THE COLONIZATION OF ANOPHELES SUBPICTUS GRASSI

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INTRODUCTION. Behavioral, physiological and vector relation studies on mosquitoes are frequently dependent upon the establishment of laboratory colonies. Since projected activities of the University of Maryland Institute of International Medicine include re-evaluation of the malaria vector potential of common anopheline species of Pakistan, it was desirable to establish colonies of as many of these species as possible.

Ward and Kitzmiller (1963) prepared a world-wide list of the laboratory colonies of anophelines currently being maintained. Anopheles subpictus Grassi was not included in this list. Roy (1940) stated that while Anopheles stephensi and Anopheles annularis would mate readily in cages measuring 2 ft. x 1 ft. x 1 ft., Anopheles subpictus would not. The paper which follows describes the colonization and methods used for the maintenance of this species.

MATERIALS AND METHODS. Initial colonization was carried out at the Institute's Pakistan Medical Research Center in Lahore, West Pakistan. The insectary was without windows, had one door, and measured 16 ft. in width, 18 ft. in length, and 10½ ft. in height. A humidifier powered by an electric motor was suspended from the ceiling in the center of the room. Lighting was provided by four double 400 watt fluorescent tubes plus four 150 watt incandescent bulbs. There were 14 hours of full “daylight,” during which time all lights were in use. At the end of this 14-hour period, the fluorescent tubes were turned off, leaving only the incandescent bulbs, which were then dimmed gradually over a period of 80 minutes by use of a powerstat transformer until there was total darkness. The process was then reversed in the morning. Insectary temperature varied during the year between approximately 80° and 90° F., while the humidity ranged between 70 and 80 percent. Temperature during the summer months was determined by the ambient building temperature, while an electric heater was used during the winter months.

Routine cattle-biting collections made at Shahzada village south of the city of Lahore during the month of May, 1965 yielded a predominance of Anopheles subpictus. An examination of the surrounding area revealed a pool containing immature stages of this species. The pool was approximately 30 feet in diameter, and was filled by seepage from a nearby well. Several larval collections made from this source were brought to the insectary. Larvae were placed in enamel pans and fed a 1:1 mixture of ground dog food and a breakfast cereal (Farex) until pupation occurred. Pupae were picked daily and allowed to emerge in a cubical cage having 2 ft. sides. The adults were fed sugar-water and raisins, and a shaved rabbit was offered nightly. The oviposition site consisted of an enamel pan measuring 12 x 7 x 1½ inches, the bottom of which was lined with wet filter paper. Eggs were hatched daily in pans of tap water, and were contained by wax paper floats which prevented stranding on the sides of the pan. Pupae developing from these eggs were subsequently introduced.

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into the same cage with the surviving parental stock.

Results. Adult mortality of the P₂ generation was extremely high and manipulation of the humidity by draping the cage with wet towels failed to reduce the death rate. Introduction of large numbers of pupae to compensate for this mortality became necessary. During the daylight period, adults were inactive but at the start of the crepuscular period, flight activity began and became more pronounced as the dimming progressed. Copulation was observed on several occasions midway through the twilight period.

Egg production was small at first but increased gradually (Table 1). Actual hatching rates were not determined but pupal production increased steadily with the exception of the 61–80 day period. Investigation at this time revealed that the lights were not being dimmed properly which may account for reduced egg fertility and subsequent loss in yield of pupae.

Discussion and Conclusions. Since Roy (1940) did not discuss in detail the insectary conditions under which he carried out his attempts at colonization, it is difficult to suggest factors which may have contributed to his failure. Barnett and Gould (1962) stated that the prospects for colonization are enhanced if very large numbers are used initially, on the assumption that this promotes the occurrence of selection processes favoring the perpetuation of those individuals best adapted to the environmental conditions. Haeger (1958) demonstrated that proper lighting conditions and crowding were two of the points of importance contributing to the colonization of Aedes taeniorhynchus. He believed that crowding adults increases flight activity which in turn stimulates mating activity.

With these considerations in mind, large numbers (average of 1000 per day for 30 days) of pupae were introduced into the colony cage to provide both crowding and maximal opportunity for selection of adaptable individuals. As mentioned previously, adult mortality was extremely high at first, and there were probably never more than 2,000 to 3,000 adults in the cage at any one time. Since copulation was observed during the crepuscular period, it is obvious that lighting was also an important factor.

At the time of writing, this colony had been vigorous and self-sustaining for over six months. A sub-colony has been established at the laboratories of the Institute of International Medicine in Baltimore.

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References


