of phenocopy. In all, 558 flies of two stocks, an isogenic wild type and the mutant aristapedia-Bridges, were irradiated. No difference in the response of the two stocks was observed. The results indicate the existence of two periods of high sensitivity to the treatment, one 70 hours after hatching and one 100 hours after hatching, just before pupation. Larvae

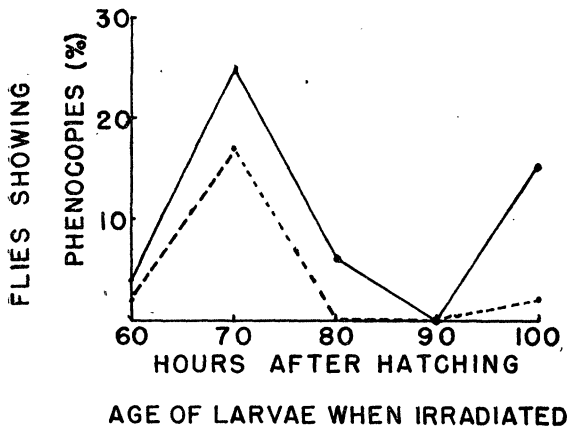

FIG. 2. Percentage of flies showing phenocopies after exposure of the larvae to light in cyanin solution. Dotted line = 30-minute exposure, solid line = 60-minute exposure.

show a gradual increase in sensitivity to ultraviolet radiation with increasing age from 50 to 100 hours after hatching (*10*) but only one period of maximum sensitivity, just before pupation.

The phenocopies produced in these experiments may be interpreted as due to the killing of individual cells or groups of cells in the larvae at the time of the exposure, which results in the failure of some part to form. More extensive damage results in the death of the larva. The larvae which pupated and then died were dissected open. These animals showed patches of black, necrotic tissue on the abdomen which resembled burns. No phenocopies resembling the pervasive changes in morphogenesis found as a result of X-radiation (*9, 11*) were produced by these treatments. Photodynamic dyes are believed (*1*) to be activated by a quantum of light and to transfer this activation to a molecule of some substrate substance in the cell which then reacts with oxygen. Since the oxidation of this substrate molecule is incompatible with life, the cell dies.

These experiments demonstrate the effectiveness of cyanin as a photodynamic dye in the production of phenocopies and suggest its use in other biological systems.

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A Qualitative Test for Penicillin

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As the list of antibiotic substances lengthens, the problem of determining whether or not a new one has been obtained becomes increasingly difficult. In a survey of new sources of antibiotic substances, the problem is further complicated by the need to identify the active substance in highly impure preparations. The fact that penicillin is widely distributed, very active, and produced in satisfactory quantities by fungi already in cultivation makes it highly desirable to determine before the solution is studied further that its activity is not due to penicillin. An enzyme from a culture of *Staphylococcus aureus*, which did not affect the activity of aspergillic acid, gliotoxin, and gramicidin but did inactivate penicillin, was used to indicate that one of the antibiotic agents formed by *Aspergillus flavus* was a penicillin (*2*).

A specific qualitative test for penicillin is available if it can be shown that penicillinase inactivates only penicillin and does not affect other antibacterial substances. Penicillinase in the form of Clarase Lot No. 1351 and Schenley Penicillinase are used in chemical (*6*) methods for the assay of penicillin concentrates and in sterility tests of penicillin preparations (*1, 4, 5*).

It was found in this laboratory that penicillinase (Clarase Lot No. 1351 or Schenley Penicillinase A) did not affect the antibacterial activity of the following substances: aspergillic acid, citrinin, gliotoxin, patulin, penicillic acid, spinulosin, streptomycin, streptothricin, the active substances formed by eight members of the higher Basidiomycetes, and a crystalline compound isolated from a green plant. The activity of one of the basidiomycete culture solutions was not affected by the Schenley Penicillinase A but was decreased by the Clarase, presumably as the result of some of the many other enzymes in it. Hence, only the Penicillinase A could be used to identify penicillin. It is possible that antibacterial agents other than penicillin will be found that are inactivated by penicillinase; but until they have been shown to occur, the failure to obtain inactivation by penicillinase may safely be taken to indicate the absence of penicillin.

The activities of the antibacterial substances were measured in serial dilution tests (*3*) set up with and without the inactivating enzyme in the broth. The test bacterium was the Heatley strain of *Staph. aureus*. Clarase at a concentration of 1 mg./ml. of broth inactivated 50 μg. of penicillin G in this test—that is, reduced the effective concentration of penicillin to less than 0.06 μg./ml. One Schenley unit of penicillinase/ml. of broth inactivated 200 μg. of penicillin G. The Clarase and Schenley Penicillinase A were used at concentrations sufficient to inactivate penicillin solutions with activities from 10 to 100 times as great as that of the antibacterial substances which were not affected by the enzyme.

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