## TECHNIQUE FOR SURVEYING LARVAL POPULATIONS OF COQUILLETTIDIA PERTURBANS

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ABSTRACT. A dipper with a 1-mm mesh screen bottom was used to sample larval populations of *Coquillettidia perturbans* from plant roots in Minnesota wetlands. This sampling technique was especially useful for large-scale larval surveys because the sampler was portable, individual sample collection and processing could be completed in <10 min and data collected were appropriate for statistical analyses. Sampling indicated that larval populations were clumped, with a negative binomial model closely describing larval distributions.

A large-scale larval control program against Coquillettidia perturbans (Walker) was initiated in 1984 by the Metropolitan Mosquito Control District (MMCD) in the Minneapolis-St. Paul area of Minnesota. For this program, sampling larval populations of Cq. perturbans posed several challenges. First, in the projected control area, Cq. perturbans were found to breed in >300wetlands. Yet in many of these sites, larval densities in some years were negligible. Thus, breeding in every prospective site needed yearly verification. Second, only a portion of each wetland typically supported larvae. Thus, each wetland needed to be surveyed intensively to identify where larvae were concentrated. This emphasis on within-site sampling significantly reduces the material and labor costs involved in control (Sjogren et al. 1986). Third, the time period appropriate for sampling these sites was short. Although Cq. perturbans larvae in Minnesota are abundant from early autumn through late spring, a determination of the extent of breeding in autumn before freeze-up allows the MMCD to precisely allocate material and labor for subsequent late-winter and spring treatments. And finally, the cattail (Typha sp.) stands in most breeding sites were dense and only could be accessed on foot. Thus, sampling equipment was needed that could be carried manually under these conditions, often for long distances.

Several techniques exist to sample Cq. perturbans larvae (McNeel 1931, Bidlingmayer 1954, Morris et al. 1985, Walker and Crans 1986). Although these techniques have proven value (Olds et al. 1989, Morris et al. 1990), large-scale surveys such as those required for the MMCD program may not be practical using these techniques; sample processing time can be long or equipment may be cumbersome. Alternatively, Barton (1964) described a very simple technique

whereby *Cq. perturbans* larvae were scraped from plant roots with a standard mosquito dipper; he later found that use of a dipper with a screen bottom increased collection efficiency. With this portable apparatus, many sites and areas within sites could be sampled rapidly, although the value of these results for statistical analyses was unknown. Here I describe how collections with a screen-bottom dipper can be standardized to yield data suitable for statistical analyses.

I used a screen-bottom dipper that had the general form of a standard mosquito dipper. Rather than the typical dipper cup, however, I used a 1-mm mesh screen unit that was 12.7 cm (5 in.) in diam. with a 4-cm-high brass rim (similar screens are available from scientific supply firms). This screen-bottom dipper was welded onto a handle made from a 1-m long section of 1-in. metal pipe.

In Minnesota, larvae attach to the roots of plants that are either emergent (i.e., anchored to the bottom) or floating (Batzer and Siogren 1986). Sampling procedures were developed for both conditions. For emergent plants, the screen-bottom dipper was oriented so that the distal edge of the screen rim was at the base of the plant. The dipper was then scraped up vertically through the plant root mass and then along the plant stem to the surface. Because larvae cannot live in consolidated substrates (Armstrong 1980, Hagmann 1980, Batzer and Sjogren 1986), underlying mud need not be sampled. However, roots covered by unconsolidated detritus (flocculence, plant material) should be sampled because they are suitable larval habitats (Batzer and Sjogren 1986). For floating plants, the dipper was oriented horizontally beneath the plant a distance of 50 cm (larval numbers under floating plants decline at distances >60 cm from open water [Batzer and Sjogren 1986]). The distal edge of the dipper rim was then pushed firmly up into the root mass and scraped out along the underside of the floating plant to open water, and then brought to the surface.

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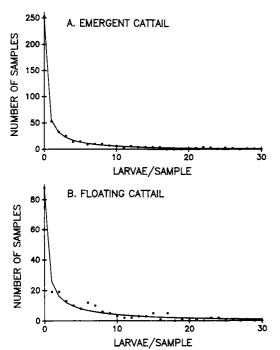


Fig. 1. Observed number of screen-bottom dipper samples (dots) with 1-30 larvae per sample and expected numbers of samples (line) if larval distributions followed a negative binomial model from (A) 25 emergent cattail stands, and (B) 24 floating cattail stands in Minnesota. Those few samples with >30 larvae are not indicated here.

Often dead cattail stems and leaves became draped over the dipper while the sample was brought from the roots to the water surface. Because these materials hindered sorting larvae, I determined the loss in sample precision that would occur if large stems and leaves were rinsed in the dipper and then discarded without examination. In 3 trials where rinsed stems and leaves were collected and then examined for additional larvae, only 3, 4 and 6% of the total larvae in samples occurred among these stems and leaves (for each trial, sampling was conducted until at least 100 larvae were collected). Thus, sorting time can be reduced with minimal loss in precision by discarding large stems and leaves without examination.

Larvae were sorted on-site by backwashing water through the screen until most of the fine sediments were eliminated. By agitating the residual detritus and tapping the dipper rim, larvae were induced to swim to the surface where they were collected with a pipette. The typical white color of the larvae made them visible among the materials in the dipper and their slow swimming action made them easy to capture. Typically, all larvae could be removed from a sample in <10 min. As many as 83 larvae/sample were recovered from floating plants and 69 larvae/sample from emergent plants.

For a sampling method to be useful, results must be repeatable. Therefore, test areas were sampled twice within 48 h to determine if similar larval densities were collected. Ten emergent cattail stands (n = 15 per sample) and 5 floating cattail stands (n = 10 per sample) were sampled. In all cases larval densities were similar in both paired samples (t-test, P > 0.05). Percent differences between pairs averaged 19.4  $\pm$  2.7% (SE). For the 30 samples, mean larval densities ranged from 1.8 larvae/dip to 22.0 larvae/dip, the coefficient of variation averaged 0.864 (SE = 0.052).

With the suction sampling methods used by Olds et al. (1989) and Morris et al. (1990), distributions of Cq. perturbans larvae were found to be clumped (i.e., followed a negative binomial distribution). To determine if sampling results obtained using the screen-bottom dipper showed similar patterns, larval distributions were determined from a 1982 survey of all known sites in a 1,400-km<sup>2</sup> area of Ramsey and Hennepin counties. Twenty-five breedings sites with emergent cattail and 24 sites with floating cattail were sampled using the procedure described above. For both situations, chi-square goodness-of-fit tests (P > 0.05) indicated that observed frequencies of samples with specific larval numbers were very similar to expected frequencies given that distributions followed a negative binomial model (emergent cattail stands, Fig. 1A; floating cattail stands, Fig. 1B). Thus, results from sampling with the screen-bottom dipper and those from previously calibrated sampling methods all indicated similar larval distributions. For populations that have the high sample variabilities that are inherently associated with negative binomial distributions, accurate estimations of densities require large numbers of samples (Southwood 1978). This pattern by Cq. perturbans underscores the need for a method where samples can be rapidly collected and processed.

Some problems were encountered when sampling with the screen-bottom dipper. As with the pump methods of Morris et al. (1985) and Walker and Crans (1986), workers using the screenbottom dipper must be aware of where roots supporting larvae are likely to occur. In the same habitats, inexperienced workers initially collected fewer larvae per sample than I did. However, as workers became experienced, we collected similar larval numbers.

Another problem with the sampler was that the 1-mm mesh size efficiently retained only 3rd and 4th instars; 2nd-instar larvae could pass through the screen. Because 3rd and 4th instars dominate larval populations in autumn (Olds et al. 1989), the inability to retain 2nd instars was not a significant problem for the autumn sampling required by the MMCD program. Similarly, spring sampling with the screen-bottom dipper should yield reliable results. A finer mesh could be used to collect 2nd instars (Olds et al. 1989) but this would increase sample processing time because more detritus would be retained.

Overall, the screen-bottom dipper proved to be a simple, reliable means of rapidly assessing Cq. perturbans larval numbers. Although this method is particularly suited for operational control programs where extensive surveys are required, the method is also appropriate for experimental studies on the ecology and control of Cq. perturbans (Batzer and Sjogren 1986, Sjogren et al. 1986, Batzer and Resh 1992).

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