat surprisingly, in view of the scantiness of the elevation of the urinary excretion, a slight increase in the concentration of lead in the blood, of both subjects The increase was irregular, late in putting in its appearance in the case of Subject S.S and of such slight proportions in both instances as to be of dubious significance. The trends, however, appear to be sufficiently persistent to be valid. Unfortunately, their interpretation is uncertain, for the reason that the lead content of the food and feces of both subjects was somewhat more than usually variable*, and the increases above the average level tended to coincide with those of the output of lead in the urine and the concentration of lead in the blood. In neither case was it possible to plot a curve that would fit, adequately, the findings in either urine or blood from which a valid extrapolation could be projected. (In Figure 8, the upward and downward trends in the concentration of lead in the blood of Subject [redacted] for several months at a time, have been indicated roughly by a dotted line in illustration of this difficulty.) It may be that the concentration of 10 micrograms of lead per cubic meter of air is near the threshold value which is being sought in these experiments. To ascribe such significanc to these findings would be to deal irresponsibly with evidence of a type that has been found previously to be misleading. These data are being examined in somewhat greater detail by members of the statistical staff to wring out of them such further relationships as may be found. Meanwhile, a further experiment is being designed to yield definitive observations on which a plausible extrapolation can be projected.

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* The gross contamination of the food of Subject [redacted] resulted from his use of the flesh of a steer killed by gunshot, with unexpected shattering and spread of minute fragments of the bullet beyond visible pathways.

Projected Experiment

One of the subjects (Subject [redacted]) of the experiment described above is regarded as suitable for employment in a further experiment (after a period of further observations intended to reveal any decrease in response following the termination of his former experimental exposure). He has found it possible to act in this capacity. Meanwhile, a new subject, (Subject [redacted]), has been found and has been inducted into the preliminary observations which should prepare him for the initiation of exposure in a regimen that will run parallel to that of Subject [redacted].

The new experiment will continue along the lines of the one now drawing to an end. Two modifications are now under consideration. Some doubt is entertained as to whether the maximum physiological response to exposure to lead at a low level has always been achieved in 16 weeks. Therefore, it seems wise to extend the first productive period of exposure (one that yields a definite positive response) over a length of time at which the maximum response will surely have been observed. This period of time can then be employed throughout the remainder of the experiment.

The second modification will be concerned with the concentration of lead to be maintained within the respiratory chamber. Considering the desirability of bringing this phase of the investigation to the earliest possible conclusion by securing data that can hardly be dubious in significance, the figure of 20 micrograms per cubic meter of air seems less likely to err on the low side, than would that of, for example, 15 micrograms per cubic meter. The evidence obtained in several extensive field investigations, (including the most recent and most extensive, geographically and numerically, in which the Public Health Service participated, among other governmental groups and ourselves, has failed to demonstrate differences in the responses of large groups in the population of cities that would seem to have differed significantly, with respect to the average yearly concentration of lead in their entire atmospheric environment. For

example, the populations of Los Angeles and Cincinnati, as investigated, were undifferentiable in terms of the concentration of lead in their blood, despite a two fold difference in the average content of lead in the air. The lead content of the air in both cities was low, to be sure, but the exposure of the population thereto must be regarded as having been continuous. These facts argue that neither was sufficient to reveal its effects in the face of the quantity and variability of the absorption of lead from alimentary tract. This fact, which seems to have escaped interpretative critique, makes for considerable doubt concerning the usefulness of slight differences in atmospheric conditions (with respect to lead) in experiments in which the effects of graduated temporal intermittency are expected to give the clue to the effects of continuous exposure.

Whatever may be the ultimate basis of judgment concerning the primary conditions of further experimentation along this line, there is little definitive information by which to be guided. The relationships under investigation have long been questioned, but there have been no answers. There is good reason, however, to believe that, with the completion of this further experiment, the results of which may be somewhat on the far (or positive) side of our objective, it may be possible to visualize, with appropriate clarity, the middle ground of a suitable physiological criterion for avoiding deleterious effects due to lead. A question will still remain as to the proper allowance that must be made for those in the general population who may be unusually susceptible, for one reason or another, to the toxic effects of the absorption of lead. It may be useful, in the connection, to assemble a few pertinent facts that may make for a degree of balanced judgment in anticipation of such a need, and to suggest that a reasonable decision may be arrived at.