

## BROOKHAVEN NATIONAL LABORATORY

## MEMORANDUM

REPOSITORY Records Holding Area Bldg. 494  
 COLLECTION Protocols-Clinical  
 BOX No. 4  
 FOLDER HUMAN Protocols 1950-1963

DATE: March 11, 1957

TO: Isotope Committee

FROM: Doctors George C. Cotzias and  
Donald C. Borg DCBSUBJECT: Request for permission to  
use  $Mn^{54}$  in patients.

We herewith request that permission be granted for the use of  $Mn^{54}$   
 in trace amounts in humans (up to 50  $\mu$ c) for investigative purposes.

Some pertinent information is herewith included to assist the committee  
 in evaluating this request.

## CHEMISTRY

We plan to administer manganese in its divalent form as a dissociable  
 salt, either as  $Mn^{54}Cl_2$  or  $Mn^{54}SO_4$ . Following administration, the  $Mn^{++}$  ion is  
 expected to be partially converted into the poorly soluble --ous hydroxide and  
 the --ic oxide. This expectation is based on the body's pH and redox potential.

In some tests, the metal will be administered as a dissociable chelate.

## TURNOVER AND METABOLISM

From our experiences in mice and rats, it appears that the primary  
 site of accumulation of  $Mn^{54}$  is the liver, as it is with  $Mn^{56}$  in animals and man  
 (11, 14). This site can be markedly favored to the point of almost complete  
 initial exclusion of the other sites by means of delivery of the isotope directly  
 into the portal vein. Under these conditions, the liver discharges a very large  
 percentage of the dose into the gut and [redistributes] the rest (is then redistributed) to other organs.  
 The route of discharge of manganese from the body is the feces, except when  
 chelating agents divert the element into the urine.

The total body turnover has been studied only in mice (11). With the

subject on a normal diet, injected  $Mn^{54}$  shows two rates of turnover with half times

1176337

March 11, 1957

of 40-56 hours and 400-500 hours respectively. The first ordinarily accounts for the loss of about 60-70% of the dose, while the latter accounts for the rest. The turnover can be markedly accelerated by a) feeding manganese salts, b) injecting any inorganic manganese compound including metal powder. It has been slowed down by constipation and starvation but has not been affected by feeding or injecting excesses of members of the first transition group or of group VII (i. e. rhenium) of the elements. On the basis of partition studies, the total body turnover is expected to become accelerated when animals are given versene injections.

#### METABOLIC ROLE

Work by Orent and McCollum (3) has shown that manganese is essential to nervous function and reproduction in mammals. The disease Perosis of the chicken is a specific manganese deficiency. Some work has suggested that manganese might play a role in normal hematopoiesis (9). There is an immense amount of in vitro work which demonstrates the essentiality of  $Mn^{++}$  ions in enzyme systems related to oxidation, hydrolysis and synthesis (12). Preliminary studies have implicated manganese itself or biological systems known to be sensitive to manganese in several widely prevalent diseases of man whose etiologies are presently unknown (11, 12).

#### CONCENTRATION AND DISTRIBUTION

Manganese in trace amounts is ubiquitously present in all animals ~~examined~~ and in all organs. The liver, pancreas, kidney and intestine are the richest sites (7), but they also constitute pools of high turnover (11). Their turnover can be materially accelerated by feeding or injecting any inorganic manganese compound, including the metal (11). Chelating compounds such as versene (EDTA) will divert manganese in transit from its normal excretion via the gut into urinary excretion and will thus expedite the turnover of the liver, the only organ so tested (11).

1176338

The total amount of body manganese has been reported as 220 mgm. for adult man (7). A recalculation of data obtained at Oak Ridge by Dixon yields only 15 mgm. of manganese for the entire body. This discrepancy cannot be reconciled as yet. We shall assume that the latter value is correct, as this will lead to safer operations.

#### MANGANAL THERAPY

The only established application is that of per-manganate salts for (external) antiseptic by virtue of oxidation. There are references in the literature, however, pertaining to treatment of human schizophrenia, human anemia and human skin diseases with manganous salts (6). These approaches are by no means established (4). In veterinary practice, berosis of chickens is both prevented and cured by feeding  $MnSO_4$ . Horses used for antiserum production will a) yield much more antitoxin, b) suffer much less amyloidosis if fed manganous sulfate (13).

#### TOXICITY

Both acute and chronic toxicity are known in man (1). The acute toxicity consists of gastrointestinal symptoms (diarrhea) when Mn salts are given in large doses (grams) by mouth. For potassium permanganate, the fatal dose by mouth is about 15 gm, and this toxicity is probably due to the specific reactivity of the permanganate ion (8). Sudden deaths have been seen in animals and suspected in man after parenteral administration of doses consisting of many milligrams of the metal (8, 9); however, there appears to be general agreement that only a very small amount of manganese is absorbed from the alimentary tract (1, 8, 10, 11, 12), and serious toxicity via this route has not been reported. Amounts of 4-5 gamma have been given intravenously by us to a large number of <sup>large</sup> humans without any reaction, and repetition of this administration several times

March 11, 1957

in some patients produced no ill effects (11).

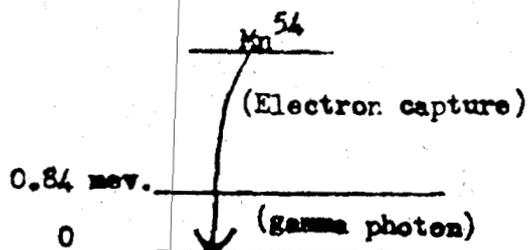
The chronic form of toxicity results in a neurological syndrome kindred to that of Wilson's and Parkinson's disease (2). This has been encountered a) in miners of manganese ores and of ferrous metals, b) in workers making battery elements, c) possibly in one population which had its water supply accidentally contaminated.

The amounts necessary for establishment of chronic poisoning have not been defined, but they appear to be massive as compared to the 4 mgm/day in average human diets and even more so as compared with our proposed doses of less than 10 gamma. This is supported by the fact that the safe concentration of manganese dusts in air is 6 mgm. of dust per M<sup>3</sup> of air (8), despite inhalation being the commonest route of systemic poisoning (2). Exposure to manganese dusts, furthermore, has to be prolonged (years to decades). Apparently, any form of manganese compound has to be suspected as capable of producing poisoning, including the trivalent oxide which is most insoluble in water. Once established, the disease has been thus far, both irreversible and progressive, but current experimental data suggest that this might not be so following the advent of the versenes.

#### RADIATION CHARACTERISTICS

Manganese 54 is produced in the cyclotron by deuteron bombardment of chromium 54. It can be purchased from Oak Ridge. It ordinarily is contaminated with some Mn<sup>52</sup> (T<sub>1/2</sub> 140 hours) but not by its isomer which has a T<sub>1/2</sub> of  $\approx$  2 minutes. It decays back into inert chromium 54 by the following decay scheme.

#### Decay Scheme of Mn<sup>54</sup>



Half life: 291 days

Total disintegration energy: 1.84 mev.

1176340

## RADIATION DOSE CALCULATIONS

In order to have the most conservative estimate of the dose from  $Mn^{54}$ , the following assumptions are made which all tend to yield a maximum estimated dose:

- (1) That the rate of energy absorption at a given site equals its rate of release from that site, thus making the dose formula ordinarily used for beta particles applicable.
- (2) That the patient weighs 70 kg and his liver 1500 gm.
- (3) That 30% of the radiation dose goes to the liver and 70% to the rest of the body and that this is distributed homogeneously in each case (11, 14).
- (4) Physical decay is neglected.
- (5) That the metabolic half life is longer than the longest seen in mice — namely 600 hours (=25 d) and that there is no short or spillover component to the turnover. (In view of the fact that the proposed study is the first of its kind, there are no available human data pertaining to turnover studies. Furthermore, <sup>accurate</sup> balance studies have not been reported, presumably because of the absence of satisfactory analytical techniques for manganese and the low ratio of metabolized manganese to dietary and fecal manganese. A conservative use of the animal data provides the best guide at the present.)
- (6) That a 50  $\mu$ c dose will be used. (See SPECIFIC PROPOSAL).
- (7) That the total disintegration energy (1.84 mev) is absorbed, rather than only a fraction of the gamma energy (0.84 mev) and of the K-electron-capture rays.

Then: For initial dose rate to liver (receiving 15  $\mu$ c)

$$d = 51.2 C_t E$$

$$d = (51.2)(0.01)(1.84)$$

$$d = .94 \text{ rad/day}$$

1176341

For cumulative liver dose

$$D_{\infty} = 73.8 C_0 ET$$

$$D_{\infty} = (73.8)(0.01)(1.84)(25)$$

$$D_{\infty} = 34 \text{ rad.}$$

For initial dose rate to whole body (receiving 35  $\mu$ c)

$$d = 51.2 C_t E$$

$$d = 51.2 (5 \times 10^{-4})(1.84)$$

$$d = 0.05 \text{ rad/day}$$

For cumulative body dose

$$D_{\infty} = 73.8 C_0 ET$$

$$D_{\infty} = 73.8 (5 \times 10^{-4})(1.84)(25)$$

$$D_{\infty} = \frac{1.7}{17} \text{ rad.}$$

## SPECIFIC PROPOSAL

The total turnover of manganese and its redistribution in man cannot be studied with the hitherto used  $Mn^{56}$  or  $Mn^{52}$ . Therefore, we propose to study the turnover and distribution of manganese in man by means of  $Mn^{54}$ . Tracer doses will be given to patients suffering from chronic diseases such as Parkinsonism, Cancer, Polycythemia, Rheumatoid Arthritis, etc. Serial blood samples will be drawn to establish blood clearance curves, and repeated in vivo scanning will be carried out to determine total body and localized (organ) uptake kinetics and redistribution. The feces will be collected as long as they are countable and so will the urines, although the latter have never been countable in previous human experiments using  $Mn^{56}$  or  $Mn^{52}$  in the absence of chelating agents. Only one administration per patient is anticipated.

Studies with  $Mn^{56}$  in man have indicated that satisfactory in vivo scanning and blood clearance data are obtainable with 10 - 20  $\mu$ c of radiomanganese. Since

1176342

slightly larger tracer doses than these may be required for long-term studies involving biological decay, it is expected that  $Mn^{54}$  will ordinarily be injected intravenously in dosages of 20 - 25  $\mu c$ , and no dose larger than 50  $\mu c$  is anticipated.

Theoretically  $Mn^{54}$  is prepared carrier-free. Spectrographic checks of several samples delivered to us have either confirmed this or have indicated but very slight metal contamination. Therefore, it is expected that the amount of manganese metal administered shall not exceed 10 gamma.

Since no other isotopes are to be used during these tests, the radiation doses should be less than those presented under the general proposal, wherein strongly maximizing assumptions were made. There is no radiological or pharmacological hazard to the patient as far as can be judged beforehand.

## REFERENCES

- (1) Goodman and Gilman: "The Pharmacological Basis of Therapeutics". Macmillan and Company. New York, 1955. Pages 993-994.
- (2) Flinn, R. H.; Neal, P. A.; Reinhart, W. H.; Dellavole, J. M.; Fulton, W. B.; and Booley, A. E.: "Chronic Manganese Poisoning in an Ore-crushing Mill". U. S. Public Health Service, 1940, Bulletin 247.
- (3) Orent, E. R. and McCollum, E. V.: "Effects of Deprivation of Manganese in the Rat". J. Biol. Chem., 1931, 92 (651-78).
- (4) Report of the Council: "Manganese Compounds for Cutaneous Diseases, Cocciogenic Infections and Blood Formation not Acceptable for New and Nonofficial Remedies". J. Am. M. A., 1940, 114 (248-49).
- (5) Sheppard, C. W.; Walls, E. B.; Hahn, P. F. and Goodwall, J. P. B.: "Studies of the Distribution of Intravenously Administered Colloidal Sols of Manganese Dioxide and Gold in Human Beings and Dogs Using Radioactive Isotopes".
- (6) Sullivan, M.: "Manganese Hydroxide in the Treatment of Acne Vulgaris, Postular Acne, Furunculosis, Sycosis Vulgaris and Psoriasis". J. Am. M. A., 1940, 114 (246-248).
- (7) Ottungen, W. F., Von: "Manganese: Its Distribution, Pharmacology and Health Hazards". Physiol. Reviews, 1935, 15 (175-201).
- (8) Thienes, Haley: Clinical Toxicology, 3rd Edition, Lea and Febiger, 1955.
- (9) Bridges: Dietetics for the Clinician, 4th Edition, Lea and Febiger, 1941, pages 133, 196, 798.
- (10) Kehoe et al: J. Nutrition, 1940, 19 (579) and ibid, 20.
- (11) Cotzias and Borg: Unpublished.
- (12) Schroeder, H. A.: "Trace Metals and Chronic Disease". Advances in Internal Medicine, 8 (259-303) 1956.
- (13) Walburn, Schmidt: Zist. F. Immun, Naturforschung. 1924, 42, (32-45).
- (14) Maynard, L. S. and Cotzias, G. C.: "The Partition of Manganese among Organs and Intracellular Organelles of the Rat". J. Biol. Chem., 1955, 214 (489-495).

1176344

COMMITTEE SIGNATURES:

*Lee E. Farr*  
Dr. Lee E. Farr

*Lewis K. Dahl*  
Dr. Lewis K. Dahl

*Robert A. Love*  
Dr. Robert A. Love

*Elmer E. Stickley*  
Dr. Elmer E. Stickley

*James S. Robertson*  
Dr. James S. Robertson

*limit studies to 5/year per patient*

*Victor P. Bond*  
Dr. Victor P. Bond

*Eugene P. Cronkite*  
Dr. Eugene P. Cronkite

*Existing, polyarthritides already have traces of <sup>60</sup>Co B<sub>12</sub> in them and probably would not be justified to use <sup>54</sup>Mn in them unless turnover more rapid and would not interfere with alternate analyses determination of <sup>60</sup>Co.*

Amendment #1, dated July 11, 1960, to isotope Request H-49 is hereby approved.

*Walton W. Shreeve*  
Walton W. Shreeve, M.D., Ph.D.  
Chairman

*John L. Bateman*  
John L. Bateman, M. D.

*Elmer E. Stickley*  
Elmer E. Stickley, Ph. D.

*James S. Robertson*  
James S. Robertson, M.D., Ph.D.  
Ex Officio

Dated July 18, 1960: