to the enzyme with consequent inactivation, or irreversible covalent changes in the apoenzyme or coenzyme–apoenzyme complex.

Our results to date do not allow us to determine which, if any, of these factors is operative here. The changes in dietary habits of the animals treated with decaborane are unlikely to have been a significant factor in the marked loss in enzyme activity since it has been shown that the activity of this enzyme is unchanged in the tissues of rats fasted overnight (10). Studies are in progress to determine if boron is bound by the aminotransferase apoenzyme or if the structural charactersitics of the enzyme are altered by boron hydrides.

These and other data from our laboratory suggest that boranes have a predilection for pyridoxal enzymes, and this hypothesis is supported by additional studies in this series indicating that lactic dehydrogenase (EC 1.1.1.2.7) activity is not altered significantly by decaborane (W. N. Scott, J. H. Landez, and H. D. Cole, in preparation). If substantiated by further studies, our findings may result in two advances: (a) an underlying hypothesis for the mechanism of the toxicity of boron hydrides; and (b) a simple and effective technique for inhibiting pyridoxal-catalyzed enzyme systems *in vivo*.

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## Zinc Metabolism in Schistosomes (32777)

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The relationship of zinc metabolism to schistosomiasis is of particular interest due to recent observations by Prasad *et al.* of a dwarfism syndrome in Egyptian males associated with low serum zinc levels (1–3). Growth failure, hepatosplenomegaly and hypogonadism manifested by absence of pubic, axillary, and facial hair; small penis, and atrophic testes were consistent findings in these patients. Both hookworm and *Schistosome haematobium* infections were observed in many patients and all had hypochromic anemia. The diet of the patients was high in cereals, low in animal protein and green vegetables. Nutrition was proposed as one factor in the deficiency, and blood loss due to parasites as another. Since schistosomiasis is present in a very high percentage of the population in the Nile delta, it was hypothesized that there might be a relationship between zinc deficiency and schistosomiasis.

Two basic experiments were set up: (i) to determine the $^{65}Zn$ uptake by schistosome worms and eggs in infected hamsters, (ii) to

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determine any differences in the biological half-life of tracer doses of $^{65}Zn$ in noninfected and infected animals.

*Materials and Methods.* A. *$^{65}Zn$ uptake by schistosome worms and eggs.* 1. Groups of hamsters were selected of similar size with 60-day-old schistosome infections of approximately 250 *S. mansoni cercaria.* Each group had been infected on the same day from one suspension of cercaria.

2. Each hamster with injected intraperitoneally with 18 $\mu$C of $^{65}ZnCl_2$ and maintained on a standard animal diet in similar cages until time of sacrifice.

3. The time intervals from injection to sacrifice were 0.5, 1, 2, 24 hours, 3, 10, 15, and 21 days. The total number of hamsters used was 31.

4. At sacrifice each hamster was weighed, injected intraperitoneally with 0.3 ml of nembutal and 0.3 ml of heparin.

5. Approximately 1 ml of blood was removed with a heparinized syringe from the right ventricle, transferred to a tared counting tube, and the tube was corked. A 0.5-gm liver sample was removed, transferred to a tared counting tube, the tube was corked, and both tubes were weighed.

6. Adult worms were collected by the perfusion method described by Browne and Schulert (4), male and female worms separated, counted numerically, and transferred to isotope counting tubes.

7. Schistosome eggs were collected separately from the liver and intestine by the method of Browne and Thomas (5), separately processed and counted. Aliquots of egg-containing solutions were used for numerical counting purposes, the remaining egg solutions were transferred to isotope counting tubes, and the number of eggs per tube were computed.

8. All samples were assayed for $^{65}Zn$. Wet weights of the worms and eggs were computed using Browne and Schulert's weight/worm or egg figures (4), and $\mu$C/gm of wet weight worm was computed.

*B. The effect of schistosomiasis on the biological residence time of $^{65}Zn$.* 1. Twenty-four male hamsters of approximately 6 weeks age and 60 gm weight were selected, weighed on day 0 and weekly thereafter to determine growth curves. Food consumption was measured for each of the two groups.

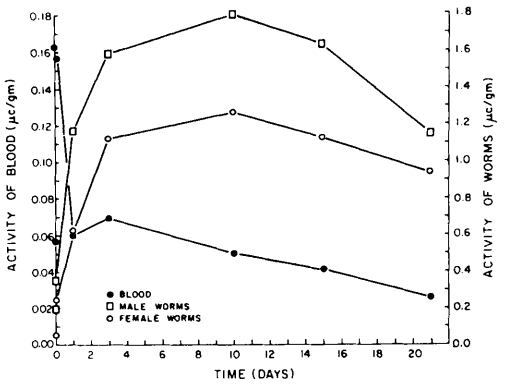


FIG. 1. Zinc-65 activity in blood and male and female schistosomes vs time.

2. Each hamster was injected intraperitoneally with 1.0 ml of a $^{65}ZnCl_2$ solution (activity 0.219 $\mu C$/ml). Each hamster was counted in a whole body radiation counter at intervals up to 50 days. The $^{65}Zn$ standards were used to correct for fluctuations in the detector.

3. On day 2 half of the hamsters were infected by exposure to a suspension containing 264 *S. mansoni cercaria.* The other half were handled similarly but no cercaria were introduced.

4. The radioactivity was measured as above and the average $^{65}Zn$ content per hamster computed for the infected group and the noninfected group.

5. The experiment was terminated on day 50. The animals were counted for a final time, killed by injection of nembutal and heparin, the schistosome worms were collected by the perfusion technique described by Browne and Schulert (4) and counted.

*Results.* The uptake of $^{65}Zn$ by the various tissues is shown on Figs. 1 and 2. Each graph shows the activity as $\mu$C/gm of wet weight tissue, corrected for the radioactive decay of the isotope.

The concentration in blood rises rapidly in the first few hours postinjection, falls considerably by day 1, and slowly declines from day 1 to day 21. Standard deviation averaged 18%.

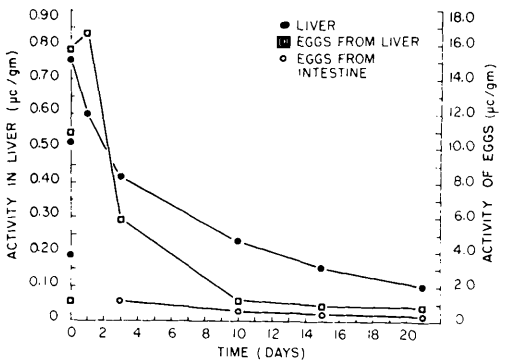


FIG. 2. Zinc-65 activity in liver and schistosome eggs vs time.

The $^{65}Zn$ concentration in the schistosome worms rises within a day to 10–20 times that of blood. The concentration continues to rise to a peak around day 10, and gradually to decline thereafter. Standard deviation averaged 28%.

Worms of both sexes had much higher concentrations of $^{65}Zn$ than blood, and that in males was consistently greater than in females. Stable zinc concentration was found to be 0.11 ± 0.3 mg of Zn/gm of worm for males and 0.09 ± .02 mg of Zn/gm of worm for females.

The ratio of worm/blood $^{65}Zn$ concentration seen in Table I shows a constant increase from day 1 to day 21 for both male and female worms.

Changes in liver $^{65}Zn$ activity vs time (Fig. 2) are similar to those in blood. The $^{65}Zn$ concentration rises rapidly in the first few hours postinjection and then gradually declines throughout the observation period. The liver concentrations, however, are approximately 5–10 times higher than the blood concentrations. Standard deviation averaged 18%.

The graph of $^{65}Zn$ activity in schistosome eggs from the liver, Fig. 2, shows a rapid rise during the first day postinjection followed by a rapid fall in activity from day 1 to day 10. From day 10 to day 21 the activity declines but at a much slower rate than previously. The standard deviation averaged 50%.

In Fig. 2 $^{65}Zn$ activity in schistosome eggs from the intestine, which should be younger eggs than those from the liver, shows a gradual decline from days 3–21. (Data for intestinal eggs were not obtained for day 0–3.) The $^{65}Zn$ concentration in eggs from the intestine was consistently slightly lower than that in eggs from the liver. The reason for this is not clear. Earlier studies of Browne and Schulert (4) demonstrate that antimony uptake averaged 50% greater in intestinal eggs than in liver eggs during the first 24 hours.

The $^{65}Zn$ concentration in schistosome eggs from the liver was 5–28 times that in the liver itself. The $^{65}Zn$ concentration in eggs from the intestine ranged from 10 to 17 times that in the blood.

It is noted (Table I) that the ratio of $^{65}Zn$ concentration in worms to that in the blood rises throughout the entire 21-day observation period. In contrast the concentration ratio in eggs from the intestine to that in the blood falls slightly between days 3 and 10 and levels off, and the concentration ratio in eggs from the liver to that in host liver tissue falls from 28 on day 1 to 8 on day 21.

The determination of biological half-life of tracer doses of $^{65}Zn$ in animals infected and not infected with *S. mansoni* was complicated by the sudden appearance of idiopathic ileitis among the hamsters which killed 15

TABLE I. Ratio of $^{65}Zn$ Concentration in the Parasite to that in Host Tissues.

| | Male worms | Female worms | Eggs from liver | Eggs from intestine |
|---|---|---|---|---|
| Day | Blood | Blood | Liver | Blood |
| 1 | 19.5 | 10.3 | 28.0 | — |
| 3 | 22.8 | 16.3 | 14.0 | 17.2 |
| 10 | 36.0 | 25.4 | 5.2 | 12.0 |
| 15 | 41.0 | 28.2 | 6.0 | 10.0 |
| 21 | 44.2 | 36.2 | 8.0 | 11.5 |

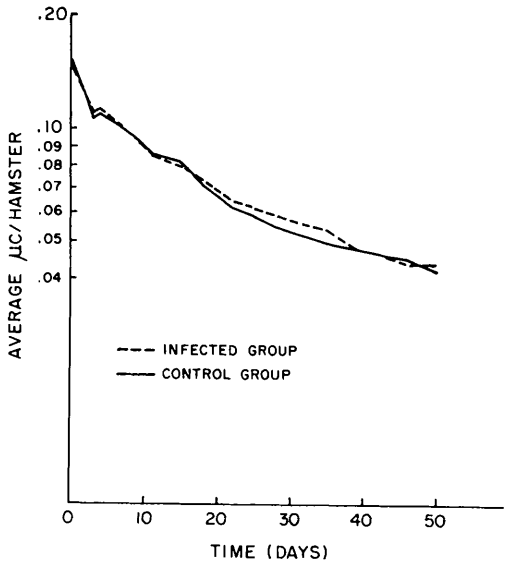


FIG. 3. Average activity/hamster vs time for infected group and noninfected group.

of the 24 experimental hamsters after a 3-week period. This left six animals in the infected group and three animals in the noninfected group. It was not practical to process additional animals and the data were analyzed on the basis of these nine animals. The biological half-life of $^{65}Zn$ in the two groups is shown in Fig. 3. Standard deviation averaged 6%. The biological half-life after 22 days is approximately 50 days.

No significant difference in the biological half-life of zinc was found between the infected and noninfected groups. Likewise there was no significant difference in food consumed or in rate of weight gain. When the infected animals were sacrificed at termination of the experiment, all animals were found to have substantial worm loads with an average of 115 worms/hamster and a range of 43–210 worms/hamster.

*Discussion.* In the determination of $^{65}Zn$ activity in blood vs time (Fig. 1) the rising first portion of the curve probably represents mixing of the isotope as $^{65}ZnCl_2$ is absorbed from the peritoneum into the blood stream. The descending portion of the curve reflects both shift of the isotope from the blood stream into other body compartments and excretion of the isotope. The portion of the curve for whole body biological half-life for this time interval, days 3–21, is approximately 25 days (Fig. 3), while the half-life for $^{65}Zn$ activity in the blood stream for this same time interval is only 12 days (Fig. 1); this would indicate the decrease in $^{65}Zn$ activity in the blood is not solely due to excretion but also to a redistribution of the $^{65}Zn$ from the blood to other body pools.

As noted, the $^{65}Zn$ concentration in worms was highest on day 10, and the worm/blood $^{65}Zn$ concentration ratio increased throughout the 21-day interval to final values of 44.2 and 36.2 for male and female worms, respectively.

The high worm/blood concentration ratio of the isotope is, of course, a reflection of a similarly high ratio for the natural zinc. The failure of the isotope to equilibrate fully with the natural element in this period of time could be due to either of two possible factors (or a combination of both): (i) Zinc uptake and turnover by schistosome worms are slow processes per se, (ii) the $^{65}Zn$ specific activity in the pool from which the worms get their nutrient increases slowly over a period of several days.

It has been found in human studies following the intravenous administration of $^{65}Zn$ that the concentration of $^{65}Zn$ in circulating erythrocytes rises to a maximum on the tenth day postinjection (6). One might therefore speculate that the nutrient for the worms arises primarily from the erythrocytes. However, there is no independent evidence for this, and further investigation is required to determine to what extent these or other factors may be involved.

The $^{65}Zn$ concentration in male worms was consistently higher than in females, in line with the observed stable element values, though the latter difference was not statistically significant. Thus sex variation for zinc differs from antimony which concentrates much more highly in the female following the administration of antimony-containing drugs (4).

A striking observation is that while the $^{65}Zn$ worm/blood concentration ratio increases over the 21-day interval, the egg/blood ratio decreases. This divergence of worm and egg $^{65}Zn$ concentration further indicates that the

eggs metabolize independently of the female worms after being laid.

The determination of biological half-life of $^{65}Zn$ reveals no significant difference between infected and noninfected animals. Differences may have existed in the distribution of $^{65}Zn$ among the various body pools which were not detectable in the biological half-life.

Although these studies do not demonstrate that schistosome infection reduces the body zinc stores, they do demonstrate relatively high zinc uptake by both worms and eggs. Schistosome infection may play a role in pathogenesis of zinc deficiency, not by reducing total body zinc stores, but by redistribution of body zinc.

*Summary.* When $^{65}ZnCl_2$ was injected intraperitoneally into hamsters infected with *Schistosoma mansoni* the incorporation of $^{65}Zn$ into the parasite, including worms of both sexes and eggs, was much greater than the incorporation of $^{65}Zn$ into host tissue. Furthermore, the worm/blood $^{65}Zn$ concentration ratio increased throughout 21 days, being 10 and 20 after 1 day for female and male worms respectively, and 36 and 44 after 21 days. The determination of biological half-life of tracer doses of $^{65}Zn$ in hamsters heavily infected with *S. mansoni* and in hamsters not infected showed no significant differences in half-life between the two groups up to 50 days postinjection.

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## Effect of Nephrectomy and a Low Sodium Diet on the Increase in Plasma Renin Levels Produced by Hemorrhage* (32778)

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The present experiments were carried out to determine if the kidneys are the source of the increased amounts of renin in the circulation following hemorrhage, and if the amount of renin secreted in response to hemorrhage is

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