mine whether they can reach, and maintain, normal mature weights on this combination.

Rats suffering from dermatitis were also supplied with the flavine preparations, but these did not retard the course of the disease in the slightest degree. As mentioned previously, wheat-germ oil heals the dermatitis. The animals gained slowly for several weeks and then declined, with no recurrences of dermatitis. These animals did not become denuded, an anomaly that is hard to explain. If, in addition to the oil, these animals are also given 1 drop daily of the flavine preparation they gain in weight and assume a normal appearance. It is too early as yet to determine whether they can reach mature weight, or whether the maximum weight can be sustained. Additional details are supplied in Fig. 1.

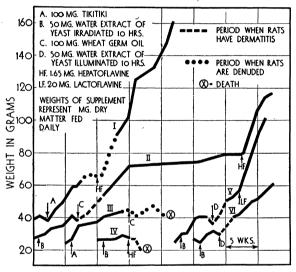


FIG. 1. I. Rats become denuded on tikitiki, and are healed by flavines. II. Dermatitis is healed by wheat germ oil, but growth does not occur unless flavines are added also. III. Wheat germ oil has no effect on denuding. IV. Flavines have no effect on dermatitis. V and VI. Illuminated vitamin B carriers heal dermatitis, but flavines in addition must be added to support normal growth.

Evidently the two conditions have little or no relation to each other. Wheat-germ oil heals dermatitis but does not relieve the denuded condition. Flavine heals the denuded condition but does not relieve the dermatitis. These facts show clearly that ultra-violet irradiation destroys at least two vitamins, the flavine, and the anti-dermatitis factor which has not been identified as yet. It is possible that still others are destroyed also.

It has been reported previously<sup>5</sup> that the anti-

<sup>5</sup> A. G. Hogan and L. R. Richardson, Jour. Nutrition, 8: 385, 1934.

dermatitis factor is not destroyed by irradiation if the ultra-violet portion of the spectrum is excluded. Since flavines are labile to the visible spectrum, additional observations on this point seemed desirable. Six rats were each supplied with 50 mg daily of the water extract of yeast that had been illuminated through plate glass for 10 hours at a distance of 10 inches with a 1,500 watt Mazda bulb. They made only slight gains in weight, but there was not a single well-defined case of dermatitis. The controls received 50 mg of the yeast extract that had been subjected to ultra-violet irradiation, and they developed dermatitis and died in the usual time. Ten other rats received the irradiated preparation until they developed definite cases of dermatitis; then they were changed to the preparation which had been illuminated with the Mazda bulb, as described above. Every animal recovered from dermatitis, but the gains in weight were slight. When the illuminated material was fortified with 1 drop daily of the flavine preparation growth was resumed, but it is too early to decide whether this combination is complete in every respect. Some additional details are shown in Fig. 1.

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## WHY HAVE SOME INVESTIGATORS BEEN UNABLE TO GROW CHILOMONAS PARA-MECIUM IN INORGANIC OR SIMPLE ORGANIC SOLUTIONS?

PRINGSHEIM<sup>1</sup> maintains that Chilomonas paramecium will grow in sterile inorganic solutions containing glycocoll and acetate, but not if the nitrogen and carbon containing compounds are less complex. Mast and Pace<sup>2</sup> conclude that it will grow if these two elements are in the form of inorganic or simple organic compounds, i.e., they maintain that it can obtain nitrogen from ammonium salts and carbon either from acetates, formates or carbon dioxide. Pringsheim<sup>3</sup> was unable to confirm these conclusions concerning the utilization of CO<sub>2</sub>; and Loefer<sup>4</sup> was unable to confirm the conclusions concerning the utilization of CO<sub>2</sub> or acetates. He says: "It was impossible to maintain bacteria-free cultures of Chilomonas paramecium-in a medium containing inorganic nitrogen, even in the presence of sodium acetate as a carbon source." Loefer and Hall<sup>5</sup> repeated the experiments of Loefer, using some of the media of Mast and Pace and their technique as well as that developed by

<sup>1</sup> E. G. Pringsheim, Beitr. z. allg. Bot., Bd. 2, S. 88-137, 1921.

<sup>2</sup> S. O. Mast and D. M. Pace, Amer. Jour. Physiol., 101: 75, 1932; Anat. Rec., 54: 101-102, 1932; Protoplasma, 20: 326-358, 1933.

<sup>8</sup> E. G. Pringsheim, Naturwiss., Bd. 23, S. 110-114, 1935.

4 J. B. Loefer, Biol. Bull., 66: 1-6, 1934.

<sup>5</sup> J. B. Loefer and R. P. Hall, SCIENCE, 81: 486, 1935.

Loefer, but they obtained growth for only a few days. They conclude: "It would seem, therefore, that our strain of Chilomonas paramecium is unable to synthesize protoplasm from ammonium compounds and other inorganic salts and is thus quite different in this respect from the strain used by Mast and Pace." Loefer<sup>6</sup> concludes that their strain can not even "utilize as a source of nitrogen any of the single amino-acids tested," i.e., "glycine, dl-valine, l-leucine, dl-leucine, dl-iso-valine, dl-\beta-phenylalanine, l-tyrosine and the compound asparagin."

At our request Loefer and Hall very generously sent us a sample of their strain of Chilomonas. This sample contained bacteria when it arrived, having, unfortunately, been contaminated en route. Loefer and Hall had for two or three years grown their strain in a bacteria-free solution containing tryptone.

We added to this solution an equal quantity of our solution D, *i.e.*, a solution containing nitrogen in the form of ammonium chloride and carbon in the form of acetate and left it 12 hours; then several specimens were removed with .1 cc of the solution and added to .3 cc of solution D and left 12 hours, after which single individuals were removed, passed through several portions of fresh solution D and cultured on depression slides in accord with the method described by us.7

Four lines of isolation cultures were thus established and carried for six weeks with daily transfers. During this time the average rate of fission was 3.06 per day, *i.e.*, it was practically the same as that obtained in our earlier experiments with our strain of Chilomonas. None of the lines died out during the experiment and at the close the specimens in all were in excellent condition and indistinguishable from those in our strain cultured in solution D. There is therefore no reason for assuming that the two strains in question differ. We have grown in the acetate-ammonium solution chilomonads collected at Woods Hole, Mass., Baltimore, Md., Durham, N. C., and Birmingham, Ala. It is therefore not probable that there are different strains of Chilomonas in reference to ability to obtain nitrogen and carbon from simple compounds.

We have repeatedly observed that if chilomonads are transferred from a glucose-peptone solution directly to an acetate-ammonium solution or from this solution directly to this solution minus acetate nearly all die immediately, and that those which do not die immediately divide infrequently and usually die after a few days. We have also repeatedly observed that if the concentration of the acetate is too low there is frequent division for a few days during which the chilomonads become smaller and smaller until they die. The failure of Loefer<sup>8</sup> and Loefer and Hall<sup>9</sup> to obtain growth in our inorganic solution (solution I) or our acetate-ammonium solution (solution D), and Loefer<sup>10</sup> to obtain growth in solutions containing but one amino-acid was therefore probably due to insufficient care in transferring the chilomonads from their tryptone solution to the solutions containing ammonium chloride and acetate or a single amino-acid, or to unsatisfactory concentrations of acetate.

Pringsheim presents no details concerning the methods used in his attempt to grow Chilomonas in inorganic solutions. We are therefore unable to offer any suggestions concerning the cause of his failure.

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## SCIENTIFIC APPARATUS AND LABORATORY METHODS

## PARADICHLOROBENZENE AS A HER-BARIUM INSECTICIDE

THE author read with interest an article recently published in this journal entitled, "Paradichlorobenzene, an Effective Herbarium Insecticide."1 The gratifying results obtained by the author of this article are similar to the results experienced by many of us who have been using this herbarium insecticide for the past several years. The use of paradichlorobenzene in herbariums goes back several years. It was used in a part of the National Herbarium prior to 1930, and also in many other herbariums prior to that date. The use of this insecticide has become so popular that the old method of periodic fumigation with

hydrogen cyanide or carbon bisulfide is almost obsolete.

Even though good results were obtained by the author of the above-mentioned article by placing the crystals of paradichlorobenzene on the bottom of the herbarium cases, it is considered to be better practice to place the crystals on the top shelves of the cases. The fumes of the insecticide are heavier than air, and thus by placing the crystals near the top better distribution of the insecticidal gas is obtained. When the crystals are placed at the bottom, distribution of the heavier-than-air gas depends upon convection currents.

P. F. Shope

<sup>&</sup>lt;sup>6</sup> J. B. Loefer, Arch. f. Protist., Bd. 85, S. 74-86, 1935. <sup>7</sup> Protoplasma, 20: 326-358, 1933.

<sup>&</sup>lt;sup>1</sup> Frank C. Gates, SCIENCE, 81: 438, 1935.

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<sup>8</sup> Biol. Bull., 66: 1-6, 1934.

<sup>&</sup>lt;sup>9</sup> SCIENCE, 81: 486, 1935. <sup>10</sup> Arch. f. Protist., Bd. 85, S. 74-86, 1935.