tion recovery offers no advantage over oral feeding as measured by long-term weight gain or mortality, it does suggest that parenteral feeding may be advantageous during the first few post-irradiation days. For this reason further work with parenteral hydrolysate administration during the early post-irradiation period is indicated. In view of the earlier work of Buchwald reporting impaired intestinal glucose absorption in the immediate post-irradiation period(3), the effect of complete parenteral feeding in the early period following radiation should be studied.

Conclusions. 1. Parenteral feeding of protein hydrolysate following radiation offers no advantage over oral feeding in the protein-depleted rat as measured by radiation susceptibility. 2. Parenteral hydrolysate administration may be advantageous in preventing early post-irradiation weight loss.

The animals were irradiated through the kind cooperation of the Biology Division, Argonne National Laboratory, Chicago, Ill.

The author wishes to express his appreciation for the aid and assistance of the following: Paul R. Cannon, Robert W. Wissler, and Randolph H. Hughes, Department of Pathology, University of Chicago and Austin M. Brues, Biology Division, Argonne National Laboratory, Chicago, Ill.

1. Meed, J. F., Decker, A. B., and Bennett, L. B., UCLA-90, 1950.

2. Martin, C. L., and Rogers, F. T., *Am. J. Roentgen.*, 1924, v11, 280.

3. Buchwald, K. W., *J. Exp. Med.*, 1931, v53, 827.

4. Barron, E. S. G., Wolkowitz, W., and Muntz, J. A., CH-3725, 1947.

5. Bennett, L. B., Chartain, S. M., Decker, A. B., and Meed, J. F., PROC. SOC. EXP. BIOL. AND MED., 1951, v77, 715.

6. Jennings, F. L., PROC. SOC. EXP. BIOL. AND MED., 1949, v72, 487.

7. Frazier, L. E., Wissler, R. W., Steffee, C. H., Woolridge, R. L., and Cannon, P. R., *J. Nutrit.*, 1947, v33, 65.

8. Cannon, P. R., Frazier, L. E., and Hughes, R., *Arch. Path.*, 1950, v50, 709.

9. Cannon, P. R., Wissler, R. W., Woolridge, R. L., and Benditt, E. P., *Ann. Surg.*, 1944, v120, 514.

10. Wissler, R. W., Woolridge, R. L., Steffee, C. H., and Cannon, P. R., *J. Immun.*, 1946, v52, 267.

11. Wissler, R. W., Steffee, C. H., Woolridge, R. L., Benditt, E. P., and Cannon, P. R., *J. Am. Diet. Assn.*, 1947, 23.

12. Bliss, C. I., *Ann. Applied Biol.*, 1935, v22, 134.

Received January 31, 1952. P.S.E.B.M., 1952, v80.

Inhibition of Growth of a Green Flagellate by the Antihistamine, β-dimethylaminoethyl (Benadryl).* (19504)

THEODORE L. JAHN AND WILLIAM DANFORTH.

From the Department of Zoology, University of California, Los Angeles.

The antihistamines and related drugs have been shown to have a variety of effects upon living systems, including growth inhibition of micro-organisms(1-5), inhibition of glutamate oxidation by rat brain tissue(6), and production of conduction block and depolarization of frog nerve(7). Of these effects, only one case of bacteriostatic activity(4) and one case of fungistatic activity(3) were reversible by histamine. Experiments in this laboratory have shown that β-dimethylaminoethyl benzhydryl ether (Benadryl hydrochloride, Parke, Davis Company) is a potent inhibitor of the growth of the green flagellate, *Chlorogonium tetragamum.*

Experiments were carried out in a modification of the medium of Hutner *et al.*(8) for fresh-water chlamydomonads, produced by reducing the concentrations of trace metals and chelating agent to one-fifth those described for the original medium. Dilution of this medium by one-sixth its volume, changes in initial pH from 5.5 to 8.0, and the addition of the metabolites and biological

* Aided by a grant from the California Cancer Institute.

materials mentioned below had no appreciable effect on the growth of *Chlorogonium* in this medium. The organisms were grown in 14 x 125 mm cotton-stoppered test tubes, and incubated at 25°C with constant fluorescent illumination. Populations were estimated by Tyndall-beam nephelometry† at approximately 3-hour intervals during the logarithmic growth period (about 12 hours) and at later times when necessary. Cultures were routinely plated on bacteriological media at the end of each experiment, and data from contaminated tubes were discarded.

Results. Benadryl‡ in concentrations of 1.7×10^{-5} M to 8.6×10^{-4} M causes a graded inhibition of the growth rate of *Chlorogonium* from almost normal growth at the lower concentration to almost complete inhibition at the higher. Concentrations in this range are not lethal, since subcultures into Benadryl-free medium after 24 hours exposure to Benadryl regularly showed normal growth. Cells maintained in 8.6×10^{-4} M Benadryl were normal in appearance, motility, and phototropic behavior after 24 hours. Higher concentrations of Benadryl, of the order of 10^{-3} M, caused actual death of the cells within a few hours.

The growth inhibitory effect of Benadryl is extremely sensitive to pH, as its action in causing nerve block(7). No inhibition is produced at pH values below ca. 6.8. Since the pH of growing *Chlorogonium* cultures rises continuously, cultures containing Benadryl and having an initial pH below this critical level grow normally for a period of time inversely related to the initial pH, after which time they show marked inhibition (Fig. 1). This dependence on pH seemingly indicates that only the free-base form of Benadryl is inhibitory. Whether this is due to differences in the pharmacological activities or in the permeability of the cell membrane to the two forms is not clear. Since many of the materials added in the experiments described below tended to acidify and/

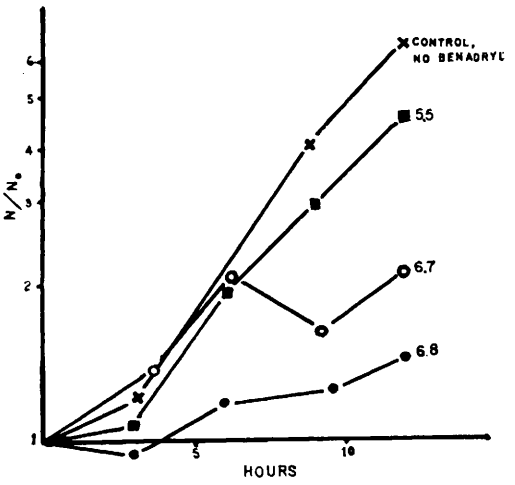

FIG. 1. Relation of Benadryl inhibition to pH. Benadryl concentration (except in control), 8.6×10^{-4} M. Points represent averages of 3 replicate tubes. Figures at ends of curves indicate initial pH. N equals population at time indicated on abscissa; N_0 equals initial population.

or buffer the medium, great care was required to prevent pH effects from interfering with the results.

Attempts were made to reverse the Benadryl inhibition by addition of various substances to the medium. Histamine in a molar ratio to Benadryl of 8:1 had no effect. Thiamin and nicotinamide in combination have been reported to reverse the inhibition of bacterial growth by another antihistamine(1), but these compounds, in molar ratios to Benadryl of 11 and 14 respectively, had no effect on the inhibition, whether added alone or in combination. The inhibition of glutamate oxidation in brain(6) and accumulation of pyruvic acid in bacterial cultures(4) caused by antihistamines suggested that these compounds might affect the Krebs cycle, but neither malic nor α-ketoglutaric acid (molar ratios of 15) nor a mixture of lactic, pyruvic, malic, α-ketoglutaric, succinic, fumaric, and citric acids (molar ratios ca. 2.5) reversed the inhibition. Neither acid-hydrolysed casein (Nutritional Biochemicals Co., "vitamin-free") nor yeast extract (Difco) (in 1% concentrations) was effective.

In a few experiments the growth-inhibitory activity of 4,4-diphenyl-N,N-dimethylbutyl-1-

† Unpublished technic developed by Roger W. Hanson.

‡ This compound and several others were generously supplied by Dr. Frederick Crescitelli.

amine hydrochloride,§ closely related structurally to Benadryl, was tested. But this compound possessed about one-fifth the antihistaminic activity of Benadryl. This compound was found to inhibit growth in a manner similar to Benadryl, but appeared to be even more potent than Benadryl itself, causing death of a large proportion of the cells in 24 hours at a concentration of 4.6 x 10^{-4} M, and producing total inhibition of growth at 2.3 x 10^{-4} M.

The lack of activity of histamine in reversing the Benadryl inhibition of *Chlorogonium* growth, and the lack of correlation between antihistaminic activity and growth-inhibitory activity in structurally related compounds appear to indicate that the growth inhibition is not related to the antihistaminic properties of the drug. The effect of Benadryl on *Chlorogonium* appears to differ from the bacteriostatic effect of antihistamines reported by Krecek *et al.*(4) and from the fungistatic effect reported by Landis and Krop(3), which were reversed by histamine, and from the bacteriostatic effect reported by De Ritis and Zanussi(1), which was reversed by a thiamin-nicotinamide mixture. Reiss and Caroline(5) have reported fungistatic effects of antihistamines which were not reversed by histamine, nor did histamine reverse the effects of Benadryl on vertebrate nervous tissue(6,7). Thus, it seems likely that the antihistaminic drugs may produce their effects by several different mechanisms.

§ Synthesized by Dr. T. A. Geissman, Chemistry Department, U.C.L.A., and tested for antihistaminic activity (guinea pig aerosol technic) by Dr. E. J. Fellows, Smith, Kline and French Laboratories, Philadelphia.

1. De Ritis, F., and Zanussi, C., *Riv. Ist. sieroter. ital.*, 1949, v24, 10.

2. Carson, L. E., and Campbell, C. C., *Science*, 1950, v111, 689.

3. Landis, L., and Krop, S., PROC. SOC. EXP. BIOL. AND MED., 1951, v76, 538.

4. Krecek, J., Sterzl, J., Kreckova, J., and Vaicenbacher, V., *Casopis lekaru ceskych*, Prague, 1950, v89, 2. Sterzl, J., and Krecek, J., *Casopis lekaru ceskych*, Prague, 1950, v89, 35. (From abstracts, *Excerpta Medica*, Sect. 2, 1950, v3, 1570.)

5. Reiss, F., and Caroline, L., *Science*, 1951, v114, 15.

6. Carlisle, E. M., and Crescitelli, F., *Science*, 1950, v112, 272.

7. Crescitelli, F., and Geissman, T. A., *Am. J. Physiol.*, 1951, v164, 509.

8. Hutner, S. H., Provasoli, L., Schatz, A., and Haskins, C. P., *Proc. Am. Phil. Soc.*, 1950, v94, 152.

Received February 13, 1952. P.S.E.B.M., 1952, v80.

Effects of Acute Radiation on the Adult Mammalian Central Nervous System.* (19505)

SAMUEL P. HICKS AND PHILIP O'B. MONTGOMERY. (Introduced by Shields Warren.)

From the Cancer Research Institute, New England Deaconess Hospital, and the Laboratories of Pathology, Harvard Cancer Commission and Harvard Medical School, Boston.

A study in progress in this laboratory has been directed to characterize the changing metabolism of the nervous system as it develops from embryo to adulthood and to relate this to its changing radiosensitivity. Data on neurectoderm, neuroblasts and neonatal nerve cells have been reported elsewhere(1-3). In general the adult nervous system is considered to be radio-resistant, but whether it is uniformly so has not been determined.

In order to learn whether a differential of radiosensitivity exists within the adult nervous system, rats and mice were irradiated either to the head or to the whole body and the acute histopathologic changes that followed were recorded. The radiation given ranged from 50 to 20,000 r.

Materials and methods. Adult rats and

* This work was done under U. S. A. E. C. Contracts, AT(30-1)901 and AT(30-1)699, U. S. P. H. S. Grant C-1042 and a Grant from the United Cerebral Palsy Associations.