

Leaching, Phytotoxicity, and Weed Control of New Formulations of Alachlor

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The use of commercially available formulations of alachlor has resulted in a serious environmental problem due to its leaching and migration to water sources. The objective of this study was to develop organo–clay based formulations that can significantly reduce leaching and improve weed control efficacy. We adsorbed the herbicide to montmorillonite clay particles whose surfaces were modified from hydrophilic to hydrophobic by preadsorbing to the clay an organic cation, such as benzyltrimethylammonium (BTMA) to reduce release of the herbicide into the soil. Alachlor adsorption was determined by gas chromatography. Organo–clay complexes of 0.5 mmol BTMA/g of clay gave larger adsorbed amounts and better formulations of alachlor as compared to BTMA preadsorbed up to the cation exchange capacity (0.8 mmol/g), whereas formulations without organic cations were not effective and gave little adsorption of the herbicide. The use of Fourier transform infrared spectroscopy showed that the optimal formulation also yielded the largest shifts of the stretching vibrations of the phenyl ring. The organo–clay formulations yielded slow release of the herbicide to the environment, maintaining the herbicidal activity in the top soil as measured by a bioassay using green foxtail and wheat as test plants. Laboratory and field experiments showed improved weed control at significantly reduced applied rate when alachlor was applied as organo–clay formulations.

Keywords: *Alachlor; montmorillonite; benzyltrimethylammonium; benzyltriethylammonium; benzyltributylammonium; hexadecylammonium; leaching; phytotoxicity; herbicidal activity*

INTRODUCTION

Alachlor is a preemergence herbicide for selective weed control in various field crops, such as corn (*Zea mays* L.), cotton (*Gossypium* spp.), peanuts (*Arachis hypogaea* L.), peas (*Pisum sativum* L.), sunflower (*Helianthus annuus* L.), and soybeans (*Glycine max* (L.) Merr.). The available commercial formulation (emulsifiable concentrate, EC) of alachlor suffers rapid loss of biological activity due to leaching down to deep layers in the soil profile and to the groundwater (Koncal et al., 1981; Cohen et al., 1986; Chesters et al., 1989; Yen et al., 1994). Alachlor and its degradation products (oxoethane sulfonate) were detected in shallow water in North America (Chesters et al., 1989; Koterba et al., 1993; Ritter et al., 1996; Thurman et al., 1996) and Europe (Riparbelli et al., 1996). Most detections correlate with the intensive use of alachlor in different crops. Loss of the biological activity may also occur due to photochemical degradation (Fang, 1977, 1979; Brady et al., 1982) and volatilization (Beestman and Deming, 1974; Taylor, 1978; Jury et al., 1980; Wienhold et al.,

1993). Volatility has been shown to contribute to alachlor phytotoxicity (Breeze, 1993). The leaching and surface mobility of this herbicide were correlated with its water solubility and soil organic matter content (Spillner et al., 1983). Thus high organic matter content may result in a decreased leaching into the soil profile. Significant reduction in alachlor movement was observed in soils modified by carbon-rich waste (Guo et al., 1993). Leaching rates increase with the rates of water application (Peter and Weber, 1985). Although some extent of leaching is necessary to achieve proper weed control, there are three problems associated with alachlor leaching: (1) leaching to the root zone of sensitive nontarget crops results in crop injury, (2) leaching below the root zone of the target weeds may reduce the herbicidal activity (Eshel, 1969), and (3) leaching may result in groundwater pollution (Jordan and Harvey, 1980; Koncal et al., 1981). The risk of contaminating the environment with herbicides or their degradation products has stimulated interest in developing less hazardous formulations. Modification of pesticide behavior by encapsulating the chemicals in a starch matrix is one experimental approach receiving increased attention (Wing et al., 1987). Herbicide formulations, such as starch encapsulated (Schreiber et al., 1987; Gish et al., 1991, 1994), microencapsulated (Vasilakoglou and Eleftherohorinos, 1997; Wienhold et al., 1993), tablets (Koncal et al., 1981; Gorski, 1993; Johnson and Pepperman, 1996), controlled release (Fleming et al., 1992; Buhler et al., 1994; Stork, 1997),

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and slow release (Margulies et al., 1994) have been shown to reduce volatilization and lower movement in soil. Microencapsulated formulations of alachlor were more persistent at high moisture level (Negre et al., 1992).

The objective of this work was to develop new organo-clay formulations of alachlor which may reduce its leaching and migration from the top layers of soils, thus improving weed control as tested under laboratory and field conditions. Our main idea of improving the herbicidal activity of alachlor was to adsorb it to clays whose surfaces were modified from hydrophilic to hydrophobic by adsorption of suitable organic cations to the clay surface. Such modified surfaces may better bind the nonpolar herbicide molecules, thus reducing their concentrations in solution, and consequently extend their biological activity in the field (Margulies et al., 1992, 1993, 1994). The adsorption of organic molecules to clays has been extensively studied (Mortland, 1970; Theng, 1974; Chu and Johnson, 1979; Narine and Guy, 1981; Boyd et al., 1988; Jaynes and Boyd, 1990, 1991). The organo-clays were prepared by adsorbing to clays substituted quaternary ammonium ions $[(CH_3)_3N^+R$ or $(CH_3)_2N^+RR']$, where R and R' are aromatic or alkyl species. This exchange modifies the clay surface from hydrophilic to hydrophobic (Mortland et al., 1986; Boyd and Jaynes, 1992). In the current study we prepared formulations of alachlor adsorbed to montmorillonite modified by preadsorption of various organic cations having a benzyl ring and trialkyl groups attached to the ammonium group.

MATERIALS AND METHODS

Materials. The clay used was sodium montmorillonite SWy-1 (Mont) obtained from the Source Clays Repository, Clay Minerals Society, Columbia, MO. Its cation exchange capacity (CEC) was 0.8 mmol/g of clay (Rytwo et al., 1991). Analytical grade alachlor, purity 99% (Chem Service West Chester, PA), was used for making the organo-clay formulation, whereas the commercial formulation (EC) (Alanex, 480 g ai/kg, Agan Chemical Manufacturers Ltd., Ashdod, Israel) was used as a standard formulation. Phytigel, HPLC-grade ethyl acetate, isooctane, benzyltrimethylammonium (BTMA), benzyltriethylammonium (BTEA), benzyltributylammonium (BTBA), and hexadecyltrimethylammonium (HDTMA) were purchased from Sigma-Aldrich (Sigma Chemical Co., St. Louis, MO; Aldrich Chemical Co., Milwaukee, WI). The chemical structures of the organic compounds are shown in Figure 1. Analytical grade NaCl was obtained from Frutarom Laboratory Chemicals (Haifa, Israel). Soils were collected from 0 to 30 cm depth at the Faculty's Experimental Farm in Rehovot and Agricultural Experimental Station of EPRI in Gaza Strip. Soils were analyzed and characterized to determine pH, organic-matter content, and particle size distribution (Table 1). Green foxtail (*Setaria viridis* (L.) Beauvois) and/or wheat (*Triticum aestivum* L., cv. Ariel) were used as test plants.

Preparation of Organo-Clay Complexes. The organo-clay complexes were prepared by dropwise addition of aliquots of a 10 mM aqueous solution of BTMA, BTEA, BTBA, or HDTMA to a 1% (w/v) aqueous suspension of the clay under continuous stirring (Margulies et al., 1992). After centrifugation (30 min, 16000g), the precipitate was washed three times with distilled water, freeze-dried, ground ($<50 \mu\text{m}$), and kept in plastic tubes at room temperature. The cationic loads were determined by CNHSO analyzer type Carlo-Erba 1108 and by a UV-vis absorption spectrophotometer (Uvikon 810).

Preparation of Formulations. The Mont-alachlor formulations were prepared by dissolving appropriate amounts of alachlor (weight per weight of clay complex) in acetone and adding it to the suspended powdered clay or organo-clay in

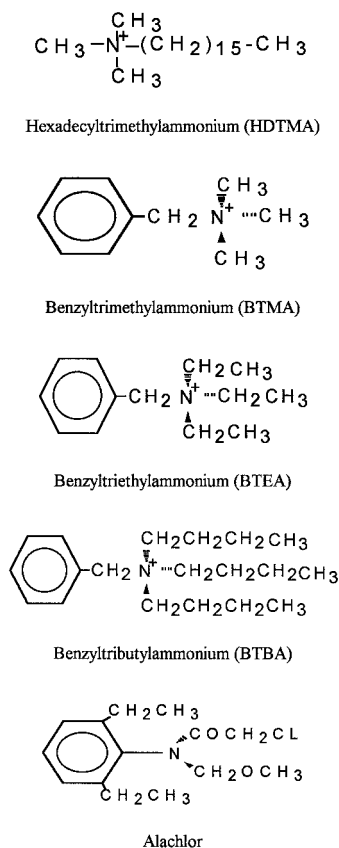


Figure 1. Chemical structure of the molecules used.

Table 1. Properties of the Soils Used in the Field and Laboratory Experiments

soil type	pH	% OM	% sand	% silt	% clay	% CaCO ₃
Rehovot	7.5	0.2	95.5	3.3	1.2	0.0
Gaza	8.0	0.7	93.0	1.0	6.0	4.9

the same solvent with a total volume of 100 mL. The solvent was evaporated under reduced pressure as previously described (Margulies et al., 1988, 1992). Several loads of alachlor on the clay complex were prepared and examined, but only results with 5 and 13% (w/w) are shown.

Adsorption Isotherms of Alachlor. The adsorption isotherms were measured in the range of 0–700 μmol of alachlor/g of Mont-organic complex with BTMA 0.5 and 0.8 mmol/g of clay. Appropriate aliquots of an aqueous stock solution of 10^{-4} M alachlor were diluted in 25 mL of distilled water and were added under continuous stirring to 5 mL of 0.5% (w/w) clay-complex suspension in a 40 mL centrifuge tube. The final concentration of the organo-clay complexes was 0.83 g/L. The samples were kept under continuous horizontal agitation for 24 h at $25 \pm 1^\circ\text{C}$. The supernatant was separated by centrifugation at 20000g for 1 h.

Sample Extraction. Sodium chloride (2.4 g) was added into a glass tube, combined with 10 mL of supernatant and 10 mL of ethyl acetate/isooctane (1:9 (v/v)). The tubes were sealed, vortexed for 2 min at the highest speed, and stored at room temperature for 1 h until the ethyl acetate/isooctane separated. The ethyl acetate/isooctane layer was collected into 25 mL volumetric flasks. The extraction procedure was repeated twice. The extracts were brought to a volume of 25 mL with the same solvent mixture, transferred to the crimp vials, sealed, and analyzed by GC.

GC Analysis. Extracts were analyzed using a Hewlett-Packard Model 6890 gas chromatograph, equipped with electron-capture detector. A Rtx-5MS capillary column (30 m \times 0.25 mm internal diameter, 0.25 μm film; Restek Corp., Bellefonte, PA) was used with nitrogen as the carrier gas at a flow rate of 2 mL min^{-1} . The nitrogen makeup flow rate was

30 mL min⁻¹. The injector and detector temperatures were 250 and 280 °C, respectively, whereas the column was programmed at 170 °C for 1 min and then the temperature was increased at a rate of 5 °C min⁻¹ to 250 °C and held there for 5 min. Blank recovery was 102 ± 3%.

Fourier Transform Infrared Spectroscopy. Fourier transform infrared (FTIR) spectroscopy was used to examine the interactions between alachlor and the clay preadsorbed with the respective organic cations as previously described (Margulies et al., 1988). The FTIR spectra were obtained using KBr pellets with a Nicolet Magna-IR-550. Spectra were recorded at room temperature in the range of 1200–4000 cm⁻¹. Difference spectra were obtained by subtracting the spectra of corresponding organo–clay or homoionic clay from the spectra of clay–organic–alachlor complexes or clay–alachlor complexes. Absolute and difference spectra were also recorded in the range of 1200–1800 cm⁻¹.

Leaching Studies in the Laboratory. Tin macrocolumns (10 × 10 cm surface area, 25 cm height) and microcolumns made of polyethylene tubes (surface area of 5 cm², 25 cm height) were filled with a sandy soil which was air-dried and sieved through a 2 mm screen. The macrocolumn surface was sprayed with various formulations of alachlor at 2.0 kg ai/ha using an atomizer, whereas in microcolumn the herbicide was applied with a pipet. The columns were carefully irrigated with 500 m³/ha applied in portions during 3–5 h with 20 min intervals. The macrocolumns were left for 24–48 h for equilibration, and then the tin columns were sliced along their lengths, hence forming two pots 10 × 5 × 25 cm each. Two test plants, green foxtail and wheat, were sown in each column half in two rows. To ensure growth, the pots were sprinkle irrigated as needed. The microcolumns were sealed at the top, placed horizontally, and sliced along their lengths. One row of green foxtail or wheat was sown along the length of the microcolumns. Shoot height and weight were determined 16 days after sowing and used as indicators to estimate the herbicide presence at different soil depths in the column. The percent of shoot growth inhibition at a soil depth was calculated according to the following formula:

$$\% \text{ growth inhibition} = 100(P_c - P_t)/P_c \quad (1)$$

where P_c and P_t are the shoot heights of the control and the treated samples at any soil depth.

Field Experiments. Experiments were conducted at two locations: in Rehovot and in EPRI, Gaza Strip, during 1996 and 1997. Beds (1 m wide and 5 m long) were prepared by using a rotary tiller and planted with green foxtail (200 seeds/m row) in summer or wheat (100 seeds/m row) in winter. Several rates of EC formulation of alachlor (2, 4, and 6 kg ai/ha) and one rate (2 kg ai/ha) of the organo–clay formulations along with the organo–clay complex without alachlor were applied preemergence using a backpack motorized sprayer at a spray volume of 300 L/ha at 245 kPa. A sprinkler irrigation system was used to water the field sites at 500 m³/ha in both locations. Herbicidal efficacy was determined by both a reduction in the shoot fresh weight and by visual evaluation of the weed control efficacy 1, 2, 4, and 8 weeks after treatment. Ratings were based on a scale of 0 (no control) to 100 (complete control). The planted test species as well as the natural weed species uniformly infesting the plots were included in the ratings. The experiment layout was a randomized block design replicated 5 times with each plot.

Leaching Studies in the Field. Experiments were conducted as described above and sprinkler irrigated with 500 m³/ha water. Soil samples were taken using half of the tin metal column used in the laboratory experiment, gently inserted to a depth of 25 cm in each plot, supported with a wide spatula to ensure complete removal of the soil in the column. The full column was carefully removed and transferred to a greenhouse. One row of each, green foxtail or wheat seeds, was sown in each column as described above. Presence of the herbicide formulations was estimated at various soil depths by measuring the shoot height or fresh weight of the

test plant. The results indicated how deep the herbicide had leached after 500 m³/ha of irrigation. The columns were arranged in a completely randomized design with five replications per treatment. The percent of growth inhibition at any soil depth was calculated according to eq 1.

Phytotoxicity of the Formulations. 1. *In Vitro Experiment.* This test was conducted to evaluate the efficacy of commercial and organo–clay formulations of alachlor under controlled environmental conditions. The procedure was as follows: in special plastic pots with a total volume of 500 mL each, 50 mL aliquots of 0.4% Phytigel containing the required concentration of the tested formulations (0.0, 0.055, 0.11, 0.22, 0.44, 0.88, and 1.76 mg ai/L Phytigel suspension) were prepared. Ten seeds of green foxtail were gently distributed on the surface of each pot. The pots were incubated in the growth chamber. The shoot fresh weight was determined 14 days after treatment. The percent of growth inhibition at each concentration was calculated as described with eq 1.

2. *Pot Experiment.* Green foxtail was planted in pots filled with 100 g of air-dried sandy soil (Rehovot). The pots had four holes at the bottom covered by tissue paper. Alachlor formulations were applied preemergence at various rates (0.0, 0.25, 0.5, 1.0, 2.0, 3.0, 4.0, and 6.0 kg ai/ha) using a chain-driven laboratory sprayer. The experiments were arranged in a completely randomized design with five replications. The shoot height and fresh weight were determined 2 weeks after treatment, and the percent growth inhibition was calculated using eq 1.

Data Analysis. The growth inhibition data were subjected to analysis of variance, and the main effects and the interactions were tested for significance using repeated measures ANOVA. Univariate comparisons of mean growth inhibition at different depths were performed by Tukey's test ($\alpha = 0.05$). The means of effects of different formulations on herbicidal activity were compared by Tukey's test at $\alpha = 0.05$, following one-way ANOVA. A semilogarithmic dose response curve was fitted to percent of growth data for each replicate in each treatment. The effective dose value that kills 50% of the test plant (ED₅₀) was interpolated for each curve. These values were compared over treatments by one-way ANOVA, and pairs of means were thereafter compared by the Tukey test.

RESULTS AND DISCUSSION

Preparation of Organo–Clay Complexes. The cationic quaternary ammonium compounds shown in Figure 1, which are solids at room temperature, are surface-active agents (surfactants) because each molecule has a nonpolar hydrocarbon group and an ionic, polar component. The molecular structures of the selected compounds include an aliphatic part with alkyl-chain lengths ranging from 1 to 16 carbon atoms and an aromatic part with one phenyl ring and trialkyl chain length ranging from 1 to 4 carbon atoms. Figure 2 shows the results of CHNSO analysis for the carbon content of the organo–clays at different added concentrations of organic cations. The amounts adsorbed followed the sequence HDTMA > BTMA > BTEA > BTBA, but the overall pattern is similar for adsorption up to the CEC of the clay. At a low concentration (0.5 mmol/g of clay), practically complete adsorption of HDTMA, BTMA, and BTEA was achieved. Unlike nonionic organic compounds, large organic cations can effectively displace inorganic ions such as Ca²⁺ and Na⁺ from the negatively charged mineral surfaces of clay and natural soil by ion exchange (Khan, 1972; Narine and Guy, 1982; Smith et al., 1988, 1990; Nir et al., 1994; Polubesova et al., 1997). As a result, organic cations are strongly retained by the clay surface for adsorption up to the cation-exchange capacity. In the case of the aliphatic cation (HDTMA), its adsorption to the clay increased above the CEC and reached approximately

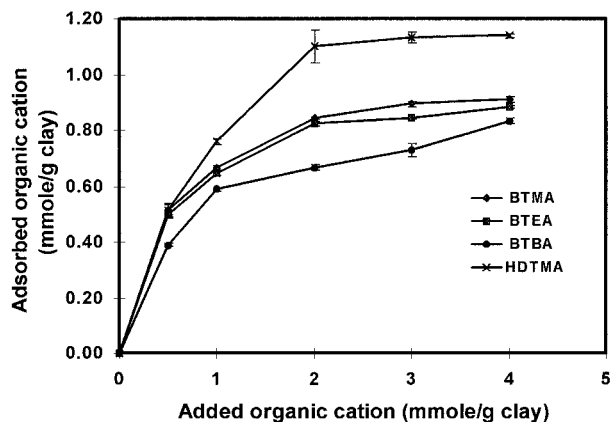


Figure 2. Adsorption of several monovalent organic cations on montmorillonite as determined by CNHSO analyzer. Bars indicate standard errors.

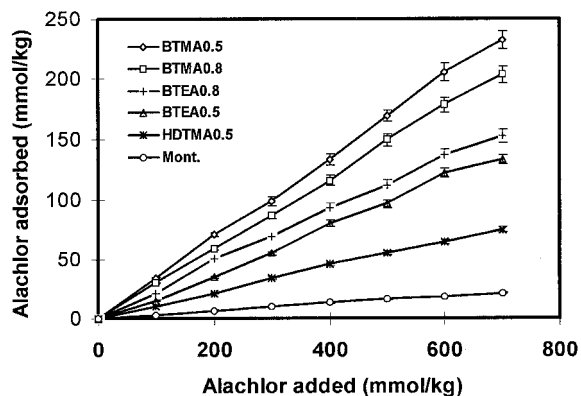


Figure 3. Adsorption isotherms ofalachlor on montmorillonite alone (Mont) and montmorillonite preadsorbed with either BTMA (0.5, 0.8 mmol/g), BTEA (0.8 mmol/g), BTEA (0.5 mmol/g), or HDTMA (0.5 mmol/g of clay). Bars indicate standard errors.

120% of the CEC. In this case, the interactions appear to be between the adsorbed and the free molecules of HDTMA via hydrogen bonding through the alkyl chains (Jaynes and Boyd, 1991). In the case of the aromatic type, the adsorption did not increase with the molecular size, unlike the case of molecules of the aliphatic type.

Alachlor Adsorption. The adsorption isotherms ofalachlor on montmorillonite (Mont), Mont-HDTMA, Mont-BTMA, and Mont-BTEA complexes are shown in Figure 3. These isotherms were generated at aqueousalachlor concentrations up to its solubility limit in water (240 mg/L). As previously demonstrated (Yen et al., 1994),alachlor was poorly adsorbed on montmorillonite alone, whereas adsorption ofalachlor on clay preadsorbed with HDTMA at a load of 0.5 mmol/g resulted in a relatively small increase in the adsorbed amounts ofalachlor. These data are in agreement with previous results reported by Jaynes and Boyd (1991). Using BTMA as a clay surface-modifying agent increased the adsorbed amounts ofalachlor 3-fold above the values with HDTMA. Partial saturation of the clay with BTMA at a load of 0.5 mmol/g clay yielded morealachlor adsorption (approximately 10% increase) than to the clay loaded up to its CEC (0.8 mmol/g of clay). These data indicate that achieving maximal transformation of the clay surface from hydrophilic to hydrophobic does not necessarily imply optimal interactions betweenalachlor and the organo-clay complex. The adsorption ofalachlor increases almost linearly as its

concentration in the equilibrium solution is increased. In the case of the less effective complex, Mont-BTEA, morealachlor was adsorbed on a clay preadsorbed by 0.8 mmol of BTEA/g of clay than with a load of 0.5 mmol of BTEA/g of clay. The low adsorption ofalachlor to the unmodified clay is due to the hydrated mineral surface of the clay and the hydrophobicity ofalachlor molecules. In contrast, modification of the clay surface with organic cations, such as BTMA at loads of 0.5 and/or 0.8 mmol/g of clay, causes a dramatic increase in the adsorption ofalachlor. This is due to the possible interaction ofalachlor molecules with the organophilic phase generated on the surface of the clay as a result of clay surface modification by organic cations. The results indicate thatalachchlor sorption to the organo-clay complex is not caused primarily by partitioning alone. Adsorption ofalachchlor followed the sequence of Mont-BTMA > Mont-BTEA > Mont-HDTMA >> Mont. These results indicate thatalachchlor adsorption to organo-clays is dependent on the size and shape of the organic cations preadsorbed on the clay, as previously suggested for other cases (Lee et al., 1989). Increasing the length of side chain from $-CH_3$ in the cation BTMA to $-C_2H_5$ in the cation BTEA resulted in a decrease of the adsorbed amounts ofalachlor. Substituting the phenyl ring with a large alkyl group ($-C_{16}H_{35}$) in HDTMA resulted in a sharp drop in the adsorbed amounts ofalachlor. The results suggest the existence of interactions between the phenyl ring of the aromatic cations (BTMA and BTEA) and the phenyl ring ofalachchlor molecules, possibly by $\pi-\pi$ interactions, and hydrogen bonding via the carbonyl and the anilidic groups, as recently suggested by Stevens and Anderson (1996). An interesting outcome was the occurrence of higher adsorbed amounts ofalachchlor to a complex of 0.5 rather than 0.8 mmol of BTMA/g of clay. The opposite trend was found for BTEA. An explanation for this unexpected result is still under investigation. However, a similar trend for Mont-BTMA formulations was also observed in leaching and FTIR experiments.

FTIR Results. Detailed information on the interactions betweenalachchlor molecules and the organo-clay complex was obtained by using infrared spectroscopy. Figure 4 shows the difference spectra ofalachchlor in its free form (a), adsorbed to the Mont-BTMA at a load of 0.5 (b) and 0.8 mmol/g of clay (c); adsorbed to Mont-BTEA at a load of 0.8 mmol/g of clay (d), adsorbed to Mont-BTBA at a load of 0.8 mmol/g of clay (e), and adsorbed to Mont-HDTMA at a load of 0.5 mmol/g of clay (f). The four peaks at 1690.3, 1458.7, 1406.2, and 1373.5 cm^{-1} in the spectrum of freealachchlor (a) were assigned to the stretching vibrations of the $-C=O$ group (1690.3 cm^{-1}), the phenyl ring (1458.7, 1406.2 cm^{-1}) and the anilidic group (1373.5 cm^{-1}) vibrations (Charles, 1981; Bosetto et al., 1993). The main differences among the spectra are as follows: (1) the peak at 1690.3 cm^{-1} of the freealachchlor (a) was shifted to a lower wavenumber at 1683.9 (b), 1684.8 (c), 1685.8 (e), and 1682.3 cm^{-1} (f) and not shifted (d); (2) the peak at 1458.7 cm^{-1} in the freealachchlor (a) was shifted to higher wavenumbers at 1471.8 (b), 1463.9 (c), 1466.5 (d), and 1462.2 cm^{-1} (e) and not shifted (f); (3) the peak at 1406.2 cm^{-1} (a), was shifted to higher wavenumbers at 1419.4 (b), 1420.5 (c), 1412.8 (d), 1421.5 (e), and 1423.3 cm^{-1} (f) and the intensity of this peak was significantly reduced (d and e); (4) the peak at 1373.5 cm^{-1} (a) was shifted into two peaks at 1375.5 and 1387.6 (b) and 1381 (c), 1381 (d)

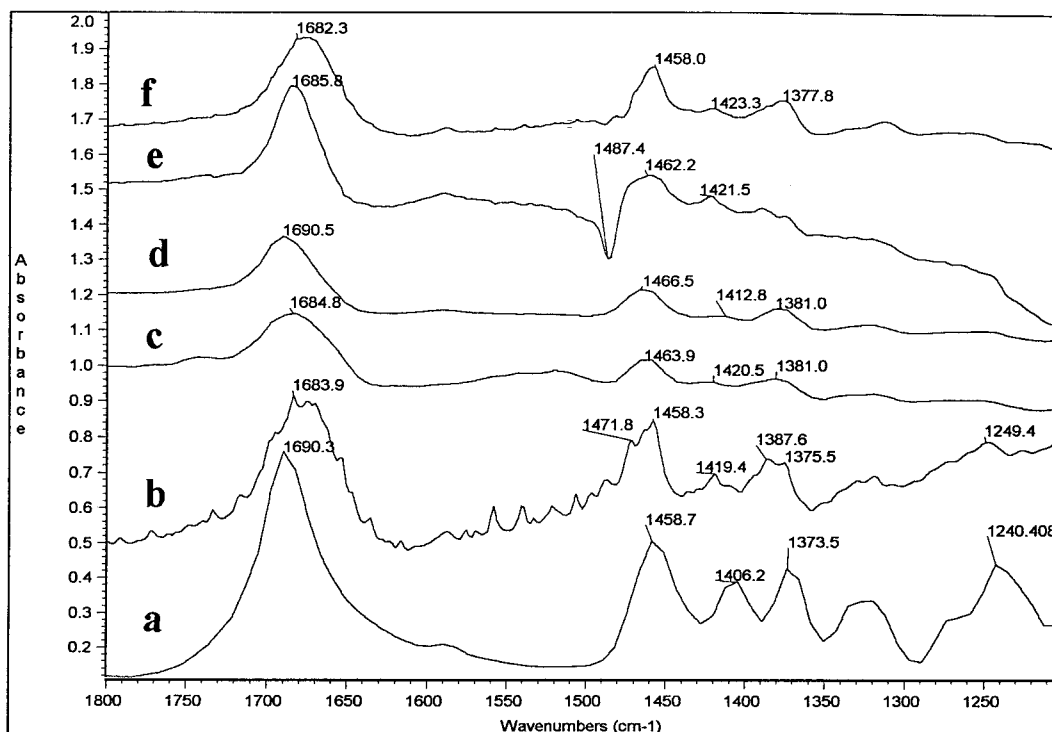


Figure 4. Fourier transform infrared (FTIR) absorption spectra of free alachlor (a) and alachlor adsorbed to montmorillonite preadsorbed by BTMA at loads of 0.5 (b), BTMA 0.8 (c), BTEA 0.8 (d), BTBA 0.8 (e), and HDTMA 0.5 mmol/g of clay (f).

disappeared at (e) and shifted to 1377.8 cm^{-1} (f). These shifts indicate that alachlor molecules interact stronger with the organo-clay complex through the phenyl rings and the anilidic group than with the carbonyl group. It can be seen from the spectra in Figure 4 that saturating the clay surface with BTMA up to its CEC resulted in a small shift of the peak at 1373.5 cm^{-1} (a) to a wavenumber of 1381 cm^{-1} (c), whereas with 0.5 mmol of BTMA/g (b) this peak shifted to 1387 cm^{-1} . The peak of free alachlor at 1458.7 cm^{-1} , which represents the stretching vibration of the phenyl ring, was shifted in all formulations involving an aromatic organic cation (b–e), but no changes were observed in case f, where the organic cation used was aliphatic (HDTMA). An increase in the number of carbon atoms in the alkyl chains of the aromatic cations to 4 [BTBA (e)] resulted in an appearance of an inverted peak at 1487.4 cm^{-1} , indicating changes in the mode of interaction.

Leaching Studies. Results of the laboratory experiments using soil columns and bioassay showed (Figure 5) that the commercial EC formulation of alachlor applied preemergence at the recommended rate (2.0 kg/ha) followed by irrigation ($500\text{ m}^3/\text{ha}$) leached to the bottom of the column. In fact no herbicidal activity was observed in the top 14 cm of the column, being detected at soil depths of 15–25 cm. Leaching of alachlor formulated with montmorillonite without any surface charge modification was reduced; however the active ingredient was completely depleted from the top 5 cm. When alachlor was formulated with clay preadsorbed with HDTMA at a load of 0.5 mmol/g, slightly decreased leaching was observed versus the alachlor formulated with montmorillonite. The other formulations resulted in an extraordinary change in the leaching behavior of alachlor. Of the several formulations shown in Figure 5 the best one was Mont–BTMA0.8–alachlor 13%, with a maximum leaching depth of 10 cm (Figure 5). It can

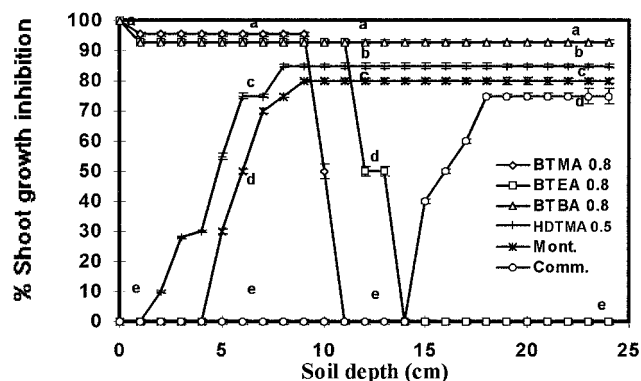


Figure 5. Leaching of different formulations of alachlor (2.0 kg/ha) in column filled with sandy soil following irrigation with $500\text{ m}^3/\text{ha}$. Green foxtail growth was used to estimate the presence of alachlor. Alachlor formulations were commercial EC formulation (comm); alachlor on clay alone (Mont); alachlor on clay preadsorbed HDTMA (0.5 mmol/g); BTBA (0.8 mmol/g); BTEA (0.8 mmol/g); and BTMA (0.8 mmol/g of clay). The clay-based formulations contained 13% alachlor. Means followed by the same letter at a representative depth are not significantly different at $p = 0.05$ level. Bars indicate standard errors.

be seen that formulating alachlor with clay preadsorbed with aromatic cations with different lengths of the alkyl chains (1 carbon atom) BTMA, (2 carbon atoms) BTEA, and (4 carbon atoms) BTBA resulted in a significant modification of the herbicide leaching behavior. With BTBA formulation, the herbicidal activity was detected through the whole column depth, indicating relatively more leaching. A possible explanation for this variable leaching pattern is the variable strength of the interactions between alachlor molecules and the molecules of the organic cations in the clay complexes, which appears to be size and type dependent. For BTMA, with the smallest size of aromatic cation, the interaction between both molecules was maximal as shown by FTIR results

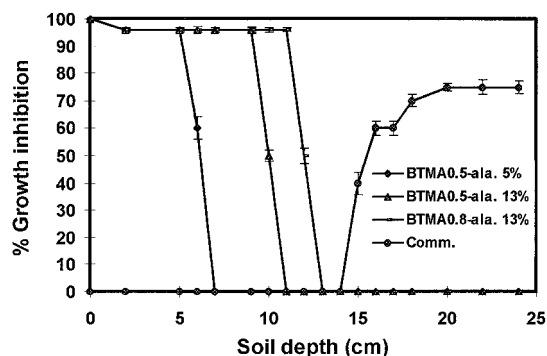


Figure 6. Effect of different loads of alachlor and BTMA on the leaching of alachlor formulations using sandy soil columns and green foxtail as a test plant. Alachlor was applied at 2.0 kg/ha followed by irrigation of 500 m³/ha. Bars indicate standard errors.

(Figure 4). With larger organic cations (BTBA) the mode of interaction is changed and the interactions are likely much weaker. Very weak interactions between alachlor and the organo-clay surface might exist with the aliphatic type molecule, HDTMA, whose leaching pattern follows largely the same sequence as the adsorption data (Figure 3). We suggest that reduced leaching is due to a slower release, which corresponds to stronger interactions between alachlor molecules and the organo-clay surface.

Several statistical tests were employed in analyzing the leaching data. Repeated measures analysis of variance was used to compare the rate of growth inhibition by treatment and depth. In all cases the differences between treatments and between depths were significant, as were the differences between depth profiles for the different treatments ($p < 0.0001$). In addition, we illustrate in Figure 5 for representative depths a comparison between treatments by Tukey's range test ($\alpha = 0.05$) following univariate analysis. According to this analysis the results are grouped as indicated by letters at representative depths.

Our results demonstrate the leaching behavior of the clay-BTMA formulation of alachlor in laboratory studies, which gave high herbicidal activity at the top of the column and minimal leaching. It can be seen that the use of aromatic and small cation (BTMA) in the formulation maintained the threshold concentration needed for weed control in the topsoil layer. As a result, a longer period of weed control may occur due to the slow release of the active ingredient to the soil environment (El-Nahhal et al., 1997).

Figure 6 shows the leaching pattern of organo-clay formulations with different loads of BTMA and alachlor. The organo-clay complex with a load of 0.5 mmol of BTMA/g of clay and a low level of adsorbed alachlor strongly inhibited leaching of the herbicide, whereas the formulation with a higher load of alachlor leached to a deeper layer (Figure 6). It can be seen that formulating alachlor at 13% ai (w/w) and a load of 0.5 mmol of BTMA/g of clay was better than formulating alachlor at 13% ai (w/w) with a load of 0.8 mmol of BTMA/g of clay in terms of leaching inhibition. Formulating alachlor at a lower rate -5% ai (w/w) with BTMA at a load of 0.5 mmol/g of clay resulted in a significantly higher degree of leaching inhibition. The explanation is that in a lower rate of alachlor, the ratio of BTMA/alachlor molecules is higher, increasing the possibility for each molecule to interact directly with BTMA

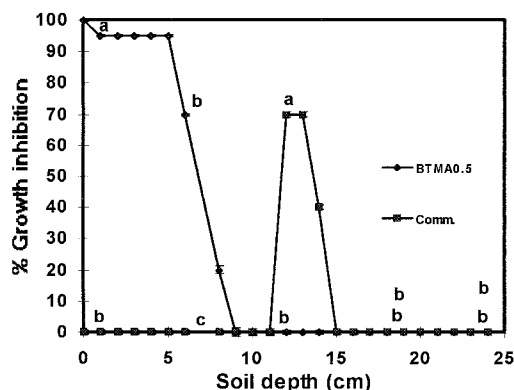


Figure 7. Leaching of commercial EC formulation of alachlor (comm) and Mont-BTMA (0.5 mmol/g of clay) containing alachlor 13% under field conditions using green foxtail as a test plant. Alachlor was applied at 2.0 kg/ha followed by irrigation of 500 m³/ha. Means followed by the same letter at a representative depth are not significantly different at $p = 0.05$ level. Bars indicate standard errors.

molecules on the clay surface. These results are in accord with the adsorption results presented in Figure 3. Higher adsorbed amounts of alachlor were observed with a lower load of the organic cation, BTMA (0.5 mmol/g of clay).

Under field conditions (Figure 7) the Mont-BTMA 0.5 (mmol/g) formulation showed herbicidal activity in the top 0-7 cm of the soil profile, whereas with the commercial EC formulation no herbicidal activity was observed in the top 12 cm and was only observed at a depth of 12-15 cm. These results are in agreement with the results of alachlor leaching under laboratory conditions and indicate that the organo-clay formulation is also stable and active under field conditions. It is evident from the results that the organo-clay-based formulation does not leach as deeply in comparison with the commercial EC formulation. Under field conditions there was less overall leaching than under laboratory conditions (Figures 5 and 6). This behavior under field conditions might be due to several reasons. First, under field conditions, alachlor may move in two directions, horizontal and perpendicular. Second, in addition to the downward movement in water rainfall or irrigation there is an upward movement of the herbicide in response to water evaporation. Third, under field conditions, significant alachlor loss by volatility was reported (Wienhold et al., 1993). The adsorption of alachlor to the organo-clay complex reduces its loss by volatility (El-Nahhal, unpublished). Since water evaporation is more pronounced under field conditions, it reduces the water volume available for downward movement. In most situations, retention of the active ingredient of the herbicide at the topsoil layer will likely result in an improvement of weed control efficacy. From the environmental point of view, the maintenance of the active ingredient of the herbicide in the topsoil layer reduces the hazard of contamination of groundwater.

Phytotoxicity. No differences in the herbicidal activity between all alachlor formulations were observed in the agar test. In this test system, environmental factors, such as adsorption and leaching did not restrict herbicide availability. The response of the test plants to different concentrations of alachlor was similar with all formulations (data not shown).

The response of the test plant (green foxtail) to different levels of several formulations of alachlor

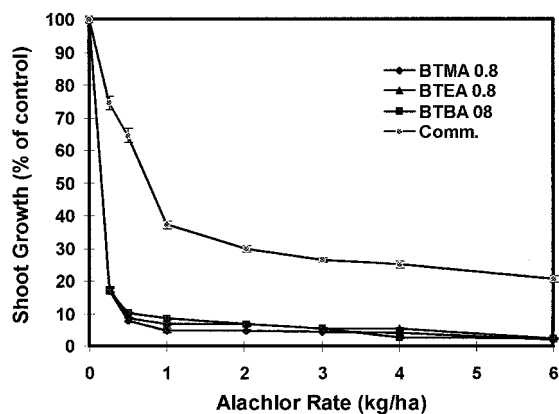


Figure 8. Response of green foxtail to different formulations of alachlor applied preemergence. Shoot fresh weight of the pot grown plants was determined 2 weeks after treatment. Bars indicate standard errors.

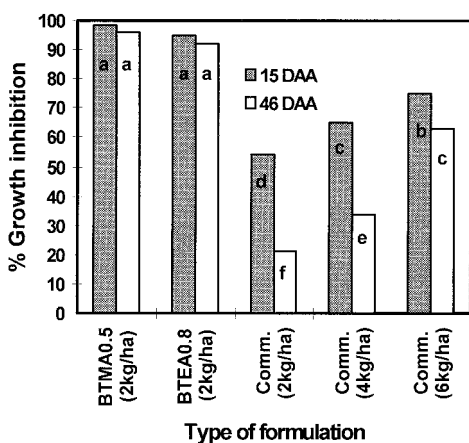


Figure 9. Effects of alachlor formulations applied in the field study on green foxtail growth as determined 15 and 46 days after application (DAA). Columns labeled by the same letter are not significantly different at the $p = 0.05$ level.

applied preemergence in pots is shown in Figure 8. Organo-clay formulations of alachlor applied at a lowest rate (0.25 kg/ha), caused severe growth inhibition (80%) of the test plant, whereas the commercial EC formulation was much less effective and caused only 25% growth inhibition. Higher rates of the organo-clay formulations enhanced growth inhibition only slightly above that obtained with 0.5 kg ai/ha. In the case of the commercial EC formulation, the increase in rates appreciably increased growth inhibition. These results are in accord with the results of the leaching experiments (Figures 5 and 6), indicating rapid dissipation of the EC formulation of alachlor from the pot soil, probably due to the sprinkler irrigation. In contrast, in the cases of the organo-clay formulations, alachlor was more slowly released from the complex and was not readily available for leaching. ED_{50} (the rate causing 50% reduction in shoot fresh weight) values interpolated for each treatment (Figure 8) were almost 9-fold smaller for the organo-clay formulations than that of the EC formulation.

Field Experiments. Following irrigation at 500 m³/ha, the formulations containing BTMA 0.5 and BTEA 0.8 mmol/g of clay were found very effective in weed control even 46 days after treatment, whereas the herbicidal activity of the commercial EC formulation was significantly lower. The data in Figure 9 present the results of field experiments in Rehovot. In both

organo-clay formulations alachlor was at load of 13% ai (w/w). The herbicidal activity of clay-BTMA 0.5-alachlor and clay-BTEA 0.8-alachlor resulted in 97 and 93% growth inhibition as determined by shoot fresh weight, in accord with the results obtained by visual rating. The high efficacy of the organo-clay formulations was similarly observed in the Gaza site (data not shown).

No phytotoxic effects were observed in the treatments employing organo-clay complexes without the herbicide (data not shown). It is evident from the results that the organo-clay formulations, unlike the EC formulation, gave excellent weed control for an extended period of time. A possible explanation for these results is the slow release of the active ingredient from the organo-clay complex into the topsoil layer.

CONCLUSIONS

This study presents an attempt to reduce the leaching and migration in soil of hydrophobic herbicides, such as alachlor. Our approach for designing clay-based formulations combined two principles:

a. The first is modification of the clay surface from hydrophilic to hydrophobic by preadsorbing to it an organic cation whose structure would enable optimal interactions between the herbicide molecules and the surface sites of the organo-clay complexes.

b. Second, the degree of coverage of the clay surface by the organic cation should be optimized. Loading the clay up to its CEC with the organic cation does not necessarily imply optimal interactions between the herbicide molecules and those of the organo-clay complex.

The amounts of alachlor adsorbed on the organo-clay complexes followed the sequence BTMA 0.5 (mmol/g) > BTMA 0.8 > BTEA 0.8 > BTEA 0.5 > HDTMA 0.5 >> Mont. The effectiveness of alachlor formulations depended on the size, type, and load of the organic cation preadsorbed on the clay surface. Alachlor leaching was reduced when formulated with organo-clay complexes according to the sequence of its adsorption. Our data indicate that the reduction in alachlor leaching was more pronounced in formulations containing a smaller organic cation, i.e., in the order BTMA > BTEA > BTBA, which also follows the pattern of shifts of infrared peaks of alachlor (Figure 4). The organo-clay formulations of alachlor enabled its slow release into the soil solution. The application of the new organo-clay formulations of alachlor may enable significant reduction in the applied rates, relative to those of the commercial EC formulation of the herbicide. These characteristics make the new formulations of alachlor more acceptable environmentally and more cost-effective.

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