Polymeric Microcapsules of Cyanazine: Preparation and Evaluation of Efficacy

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In two separate studies, the herbicide cyanazine was microencapsulated within the following polymers: cellulose acetate butyrate (CAB); ethyl cellulose of two different viscosities (EC22 and EC100); low and medium molecular weight poly(methyl methacrylate) (PMML and PMMM); poly-(α-methylstyrene) (PMS). Pesticide efficacy studies on five weed species (barnyardgrass, smallflower morningglory, crowfootgrass, Florida beggarweed, and Palmer amaranth) were conducted under greenhouse conditions over a period of 12 weeks for study 1 and 25 weeks for study 2, using the commercial formulation Bladex 90 DF for comparison purposes. In both studies, the poly(α-methylstyrene) formulations were consistently comparable or superior to the commercial formulation in long-term control.

Keywords: *Cyanazine; ethyl cellulose; formulation; greenhouse; groundwater; herbicide; microcapsule; poly(α-methylstyrene); polymer; weed*

INTRODUCTION

In the last two decades, there has been increased concern over the contamination of groundwater and surface water by pesticides (Wauchope, 1978; Squillace and Thurman, 1992; Pereira and Hostettler, 1993). In 1986, the U.S. Environmental Protection Agency disclosed that at least 17 pesticides used in agriculture had been found in groundwater in 23 states (Cohen et al., 1986). According to a 1988 interim report, 74 different pesticides have been detected in the groundwater of 38 states from all sources. Contamination attributable to normal agricultural use was confirmed for 46 different pesticides detected in 26 states (Williams et al., 1988).

The herbicides atrazine, cyanazine, simazine, metribuzin, alachlor, and metolachlor have been frequently implicated in groundwater and surface water contamination (Cohen et al., 1986; Williams et al., 1988; Squillace and Thurman, 1992; Pereira and Hostettler, 1993). According to the National Alachlor Well Water Survey (Holden et al., 1992), within the targeted alachlor use area, alachlor was detected in 0.78% of the private rural domestic wells, atrazine in 11.7%, cyanazine in 0.28%, metolachlor in 1.02%, and simazine in 1.60%. It was estimated that the following amounts of the five major herbicides were discharged into the Gulf of Mexico in 1991: atrazine, 160 metric tons (t); cyanazine, 71 t; metolachlor, 56 t; alachlor, 18 t; simazine, 10 t (Pereira and Hostettler, 1993).

Controlled-release technology, particularly microencapsulation, should be useful in reducing environmental contamination (Bahadir and Pfister, 1990; Riggle and Penner, 1990; Seaman, 1990; Williams, 1984; Scher, 1977). Microencapsulated pesticides should be safer to handle, reduce the total amount of pesticide used, and have reduced potential for leaching in the soil profile while maintaining effective biological activity. For example, Petersen et al. (1988) reported that microencapsulated formulations of alachlor and acetochlor exhibited increased persistence in the soil in comparison to emulsifiable concentrate formulations. Fleming et al. (1992) presented similar findings for microencapsulated alachlor. Huang and Ahrens (1991) and Green et al. (1992) reported that microencapsulated alachlor prolonged weed control, persisted longer in the soil, and reduced leaching in comparison to an emulsifiable concentrate formulation. Previously, we reported the preparation of *β*-cyclodextrin complexes of atrazine, metribuzin, and simazine and evaluation of their efficacy as herbicides under greenhouse conditions (Dailey et al., 1990).

The chief objectives of our research are to develop herbicide formulations that will maintain or increase efficacy on target weeds and that will not adversely impact on the environment. We are investigating a number of methods for the microencapsulation of herbicides. Among these is the solvent evaporation process which yields microcapsules of herbicide incorporated within a polymer matrix. We have reported the preparation and evaluation of polymeric microcapsules of atrazine, metribuzin, alachlor, and metolachlor (Dailey et al., 1993; Dailey and Dowler, 1995a). In this paper, we will describe the preparation of polymeric microcapsules of cyanazine and the evaluation of their effectiveness in controlling weeds in the greenhouse, as conducted in two separate studies. We have made a preliminary report on the results of the first of these studies (Dailey and Dowler, 1995a,b). The greenhouse studies constitute an initial step in evaluating these polymeric microcapsules as a potential new source of controlled release formulations.

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MATERIALS AND METHODS

Chemicals and Reagents. Technical cyanazine (97.2%) was provided by Du Pont, Wilmington, DE, and was used without further purification. Samples of the 88% hydrolyzed poly(vinyl alcohol)s Airvol 205 (low viscosity) and Airvol 523 (medium viscosity) were provided by Air Products and Chemicals, Inc., Allentown, PA. Stock 0.5% solutions of Airvol 205 and 523 were prepared by adding the poly(vinyl alcohol) in a steady stream to the vortex of cold stirred water followed by heating at 85 °C for 30 min. The following polymers were purchased from Aldrich Chemical Co., Inc.: cellulose acetate butyrate, butyryl content 17%, T_m 235 °C (CAB); ethyl cellulose, ethoxyl content 48%, viscosity (5% solution in 80/20 toluene/ethanol) 22 centipoises [EC22]; ethyl cellulose, ethoxyl content 48%, viscosity 100 cps (EC100); poly(methyl methacrylate), low molecular weight (PMML); poly(methyl methacrylate), medium molecular weight (PMMM); poly (α-methylstyrene), medium molecular weight (PMS).

Preparation of Polymeric Microcapsules. In a typical microcapsule preparation for study 1, a solution of 2.50 g of cyanazine and 10.0 g of polymer in 200 mL of dichloromethane was added slowly to the vortex of 1000 mL of a 0.25% Airvol 205 or 523 solution, stirred at 350 rpm. A Lightnin Labmaster variable-speed high-torque mixer (Mixing Equipment Co., Inc.) equipped with a 5.0-cm diameter six-bladed turbine impeller was used for all stirring. Stirring at 350 rpm was continued for 24 h, at which time evaporation of the organic solvent was complete. After the stirring was halted, the microcapsules were allowed to settle (1.0−1.5 h). The supernatant liquid (including floating solids) was decanted, 1000 mL of distilled water was added, and the mixture stirred for 0.5−1.0 h. After settling, the microcapsules were filtered, allowed to air-dry, and finally dried in a vacuum desiccator until a constant weight was obtained. The microcapsules for study 2 were prepared at 60% of the scale used for study 1.

In subsequent discussions, a polymeric microcapsule formulation will be referred to in abbreviated form, such as *CAB-205*, indicating the use of the polymer cellulose acetate butyrate and the emulsifier Airvol 205.

The herbicidal content of all the polymeric microcapsules prepared for study 1 was determined on the basis of nitrogen and chlorine microanalyses (Galbraith Laboratories, Inc.). The cyanazine content of all the polymeric microcapsules prepared for study 2 was determined by reverse-phase high performance thin-layer chromatography with densitometry (Dailey and Johnson, 1995). Based upon the amounts of materials used, each of the polymeric microcapsule formulations should contain 20% active ingredient.

Greenhouse Studies. A 10−10−10 fertilizer was thoroughly mixed at the rate of 1000 kg/ha into an air-dried Tifton loamy sand top soil (fine-loamy, siliceous, thermic, Plinthic Paleudults) with 83% sand, 10% silt, 7% clay, and 1.0% organic matter) and placed in 20 × 35 × 9 cm deep galvanized steel flats. The soil was then uniformly moistened by sprinkler from the top and allowed to equilibrate for 24 h. The indicator crop (corn) and the selected weed species were then planted in rows 3 cm apart (15−20 seeds per row) in each flat. The flats were again lightly moistened with overhead sprinklers and the herbicides applied preemergence to crops and weeds. The commercial cyanazine formulation Bladex 90 DF was applied with an enclosed chamber sprayer using a Tee Jet 80067 flat fan spray tip, operating at 160 kPa which delivered a volume of 187 L/ha at 0.45 m/s. Spray height was 46 cm. The polymeric formulations were weighed for each individual flat, placed in a small paper envelope, and spread evenly over the soil surface by hand. The treated flats were placed in a greenhouse with day length maintained at approximately 14 h by natural or supplemental fluorescent lighting (80-W double fluorescent tubes 8 feet in length) and the temperature ranging from 20 to 34 °C. The experimental design was a randomized complete block with four replications.

The weed species used in these experiments were barnyardgrass [*Echinochloa crus-galli* (L.) Beauv.], crowfootgrass [*Dactyloctenium aegyptium* (L.) Willd.], smallflower morningglory [*Jacquemontia tamnifolia* (L.), Griseb.], Florida beggarweed [*Desmodium tortuosum* (Sw.) DC], and Palmer amaranth (*Amaranthus palmeri* S. Wats). These species were chosen for their extensive occurrence in the southeastern United States and for their different levels of tolerance to cyanazine (Weed Survey, 1989).

There were two series of greenhouse efficacy studies for cyanazine. In each study, the herbicides were applied at the rate of 1.7 kg/ha. The first study involved nine polymeric cyanazine formulations (CAB-205, CAB-523, EC22-205, PMMM-205, PMMM-523, PMS-205, PMS-523, EC100-205, and PMML-205) and the commercial formulation Bladex 90 DF. The initial planting date and date of herbicide treatment (week 0) was November 2, 1993. Efficacy on selected weeds was measured by visually estimating control compared to the untreated check 20−21 days after each planting. Percent control was recorded based on 0 = no effect and 100 = complete kill. The flats were then allowed to air-dry, the tops of dead plants carefully removed without disturbing the soil, and the same crop and weeds replanted 4, 12, and 25 weeks after initial treatment in order to determine herbicide persistence or release. The second study involved eight polymeric cyanazine formulations (CAB-205, CAB-523, EC22-205, EC22-523, EC100-205, EC100-523, PMS-205, and PMS-523) and Bladex 90 DF. The initial planting date and date of herbicide treatment (week 0) was August 29, 1995. Efficacy on selected weeds was measured by visually estimating control compared to the untreated check 20−21 days after each planting. After removal of dead plants, the same crop and weeds were replanted 4, 8, 12, 18, and 25 weeks after initial treatment in order to determine herbicide persistence or release. As part of this study, blanks consisting of polymeric microcapsules containing no active ingredient were applied to the flats. None of the polymers had any effect on any of the weed species.

Statistical Analysis. Duncan's Multiple Range Test was employed for comparison of all treatments with cyanazine formulations. The significance level was chosen to be $P = 0.05$.

RESULTS AND DISCUSSION

For study 1, the cyanazine content of the polymeric microcapsules ranged from 7.9% to 19.2% (theoretical amount: 20%). For study 2, the amount of active ingredient ranged from 6.85% to 19.2% (theoretical amount: 20%).

To estimate the particle size distribution of the microcapsules, representative samples of dried material were examined under an Olympus research microscope equipped with a Polaroid camera. The following particle size distributions (diameter of microcapsules measured in microns) were determined: CAB-205 (60−160 range, 125 mode); CAB-523 (30−225, 200); EC22-205 (25−175, 125); EC22-523 (20−175, 125); EC100-205 (40−240, 175); EC100-523 (120−425); PMS-205 (20−250); PMS-523 (25−225, 100); PMMM-523 (25−80).

For study 1, the results of the greenhouse studies are illustrated by the bar graphs in Figures 1−5. The results of the Duncan's Multiple Range Test are shown in each figure. The commercial and polymeric formulations all exhibited very low or no activity after 25 weeks; accordingly, only percent control data for 0, 4, and 12 weeks after initial treatment were used. Data for PMMM-205 and PMML-205 are not shown. Formulation PMMM-205 consistently showed very low or no activity for all weed species; PMML-205 was inactive at 0 and 4 weeks, but gave 23−50% control after 12 weeks. The following order of herbicidal activity was observed: Bladex 90DF = EC22-205 = PMS-205 = PMS-523 > CAB-523 = EC100-205 > CAB-205 > PMMM-523 > PMMM-205 = PMML-205. Both poly(α-methylstyrene) formulations and EC22-205 exhibited

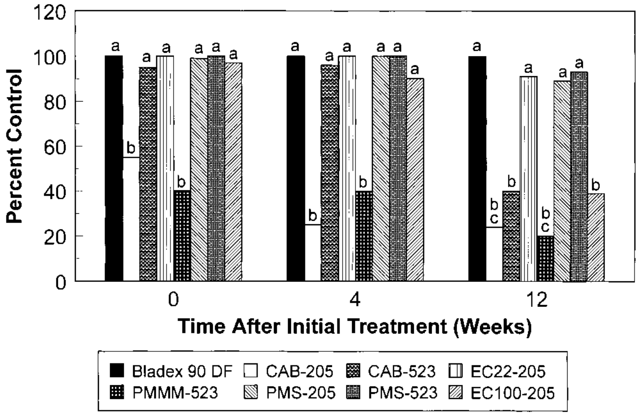

Figure 1. Efficacy of polymeric cyanazine formulations on barnyardgrass: study 1.

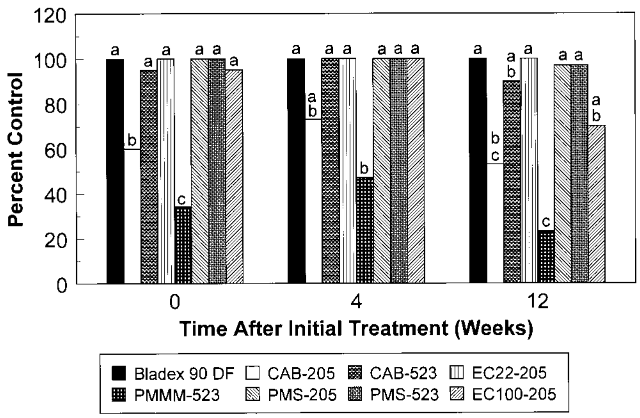

Figure 2. Efficacy of polymeric cyanazine formulations on smallflower morningglory: study 1.

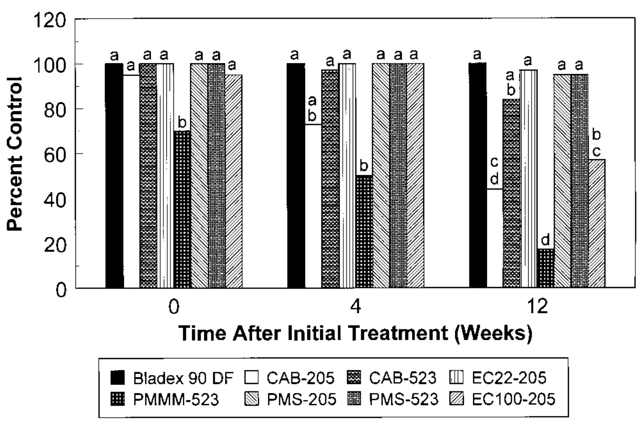

Figure 3. Efficacy of polymeric cyanazine formulations on crowfootgrass: study 1.

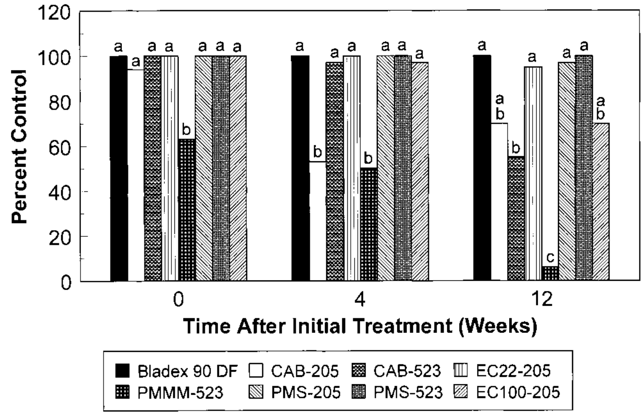

Figure 4. Efficacy of polymeric cyanazine formulations on Florida beggarweed: study 1.

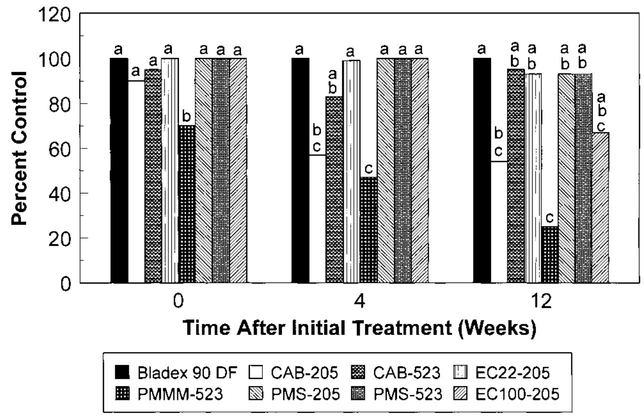

Figure 5. Efficacy of polymeric cyanazine formulations on Palmer amaranth: study 1.

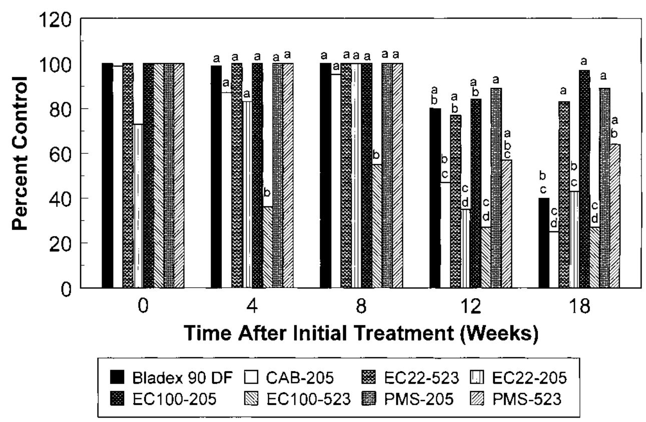

Figure 6. Efficacy of polymeric cyanazine formulations on barnyardgrass: study 2.

high activity comparable to the commercial formulation throughout the 12 weeks. Formulation CAB-523 exhibited high activity comparable to Bladex 90 DF through the entire 12 weeks against smallflower morningglory and Palmer amaranth but only for the first 4 weeks against the other weed species. For CAB and PMMM formulations, microcapsules prepared using Airvol 523 consistently exhibited higher activity than those prepared using Airvol 205. In previous studies (Dailey et al., 1993; Dailey and Dowler, 1995b), the emulsifier used generally did not have any significant effect on herbicidal activity.

In our previous studies of polymeric microcapsules of atrazine, metribuzin, alachlor, and metolachlor, CAB and EC formulations have consistently exhibited controlled-release properties and herbicidal activity equal to or greater than the commercial formulations; whereas microcapsules prepared with PMS initially exhibited high activity, but activity dropped dramatically after 4 or 8 weeks.

The unprecedented high activity observed in the PMS formulations in study 1 prompted the performance of a second study in order to confirm this activity. For study 2, the results are illustrated in Figures 6−10. The results of the Duncan's Multiple Range Test are shown in each figure. There was no significant difference among the formulations when letter designations are not shown. The four polymers which gave the best

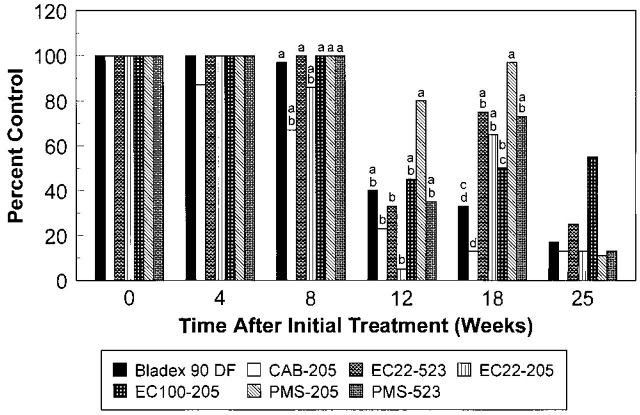

Figure 7. Efficacy of polymeric cyanazine formulations on smallflower morningglory: study 2.

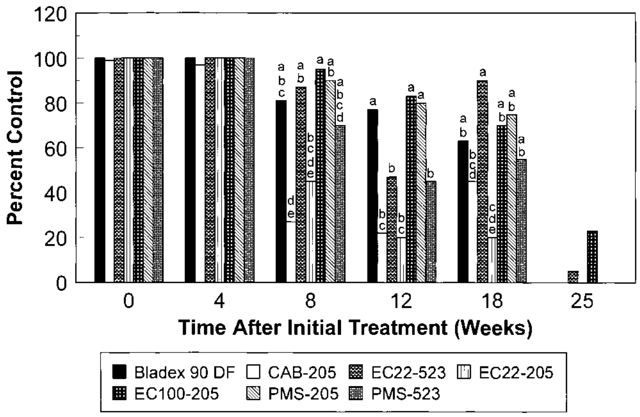

Figure 8. Efficacy of polymeric cyanazine formulations on crowfootgrass: study 2.

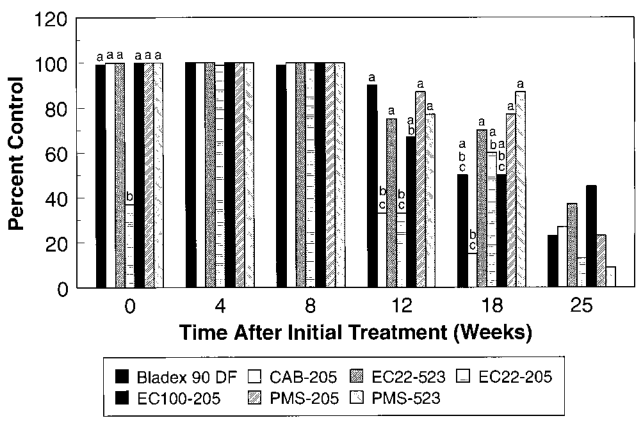

Figure 9. Efficacy of polymeric cyanazine formulations on Florida beggarweed: study 2.

results in study 1 were used for these studies: cellulose acetate butyrate (CAB); ethyl cellulose of two different viscosities (EC22 and EC100); and poly(α-methylstyrene) (PMS). No data are shown for CAB-523, which exhibited the lowest activity against all the weeds except Palmer amaranth. Formulation EC100-523 also consistently exhibited low activity, and data are shown only for barnyardgrass (Figure 6). No data are shown for week 25 for barnyardgrass since all formulations except EC100-205 were inactive at this time. Formulation EC100-205 showed the greatest efficacy against barnyardgrass and Palmer amaranth (Figure 10). Formulation PMS-205 showed the greatest efficacy against

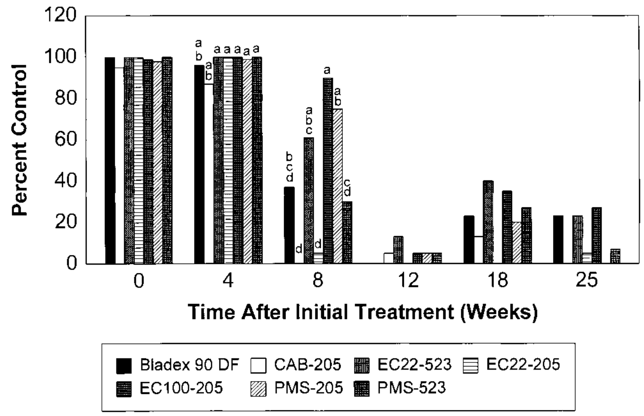

Figure 10. Efficacy of polymeric cyanazine formulations on Palmer amaranth: study 2.

smallflower morningglory (Figure 7). In general summary, the following order of herbicidal activity was observed: EC100−205 ≥ PMS-205 ≥ EC22−523 ≥ PMS-523 = Bladex 90 DF > CAB-205 = EC22−205 >EC100−523 ≥ CAB-523.

CONCLUSIONS

In previous studies (Dailey et al., 1993; Dailey and Dowler, 1995b) of polymeric microcapsules of atrazine, metribuzin, alachlor, and metolachlor, CAB and ethyl cellulose formulations have consistently exhibited controlled-release properties and herbicidal activity over time equal to or greater than the commercial formulations, whereas microcapsules prepared with poly(α-methylstyrene) initially exhibited high activity, but activity dropped dramatically after 4 or 8 weeks. In both of our studies of cyanazine formulations, the PMS formulations were consistently comparable or superior to the commercial formulation in herbicidal activity over time. In both studies, CAB formulations were less active than the commercial formulation. For EC100 and CAB formulations, microcapsules prepared using Airvol 205 consistently exhibited higher activity than those prepared using Airvol 523 in study 2, whereas CAB-523 consistently exhibited higher activity than CAB-205 in the first study. In studies of polymeric microcapsules of other herbicides, the emulsifier used generally did not have any significant effect on herbicidal activity.

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