vitamin A-deficient diet. The data indicate that no improvement in the results obtained in the usual biological assay for vitamin A was achieved by supplementing the basal diet with crystalline ascorbic acid. Somewhat different results might have been obtained, however, had the B-vitamins been furnished in the crystalline form, as in the studies of Mayer and Krehl (*3*), instead of in the form of dried brewers' yeast, as in this study.

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The Effectiveness of Carbarsone in Rendering Young Mice Free of Intestinal Protozoa

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Carbarsone (*p*-carbamino-phenyl-arsonic acid) was used in an attempt to eradicate the intestinal protozoa of young mice needed for other work. These results are presented in the hope that they will be of value to others who are relying upon this arsenic derivative to obtain amoebae-free animals prior to infection with *Endamoeba histolytica*. All the mice used here were raised and maintained on an adequate diet prepared in the laboratory (*4*). Care was taken in the preparation of this diet to prevent contamination, and it was stored in a deep freezer until used. The drinking water provided was sterilized. The doses of the chemical used were suggested by the work of Reed, *et al.* (*5*) on monkeys and rabbits and that of Gabaldon (*2*) on rats. The doses, dissolved in alkaline aqueous solution, were forced into the stomach through a blunted, 18-gauge needle attached to a 1-cc tuberculin syringe. The cages containing the mice undergoing treatment were kept in a fume hood located in a room outside of the animal quarters. Before use, the hood was disinfected and made dust free by use of an air filter. In a further attempt to minimize the possibility of reinfection from outside sources, all cages were sterilized just prior to their use.

In the first experiment, 60 mice, 6 weeks old, were selected. These were divided according to sex into 6 treatment groups of 10 mice each. Each group was confined in a separate cage and after overnight fasting was given 1 through 6 mg of carbarsone daily, respectively, for 10 consecutive days. The average weight of the mice was 20 gm, so that the dosage for those given 1 mg was at the rate of 50 mg/kg (*5*). No mice were lost during treatment, and there were no visible signs of toxicity. A 7th group of 10 mice of the same age was included as untreated controls. These were not confined to the fume hood but were fed the same diet and were fasted for the same periods as the treated mice. As expected, there was loss of weight in all groups from the fasting, but this was regained soon after treatment. At intervals following treatment, one mouse from each of the 7 groups was sacrificed to determine, by repeated cecal examinations, the effectiveness of the treatment. The first checks were made 4 days post-treatment, and thereafter checks were made at 2-day intervals. In all cases, the control mice showed large numbers of flagellates, including *Giardia muris* (*3*) in about one-half, and 8 of the 10 had infections with an amoeba tentatively designated as *E. muris*. Of the mice given 1-, 2-, and 3-mg treatments of carbarsone, respectively, those killed 4–10 days after treatment showed no amoebae, but flagellates were found in decreasing numbers correlated with the increasing doses. The remainder of the mice, killed 12–22 days post-treatment, showed flagellates and/or amoebae. It was thus clear that carbarsone in these doses was not effective.

In the second experiment, the dosage of carbarsone was increased according to the recommendation of Gabaldon (*2*). Mice of the same age as above were divided according to sex into 3 groups of 10 each and received, after overnight fasting, 10, 20, and 30 mg, respectively, for 5 days. Three mice of the group treated with 20 mg and one of the group treated with 30 mg died soon after treatment. These deaths probably resulted from toxicity, but diarrhea and the nervous symptoms ascribed by Gabaldon (*1*) to such toxicity were not observed. It should be pointed out that the largest dosage used here (30 mg) was considerably less than that found by Gabaldon to be the minimal dose producing toxic symptoms in rats (4,000 mg/kg of body weight, or equivalent to 80 mg/20 gm of body weight). The 10 untreated controls showed about the same rate and intensity of protozoan infections as those above. For 20 days post-treatment, none of the treated mice showed protozoa, except for an occasional *G. muris*. However, those mice killed 22 or more days after treatment showed large numbers of *E. muris*.

While it is not possible to exclude reinfection by airborne amoebae cysts from wild mice, this seems extremely unlikely in view of the precautions mentioned above. It appears from these results, therefore, that carbarsone, as used, exerted an amoebastatic effect but did not eradicate *E. muris*. This could lead to error in interpreting results of experimental *E. histolytica* infections following such carbarsone treatment, since the active, unstained trophozoites of the two amoebae are quite similar in certain morphological details and in type of motility.

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