

be concerned, as is neonatal, with the child, and postpartum with the mother.

HARRY E. HANDLEY

*The Commonwealth Fund
New York City*

A Chamber for Observations on Living Larvae of Anopheline Mosquitoes

IN CONDUCTING certain physiological and toxicological studies on the first two instars of *Anopheles quadrimaculatus* Say, the larvae had to be kept relatively quiescent and observed with a minimum of disturbance for long periods of time under high magnification. In addition, in some experiments, control of the temperature was important.

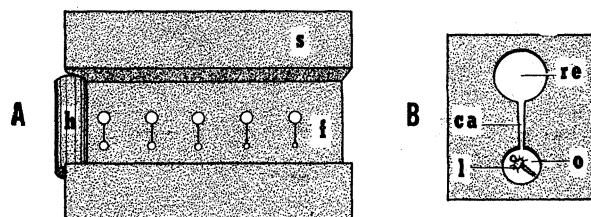


FIG. 1. A, aluminum insert (top view) which fits into an electric microscope stage incubator. B, enlargement of a portion of the floor of the insert. *f*, floor of insert; *h*, case around thermometer of stage incubator; *s*, side arm supports of insert; *o*, observation cells; *ca*, connecting canal; *re*, reservoir; *l*, larva.

The problem was solved by the design of a simple chamber in which larvae are suspended in hanging drops of water or other test fluids. The chamber, shown in Fig. 1, is formed of two parts: (1) A metal insert that fits snugly into (2) an ordinary electric microscope stage incubator (manufactured by the Fisher Scientific Company). The insert (A), fashioned from an aluminum sheet 1 mm thick, is 75 × 55 × 12 mm. One portion of the insert (*h*) encases the thermometer bulb of the Fisher stage incubator. A series of holes, 0.5–3.5 mm in diameter, is bored through the floor (*f*) of the insert to serve as observational cells (*o*) for larvae. Each of these is connected by a small canal (*ca*) (0.5 mm wide) to a hole 3 mm in diameter, which serves as a reservoir (*re*) for water or other test substances.

An ordinary medicine dropper with a narrowed tip is used to place larvae in the observation cell (*o*) and to fill the reservoir and the canal. Larvae tend to remain remarkably quiet within the hanging drops in the open observation cells. This arrangement permits detailed observations on intact organs of the larvae at magnifications of 400 diameters.

To prevent quick changes in temperature in studies where this is a critical factor, it is necessary to cover the assembled chamber. A plexiglass sheet was found suitable. Since this sheet tends to fog, it is necessary in some experiments to make a small opening in it in order to observe the larvae. When the cover is in use, larvae can be observed at 75 diameters with a dissecting microscope, but for higher magnifications with

the compound microscope it is necessary to remove it.

This chamber, suitably modified if necessary, might profitably be used to study a number of problems pertaining to mosquito larvae and possibly to a number of other aquatic organisms.

JACK COLVARD JONES

*Laboratory of Tropical Diseases
National Microbiological Institute
National Institutes of Health, USPHS
Bethesda, Maryland*

The Biological Activity of Mixtures of Lycomarasmin and Glutamic Acid, Glutamine Glutathione, or Cysteine

LYCOMARASMIN, isolated from culture filtrates of *Fusarium oxysporum* f. *lycopersici* by Clauson-Kaas, Plattner, and Gaumann (1), has been postulated to be an active toxin in *Fusarium* wilt of tomatoes. It is a dipeptide, and its structure has been determined by Woolley as N-(α -(α -hydroxypropionic acid))-glycylasparagine (2).

Strepogenin, a peptide of glutamic acid, augments the growth of *Lactobacillus casei*, as also do glutamine and glutathione (3). Because strepogenin reduces the toxicity of lycomarasmin to tomato cuttings, and lycomarasmin reduces the activity of strepogenin on *L. casei* (3), Woolley has suggested that lycomarasmin may be an inhibitory analogue of strepogenin (3, 4). On this basis one would expect glutamine and glutathione to reduce the toxicity of lycomarasmin to tomato cuttings; in fact, Albert has stated that the high cost of glutathione is all that prevents its use to control *Fusarium* wilt of tomatoes, said to be caused by the analogous polypeptide lycomarasmin (5).

Another suggestion for a lycomarasmin antidote comes from studies of the effect of patulin on living cells by Miescher (6). Miescher has pointed out that patulin, being inactivated by compounds containing free SH groups, probably reacts with essential metabolites containing SH groups and with SH-containing enzymes. Patulin affects cells somewhat similarly to lycomarasmin (7). Glutathione is an essential metabolite in many cells and, if lycomarasmin should be toxic because of its reactivity with SH groups, it will be inactivated by both glutathione and cysteine.

Either of these hypotheses, if correct, might lead to control of any portion of the syndrome of *Fusarium* wilt of tomatoes, which is caused by lycomarasmin; therefore, the authors investigated the ability of glutamic acid, glutamine, glutathione, and cysteine to reduce the toxicity to tomato cuttings of lycomarasmin in the presence of iron, which potentiates the toxicity of lycomarasmin (1).

Lycomarasmin¹ was mixed with ferrous or ferric sulfate and with water, glutamic acid, glutamine, glutathione, or cysteine. The concentration of each component of the mixture was 0.001 M. A small tomato

¹ Crystalline lycomarasmin was obtained through the kindness of Ernst Gaumann.