more difficult to prepare and decipher, while a neatly typed label on an ordinary white filing card is more readable and much easier to prepare.

The powdered acrylic resin is given an initial pressure of nearly 250 pounds. The resin is then heated to 125° C. When this temperature is attained, a pressure of 4,000 pounds per square inch (1-inch mold) is applied before removing the heater. This final pressure is maintained throughout the cooling period. The briquet is driven out at 75° rather than at 80° C.

Opaque, white, cloudy spots usually form if the older directions are followed with the label insert. The spots result from the retention of the inhibitor, which is readily driven off in spite of the label providing no pressure is maintained during the heating period.

Ultraviolet Radiation as a Means of Sterilizing Tissue Culture Materials[1]

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The efficacy of ultraviolet radiation of wave length 2,537 A. in killing bacteria, fungi, and viruses has been adequately demonstrated (*1*). It can be used, therefore, under certain conditions as a substitute for heat sterilization. If it is to be effective, however, certain requirements and limitations must not be overlooked. For sterilizing solutions containers of clear, transparent, fused quartz or of special glass transmitting wave length 2,537 A. must be used, because pyrex, plastics, and ordinary glass absorb this wave length. The surfaces of these containers and of dissecting instruments, slides, and cover glasses to be sterilized must be free of any extraneous matter that will absorb ultraviolet, and must be arranged in such a way in relation to the radiation source that all parts requiring sterilization are exposed to the radiation. Solutions must not contain in high concentration constituents that absorb appreciable amounts of ultraviolet radiation of this wave length, e.g. many organic substances—particularly proteins and sterols—as well as certain inorganic compounds. Solutions in which these substances are present in low concentration can be sterilized by rotating the flask in such a way that all parts of the solution receive similar amounts of ultraviolet energy. Spectrographic analysis of our three concentrated stock solutions[2] fails to indicate any significant absorption of wave length 2,537 A. to a depth of 1 cm.[3]

[1] This work was supported in part by a grant from the University Research Committee of the University of Alabama.

[2] Solution A: 7 per cent NaCl, 0.2 per cent KCl, 0.2 per cent $CaCl_2$, 0.1 per cent $MgCl_2$, 0.2 per cent NaH_2PO_4; Solution B: 0.12 per cent $NaHCO_3$; Solution C: 8 per cent dextrose. These constituents, if combined in a single concentrated stock solution, will undergo chemical reaction and precipitation after a short storage period. If kept in three separate solutions, however, and stored in a refrigerator, they will remain usable for a long period. Our dilute, ready-to-use solution consists of 1 part of each of Solutions A, B, and C to 7 parts of water.

[3] We are indebted to Mrs. Dorothy C. Peterson, of the National Institute of Health, for this analysis.

In sterilizing physiological salt solutions, ultraviolet radiation has a number of advantages over heat sterilization. First, there is no loss of water through evaporation and no consequent increase in osmotic pressure. Second, heat-induced chemical reactions between the constituents of the different stock solutions, viz., certain inorganic salts and dextrose, are

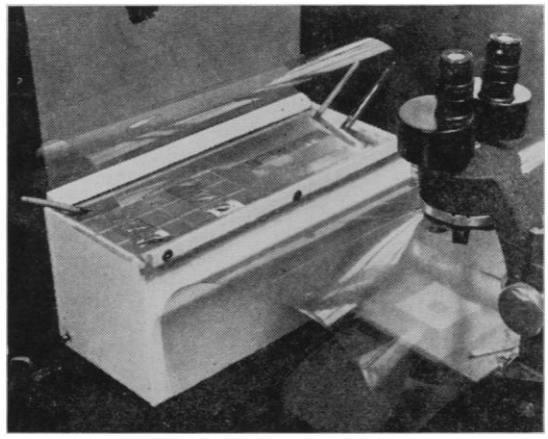

Fig. 1. Exterior of sterilization box, showing dissecting instruments, cover glasses, and slides in position for sterilization.

eliminated; therefore, sterilization can follow mixing and dilution. This dilute solution will remain stable at room temperature for at least several weeks if sterilized with ultraviolet after each opening of the container. Third, unsterilized pipettes

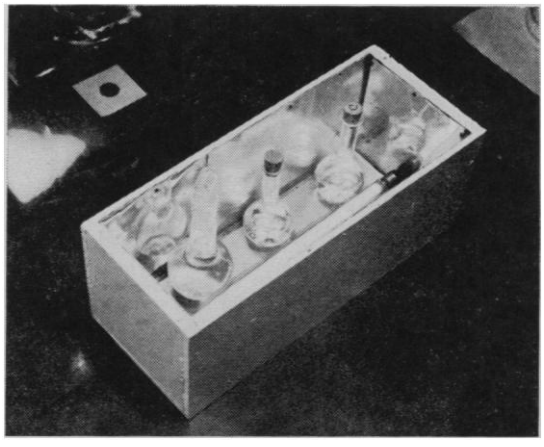

Fig. 2. Interior of sterilization box, showing aluminum lining, ultraviolet lamp, and solutions in quartz flasks.

can be used in making the dilution, for asepsis can be accomplished quickly by subsequent irradiation of the final solution, and the stock solutions can be readily resterilized with ultraviolet after opening. Fourth, the whole method is simpler and less time consuming than autoclaving, irradiation for about 5 minutes being sufficient. It must be remembered, however, that excessive exposure of some substances to ultraviolet produces chemical changes.

Our device is shown in Fig. 1. The transparent shield that covers the box containing the ultraviolet lamp protects the

eyes from direct and scattered radiation. It is a sheet of unexposed, processed X-ray film, which absorbs practically all the 2,537 A. ultraviolet striking it. The aluminum cover of the box has openings for dissecting instruments, cover glasses, and slides. The dissecting instruments are kept sterile by exposure to radiation whenever they are not in use. A few minutes before a set of preparations is to be made, cover glasses and depression slides are inverted over the larger openings.

The interior of the box is shown in Fig. 2. The ultraviolet source is an 8-watt, commercial, low-pressure, mercury vapor lamp, known by the trade name "germicidal lamp" or "sterilamp," with lamp holders, ballast, and starter. The box is lined with aluminum, which is a particularly good reflector of ultraviolet and insures irradiation of all sides of the instruments and of the mouths of cork-stoppered flasks. We have found it necessary to insert a piece of cellophane under the cork stoppers to prevent mold growth on their lower surfaces. Further, since many fused quartz flasks do not have perfectly round mouths and therefore seal with difficulty, the projecting edges of the cellophane help guard against the entrance of contaminants.

Every precaution should be taken to prevent exposure of the eyes of the worker or others in the room to direct or reflected radiation from the lamp. Failure to observe this may result in severe temporary pain commencing several hours after exposure. Unexposed, processed X-ray film, pieces of window glass, and most opaque materials of appreciable thickness are effective absorbers of the harmful radiation.

Reference

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The Production of Phenocopies in *Drosophila* Using Visible Light and a Photodynamic Dye

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Phenocopies, changes in development produced experimentally which mimic the phenotypes of mutant genes, have been produced by high temperatures (*5, 8*), low temperatures (*6*), X-rays (*3, 9, 11*), chemical agents (*7*), and ultraviolet rays (*2, 4, 10*). The present study was undertaken to see whether phenocopies could be produced by visible light and a photodynamic dye and to explore the possibility of using cyanin as a photodynamic dye. The dyes used previously by other experimenters have the disadvantage of being difficult to remove from the organism treated after their use so that the photodynamic action may continue. Cyanin (quinoline blue) is an indicator dye that is colorless at pH 7 and blue at pH 8, which permits its photodynamic action to be controlled by adjustment of the pH.

Drosophila larvae of known ages were obtained by allowing large numbers of flies to lay eggs for a two-hour period on corn meal-molasses-agar food in half-pint bottles. The cultures were kept at 25.5° C. before and after irradiation. For irradiation, the larvae were placed in small Petri dishes, covered with 0.04 per cent cyanin solution adjusted to pH 8, and exposed to the light from a 100-watt projection lamp at a distance of 10 inches. After irradiation the cyanin solution was made colorless by the addition of a few drops of 0.1N HCl, and the larvae removed, washed twice in Ringer's solution, and placed in shell vials in the incubator to complete development. The cyanin solution, made by grinding 0.1 gram of crystalline cyanin (Grübler) in a mortar with 20 cc. of 0.01N NaOH and diluting to 250 cc. with distilled water, is not very stable and gradually loses its blue color.

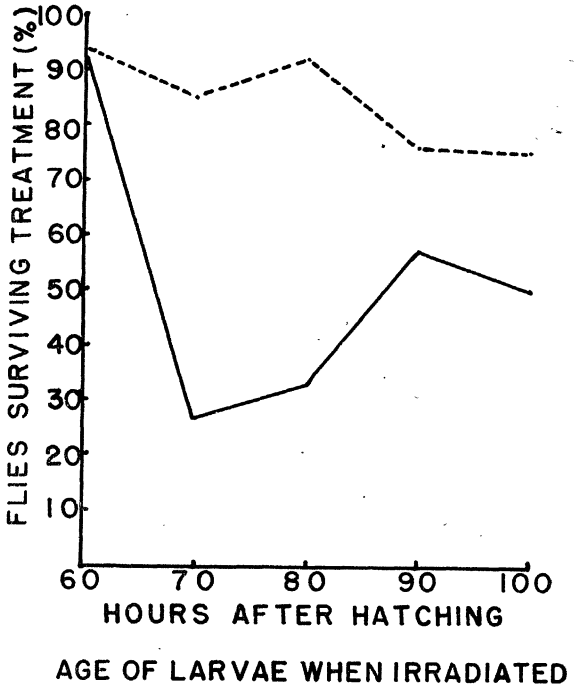

FIG. 1. Percentage of flies hatching from larvae exposed to light in cyanin solution. Dotted line = 30-minute exposure, solid line = 60-minute exposure.

Larvae 60–100 hours old (after hatching from the egg) were exposed for 30 or 60 minutes to the beam of light. The absolute intensity of the light is not known, but the constancy of its intensity throughout the experiment was checked by a photocell and milliammeter. As a result of the light a certain percentage of the larvae were killed in each experiment (Fig. 1); the larvae usually died within 12 hours of the irradiation, although some died after pupation. Other larvae, placed in the cyanin solution and subjected to the same changes in pH and washings as the irradiated flies, but not irradiated, showed no mortality. A second effect of the light was the production of a certain number of phenocopies: abnormal abdomen, dark eye color, bent or missing bristles, and folded wings (Fig. 2). The various types of phenocopies occurred at random throughout the period tested; no indication was found that an irradiation at a particular stage tended to produce a particular kind