CONTROL OF CHIRONOMID LARVAE (DIPTERA: CHIRONOMIDAE) IN ESTABLISHING RICE CROPS USING STARCH-BASED CHLORPYRIFOS PELLETS

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ABSTRACT. Starch-based pellets containing 10% (w/w) chlorpyrifos were evaluated for control of chironomid larvae in rice crops immediately after flooding. Chlorpyrifos pellets at rates of 2.0, 3.5, and 6.0 kg/ha (0.2, 0.35, and 0.6 kg AI/ha), and, as a reference treatment, 150 ml/ha of 500 g/liter chlorpyrifos emulsifiable concentrate (EC) (0.075 kg AI/ha) were evaluated in New South Wales, Australia. The pellets reduced total midge larvae by 93–94% over the initial 36 days posttreatment, with some indication of residual control 50 days posttreatment at all rates. Chironominae were reduced by 80–85%, and other chironomids (predominantly Tanypodinae) by 98% during the first 36 days posttreatment. Increasing the pellet application rate above 2.0 kg/ha did not improve control. Although the EC formulation provided 100% control of all larvae for 12 days, control declined thereafter. The EC formulation reduced total larval numbers by 55% over the first 36 days posttreatment, with Chironominae increasing by 28% and other chironomids being reduced by 84%. Significantly (P < 0.05) higher water column toxicity levels were recorded for the EC formulation in the first 3 days after application.

INTRODUCTION

Chironomid midge larvae cause extensive damage to newly sown rice crops in southern New South Wales (NSW), Australia (Stevens 1991). Larvae either enter the seed and consume the endosperm and embryo, or attack the developing root systems, retarding plant growth and rendering aerially sown crops vulnerable to wind damage. High levels of larval activity destabilize the soil surface, resulting in poor plant anchoring, and lead to turbidity, which reduces photosynthesis in fully submerged seedlings.

Chironomus tepperi Skuse is the most important midge species associated with rice plant damage in NSW. In untreated rice bays *C. tepperi* generally has only a single generation during crop establishment. Eggs are laid immediately after fields are flooded, and peak adult emergence occurs approximately 16–17 days later. First generation adults rarely oviposit in the fields from which they have emerged (Stevens 1994).

New South Wales rice fields support a diverse chironomid fauna (Stevens 1994), as do rice fields in the USA (Darby 1962, Clement et al. 1977). Although the biology and control of *C. tepperi* has been the subject of considerable research, little is known about the impact of other Australian chironomids on rice cultivation. However, all sediment-dwelling species contribute to root destabilization and water clarity problems, and consequently larval control needs to be maintained for at least 30 days after sowing.

Current control procedures involve treating pregerminated seed with malathion or trichlorfon, and applying an additional treatment (usually chlorpyrifos) 4–6 days after sowing. A second chlorpyrifos treatment 18–22 days after sowing is sometimes required, particularly if cold weather retards plant establishment.

Organophosphorus granules and pellets are well known for residual efficacy and elimination of spray drift. Chlorpyrifos and temephos pellets provide effective residual control of mosquito larvae (Nelson et al. 1976, Nasci et al. 1994), and granules effectively control chironomid larvae (Mulla et al. 1973, Ali and Mulla 1976). We conducted this study to determine whether a starch-based chlorpyrifos pellet formulation has the potential to provide effective, single application control of larval chironomids in NSW rice fields at environmentally and economically acceptable application rates.

MATERIALS AND METHODS

The trial was conducted using 2 parallel rows of rice bays, each bay 14.5×5 m. The bays were constructed on a Birganbigil clay loam (van Dijk 1961) at Yanco Agricultural Institute (34°37'S, 146°26'E) in southwest NSW. Ten bays were used in the experiment, with alternate flooded bays serving as buffers to eliminate any contamination due to insecticide seepage. No fertilizers or herbicides were used during the trial. Bays were flooded on October 28, 1992, from a common supply using plastic siphons, with an overflow system in the supply being used to maintain a constant water depth of 10.8 cm (SE \pm 0.15 cm, n = 160). Pregerminated rice (Oryza sativa Linn., cv. 'Amaroo') was broadcast by hand into all treatment and control bays at 120 kg dry weight/ ha immediately after flooding.

Starch-based chlorpyrifos pellets (10% AI by weight, 2 mm diam, formulation G01S05) supplied by Crop Care Australasia Pty. Ltd., Brisbane, were evaluated at 2, 3.5, and 6 kg/ha (0.2, 0.35, and 0.6 kg AI/ha, or 0.186, 0.324, and 0.556 ppm total AI input). A completely randomized design was used. Each treatment was evenly applied by hand to 2 randomly selected bays immediately after flooding. Another 2 bays were treated with 500 g/liter emulsifiable concentrate (EC) chlorpyrifos (Lorsban[®] 500, DowElanco Australasia Ltd., Sydney) at 150 ml/ ha (0.075 kg AI/ha, or 0.069 ppm total AI input) using a backpack sprayer, and 2 untreated bays were maintained as controls.

Larval population assessment: Three mud core samples were randomly taken from each treatment and control bay at 4-day intervals until 36 days posttreatment. A final set of mud samples was taken 50 days posttreatment. Samples were obtained by driving a plastic cylinder (9.6 cm diam, 12.5 cm long) to a soil depth of 4 cm, and then sliding a thin plastic sheet underneath the cylinder to prevent any soil loss prior to lifting the cylinder out of the water (Stevens and Warren 1992). Samples were transferred to plastic containers and frozen until analysis. Larvae were extracted from the thawed samples using MgSO₄ flotation (Stevens and Warren 1992).

Larvae were sorted into Chironominae and "other chironomids", the later group being heavily dominated by Tanypodinae. Only 6 pupae were recovered from the samples; all were Chironominae and were incorporated into the larval data for that group. Data were transformed to $y' = \log_e(y + 1)$ prior to analysis to account for the wide range of values and skewed distribution caused by high numbers of zero counts (Goulden 1952). Results for each population category were compared to corresponding control and EC-treated larval populations using multifactor ANOVA and Tukey's HSD (honest significant differences) test (Manugistics Inc. 1992).

Water column toxicity levels: Three 150-ml water samples were taken from each of the 10 bays 1, 2, 3, 5, 7, 10, 14, and 18 days after treatment and frozen at -17° C for later evaluation. Chironomus tepperi egg masses were collected from temporary rainwater pools at Yanco Agricultural Institute and used to establish larval cultures using the technique of Stevens (1991). Cultures were maintained at 25 ± 1°C with a 15L:9D photoperiod.

Water samples were thawed and aerated at 25°C prior to use in bioassays against 4th-instar *C. tepperi* larvae. Bioassays were conducted using 5 25-mm internal diam flat-bottomed glass tubes for each sample. A small quantity of ethanol-sterilized shredded tissue paper was added to

each tube to minimize contact between larvae, together with 20 ml of water sample and 10 4thinstar *C. tepperi* larvae. Bioassays were held at $25 \pm 1^{\circ}$ C and 15L:9D for 24 h before assessment. Larvae unable to make a sustained coordinated response when lightly grasped with a pair of fine forceps were considered dead. Mortalities were transformed to percentages, and corrected using Abbott's formula (Abbott 1925) and the average mortality in the control samples taken on corresponding days. The data were then subjected to arcsine transformation (Goulden 1952) prior to analysis using multifactor ANOVA and Tukey's HSD test (Manugistics Inc. 1992).

Environmental conditions: Water temperature, rainfall, and dissolved oxygen concentrations were monitored in the field, and pH, conductivity, and suspended solids were assessed in the laboratory using field-collected water samples. Rainfall was recorded daily throughout the trial; all other variables were measured every 2nd day, in both the morning and afternoon (0800– 0915 and 1600–1820 h daylight saving time) until 35 days posttreatment.

Water temperature was measured with immersed maximum/minimum thermometers in each of the control bays, and rainfall was recorded using an automated weather station. Suspended solids were assessed for nonfilterable residue (APHA 1971) with 9-cm GF/C glass fiber filters (Whatman International Ltd., Maidstone, England) and an oven temperature of 105°C. Dissolved oxygen, pH, and conductivity were assessed using electronic meters. Two oxygen measurements were made in each control bay at each sampling, and a single 250-ml water sample from each control bay was used for pH and conductivity measurements.

RESULTS

Larval populations: Figure 1 shows the results of larval population assessments. Control populations increased progressively until day 24, then Chironominae declined rapidly and other chironomids (predominantly Tanypodinae) predominated. The chlorpyrifos EC treatment totally eliminated all chironomids for 12 days. Over the initial 36 days of the trial, 55% overall control was achieved. Chironominae increased by 28% relative to control populations, but other chironomids were reduced by 84%. All application rates of the pellet formulation reduced total larval numbers by 93-94% during the initial 36 days of the study. Chironominae were reduced by 80-85%, and other chironomids by 98%. There was some evidence of residual control at all application rates 50 days posttreatment.

Water column toxicity levels: Water column



Fig. 1. Populations of chironomid larvae in control and chlorpyrifos-treated rice bays. a. Significantly lower than equivalent control population; b. Significantly lower than equivalent chlorpyrifos EC-treated population (ANOVA, data transformed to $y' = \log_e[y + 1]$, Tukey's HSD test [P < 0.05]).



Fig. 2. Mortality of 4th-instar Chironomus tepperi larvae in bioassays of water taken from bays treated with chlorpyrifos EC and pellets. a. Significantly higher mortality than other treatments sampled on that day (ANOVA, arcsine transformed data, Tukey's HSD test [P < 0.05]).

toxicity assessments are shown in Fig. 2. The chlorpyrifos EC treatment provided significantly (P < 0.05) higher levels of water column toxicity in the first 3 days after application, and, with the exception of the 6 kg/ha treatment at 10 days posttreatment, all other treatments were statistically similar on all sampling days.

Environmental conditions: Conditions during the trial are summarized in Table 1, and were broadly similar to those experienced at the study site in previous seasons (Stevens 1991; Stevens and Warren 1992, 1994). Rainfall during the early stages of the trial was, however, uncharacteristically heavy, with 5.35 cm falling in the first 5 days after treatment application. Total rainfall over the 50 days was 17.43 cm.

DISCUSSION

Starch-based chlorpyrifos pellets provided significantly better residual control of chironomid larvae than the EC chlorpyrifos formulation, the standard postsowing treatment. Under normal field usage, EC chlorpyrifos is applied 4-6 days after sowing, the seed being protected initially by a 600 ml/ha (0.3 kg AI/ha) EC malathion seed treatment. The maximum effective period of rice crop protection provided by these 2 treatments is between 18 and 22 days, in comparison to at least 36 days obtained by a single application of chlorpyrifos pellets. The optimal application rate for the pellets is 2 kg/ha (0.2 kg AI/ha) or less; higher application rates did not improve control. The low levels of water column toxicity provided by the pellet formulation are presumably a consequence of chlorpyrifos adsorbing strongly onto

sediments and suspended particles (Mulla et al. 1973, Merriam et al. 1981).

The LC_{90} of chlorpyrifos against 4th-instar C. tepperi larvae under laboratory conditions is approximately 0.0019 ppm (Stevens 1992). The EC chlorpyrifos formulation applied at 150 ml/ha (0.075 kg AI/ha) provides an Expected Environmental Concentration (EEC) (Ross et al. 1994) of approximately 0.07 ppm when applied to water 10.8 cm deep, almost 37 times the LC₉₀ concentration. It is apparent from our results that the maximum water column chlorpyrifos levels arising from the pellet formulation are only a fraction of the EEC for the emulsifiable concentrate treatment. Similar differences in the EECs of liquid formulations and water column concentrations produced by sustained-release methoprene formulations have been reported by Ross et al. (1994).

The results of this study support previous investigations into the impact of pelletized and sus-

Table 1. Environmental parameters in experimental rice bays, Yanco, NSW, Australia (October 28-December 2, 1992).

Mean	Range
7.43	6.40-9.13
153	91-188
14.3	1.0-39.3
9.2	7.6–11.4
102.9	79.3–145.1
22.1	8.0-36.0
	Mean 7.43 153 14.3 9.2 102.9 22.1

tained-release formulations on chironomid larvae. The use of chlorpyrifos pellets as an alternative to liquid formulations for larval chironomid control will reduce both application costs and the risk of spray drift associated with the

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aerial application of chemicals to NSW rice fields.

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