DEVELOPMENT OF A NEW DESIGN OF AN INSECTARY MODEL FOR REARING AND ENVIRONMENTAL ASSESSMENT STUDIES ON CHIRONOMID MIDGES

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Chironomus ramosus Chaudhuri (1992), is one of the common tropical midges that form an abundant group of sediment-dwelling macro invertebrates of slow-flowing freshwater ecosystems of the Indian subcontinent, vulnerable to numerous fluctuating environmental conditions. Midges not only play an important role in maintaining a balanced aquatic ecosystem, being a major prey for the invertebrate and vertebrate fauna and preying upon lower invertebrates, but are also important rate determining conduits useful as indicators for water quality bioassessment (Merritt et al. 2009). In recent times, chironomid midges have emerged as a useful model system for environmental biomonitoring studies of acute and chronic toxicity of large number of pollutants of the aquatic ecosystem.

Although laboratory based studies may provide a preliminary assessment of various biotic and abiotic factors circulating in the environment, they are mere auxiliary tools to the interpretation of *in situ* bioassays. For this reason, assessment of the responses of test organisms to laboratory exposures along with field based studies would help minimize the biases associated with lab-to-field extrapolation. Unfortunately, very few attempts have helped clarify this issue. A few of such studies have exploited midges, thus providing a better understanding of whether *in situ* and laboratory tests demonstrate comparable results (Faria et al. 2007). However, the absence

of an *in situ* control group of the biota is one of the limitations noticed in many contemporary experimental designs.

In an attempt at overcoming the shortcomings of comparative studies comprising laboratory and field samples, our laboratory team has come up with a novel *ex situ* technique suitable for rearing and carrying out simulation studies on *C. ramosus* in an insectary. This insectary is specially designed to meet the conditions as similar as possible to the prevailing natural environment. Further, rearing is carried out in small water tanks supplemented with phytoplankton and protists, thus mimicking the conditions in a lentic water body. Of the two tanks inside the insectary, one serves for *ex situ* control studies and the other for corresponding experimental studies in which stressor concentrations can be manipulated as per the requirement of the experimental design. The insectary is kept covered with a shade net in order to prevent the entry of other insects. The design of the insectary is depicted in Fig.1 a-c. This technique has been successfully standardized for the maintenance and breeding of *C. ramosus* and is cost effective involving less labour input. To the best of our knowledge, there are no reports of similar techniques practised for chironomid midges where natural populations can be maintained and subjected to natural seasonal variations.

The tanks were constructed of clay bricks reinforced with concrete columns. To begin with, egg masses collected from the field were reared and developmental stages were validated for taxonomic identification using morphological and cytotaxonomic keys (Chaudhuri et al. 1992, Nath and Godbole 1997). To initiate rearing, ≈100 late fourth instar larvae each were transferred to rearing tubs (Ø=35 cm) placed in a net cage under laboratory conditions (Fig. 1 d), which were maintained exactly as described by Nath and Godbole (1998). Similarly, ≈100 larvae were transferred to the insectary tanks, covered with net cages and the base of the tanks was layered (2 cm) with silt and finely ground moss that served as soft substratum for tube building. Special food formulation (Nath and Godbole 1998) was supplied to the developing cultures every two days.

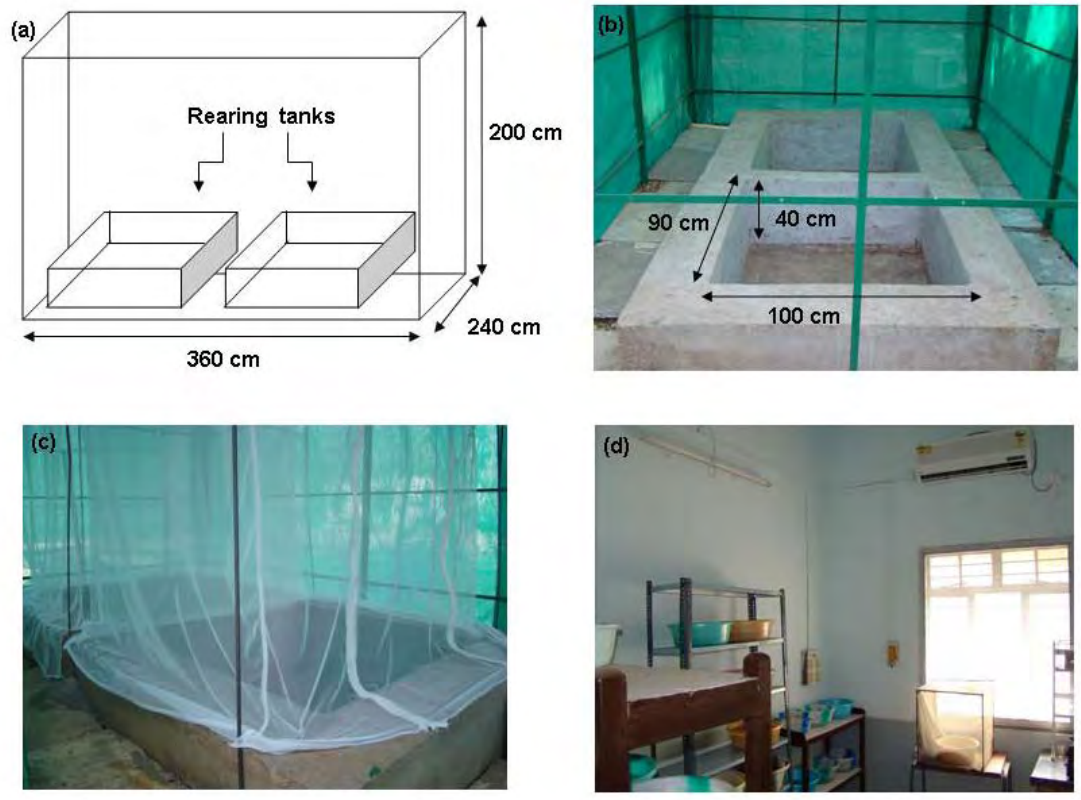

Fig. 1: a) Blue print of the insectary showing the location of the rearing tanks within the insectary. **b)** Interior view of the insectary showing the two rearing tanks. The insectary is shown covered with a shade net. **c)** Interior view of the insectary showing the two rearing tanks covered with net cages and each filled with about 130 litres of water. **d)** Special laboratory facility for rearing populations under ambient laboratory conditions.

The absence of predators of *C. ramosus*, placed it at the last trophic level of the food chain in the rearing tanks. It was found unnecessary to change the water of the tanks on a routine basis, thus saving both time and effort. However, whenever cleaning of the ponds was demanded, the water was flushed out through a small outlet present at one of the basal corners of the tanks. The desired level of water in the tanks was maintained through appropriate replenishments.

Our observations indicated that the life cycle of *C. ramosus* from egg to adult in the laboratory reared populations was completed in about 36 days, while it took about 33 days for the insectary reared population. The differences of growth parameters found in the two groups was found to be negligible (ANOVA, P> 0.05) (Table1).

Table 1: Comparison between lab reared (LR) and insectary reared (IR) populations for various growth, developmental and emergence parameters. d = duration in days.

Parameter	**LR population**	**IR population**
Hatching of eggs	2d	2d
1^{st} to 4^{th} instar stage	29 ± 1.25 d	25 ± 2.06 d
Pupal stage	3 ± 0.11d	3 ± 0.21 d
Adult life span	2.5 ± 0.68 d	3 ± 0.32 d
# of swarming adults on the day of egg laying	20 ± 3.25	25 ± 2.66
Sex ratio on the day of egg laying	1:1	1:1
Total egg masses obtained	6	9

The suitability of this novel technique proved successful when egg masses (i.e. 1^{st} generation) were obtained from the founder population. When reared in a similar manner, these egg masses perpetuated to successive generations (so far, 4 generations have been successfully reared in the tanks). Therefore, the rearing conditions maintained in the insectary showed future potential for undertaking genetic toxicological studies. Most of the ecotoxicological research has been aimed at investigating the responses of field samples under laboratory experimental regimes. But what these studies lack is an *in situ* understanding and implications of the stressors. The novel approach adopted by us, would provide an alternative design for *ex situ* assessment of toxicants using chironomid midges as model systems and gaining useful insights to queries of the ecological implications of the abiotic factors that prevail in the natural environment and whose role in the various developmental processes cannot be played-out. We hope that the model of the insectary presented here, would attract the attention of chironomidologists for designing simulation experiments, often required in environmental assessment studies.

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