Fate of 3,6-Dichloropicolinic Acid in Soils

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This study in applied research under field conditions determined the fate of the experimental herbicide 3,6-dichloropicolinic acid (DCP) in three alkaline soils (a black chernozem, brown chernozem, and an ortho eutric brunisol) in Alberta, Canada. The herbicide was found to be degraded mainly by microbial means. Degradation rates were fastest in moist soils ($t_{1/2}$ approximately 2 months) and were inversely related to the organic carbon content of the soil. The rate of degradation was greatly reduced during both dry and cold periods. Furthermore, after having been in the soil over a winter period, the herbicide gave degradation rates that were further greatly retarded. Thus, although 35% of DCP degraded during the first summer period (the first 3 months after application), only a further 31% degraded during the following 12 months. The extent of leaching of the herbicide was also inversely related to the organic carbon content of the soil, being most pronounced in the brown chernozem and brunisol soils. Predictions for depth of leaching under field conditions, based on a chromatographic model of leaching, were reasonably accurate for the chernozem soils. Adsorption of the herbicide was directly related to the organic carbon content of the soil, thus explaining the relative order of leaching observed. Adsorption distribution coefficients (K_d) were obtained by two independent methods—column leaching and batch-type equilibration of a 1:1 soil/solution slurry—the latter yielding the lower values of 0.12, 0.01, and 0.05 μg DCP/g of soil for the black chernozem, brown chernozem, and brunisol, respectively.

The experimental herbicide 3,6-dichloropicolinic acid (DCP), formulated as DOWCO 290 (trademark of the Dow Chemical Co.), has been introduced by the Dow Chemical Co. for the control of phenoxy-tolerant brush, woody rangeland, and deep-rooted perennial broadleaved species. A similar compound, 4-amino-3,5,6-trichloropicolinic acid (picloram), has been in use for such control but its phytotoxicity and relatively long persistence [half-life varies from 1 month to 4 years (National Research Council, 1974)] has led to some environmental damage. As a result, use of picloram in Canada has been confined mainly to rights-of-way along utility lines, pipelines, highways, and railways, and to limited spot treatment in croplands. It was the desire to use the phytotoxic properties of picloram on a wider scale in croplands that led to the introduction of DCP. This herbicide displays essentially the same range of phytotoxicity as picloram. According to the work of Naik et al. (1972) DCP is also as persistent as picloram but less phytotoxic. Extensive testing of the herbicide by the Dow Chemical Co. is still in progress. In trials so far (Dow Chemical Co., 1974) plant species that have shown susceptibility to DCP include Canada thistle, buckwheats, and knapweeds, as well as brush species such as velvet mesquite and whitethorn. It is relatively nontoxic to animal species as also is picloram. Small grains, corn, sorghum, and flax were found to be highly tolerant to the herbicide, and limited degradation studies at Davis, Calif. (Dow Chemical Co., 1974) indicated a half-life one-quarter of that for picloram. The present research was aimed at testing the fate of this herbicide under conditions in which it is expected to be used, i.e., in alkaline soils under the cold temperate climate of the western Canadian prairie provinces. The need for such local testing is becoming increasingly important since persistence data obtained in Europe or the southern states of America do not apply to the cool summers and cold, dry winter conditions prevalent in western Canada. Specifically, the research involved measuring the overall disappearance of DCP in three soils at two different locations in southwestern Alberta. The extent of leaching and strength of adsorption were also measured. Further experiments were performed to determine whether the degradation of the herbicide was a microbial and/or a chemical process and whether any metabolites accumulated during the degradation process.

EXPERIMENTAL SECTION

Apparatus. (a) Gas chromatograph (GC): Hewlett Packard, Model 402 equipped with a ^{63}Ni electron capture detector, 6 ft × $^1/_8$ in. i.d. glass column, packed with 3% OV-17 on 80–100 mesh chromosorb W(HP). Operating conditions: injector block, column, and detector temperatures of 225, 185, and 240 °C, respectively; pulse mode of 150 μs; carrier gas flow of 5% methane/argon at 90 mL/min; electrometer range and attenuation at 1 and 32, respectively. Retention time of 3,6-dichloropicolinic acid methyl ester is 2.8 min. (b) UV-VIS spectrophotometer: Varian Techtron, Model 635 used in the concentration mode with 1 cm cells. (c) GC–mass spectrometer (GC–MS): Finnigan, Model 1015 equipped with a 100 ft OV-17 capillary column coupled directly into the ionization chamber. Ionizing current of 450 μamps at an energy of 70 eV. Column temperature was 170 °C.

Reagents. (a) DCP (formulation DOWCO 290, M-3972), active ingredient 3,6-dichloropicolinic acid as the monoethanolamine salt (analyzed at 2.6 lb/gal acid equivalent), (b) 3,6-dichloropicolinic acid methyl ester, 99+% (both chemicals supplied by Dow Chemical of Canada, Ltd., Sarnia, Ontario, Canada).

Experimental Procedures. *Overall Disappearance Study.* This experiment served to determine the disappearance of DCP from the surface layers of three typical Alberta soils. The soils were selected from regions of the province generally designated as having agricultural soils commonly known as loam and sandy loam, and a forest soil from a mountain valley in the eastern slopes of the Rocky Mountains bordering the great plains. Analyses of the particular soil samples used in the study (Table I) showed that the soils were respectively a black chernozem, brown chernozem, and ortho eutric brunisol according to

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Table I. Characteristics of Soils Used in Persistence Studies for DCP Herbicide

Soil subgroup[a]	Source of soil sample[b]	Local designation[c]	pH[d]	Field capacity, %	Texture[e]			% organic carbon[f]
					% clay	% silt	% sand	
Black chernozem	Balzac	Loam	7.7	30.2	10.5	71.7	17.8	4.8
Brown chernozem	Strathmore	Sandy loam	8.1	21.3	2.7	8.7	88.6	0.68
Ortho eutric brunisol	Kananaskis	Luvisol	7.2	31.3	10.5	47.2	40.8	1.3

[a] Identification after Canada Department of Agriculture. [b] See map in Figure 1. [c] Generalized soil maps and local usage frequently employ these terms for the localities from which the samples were taken; by textural analysis the soils actually used were, respectively, a silt loam, a sand, and a loam. [d] Determined on a 1:2 soil/water slurry. [e] Determined by sieve and pipet analysis (Department of Geology, University of Calgary). [f] Determined by automated carbon analyzer (Institute of Petroleum and Sedimentary Geology, Calgary).

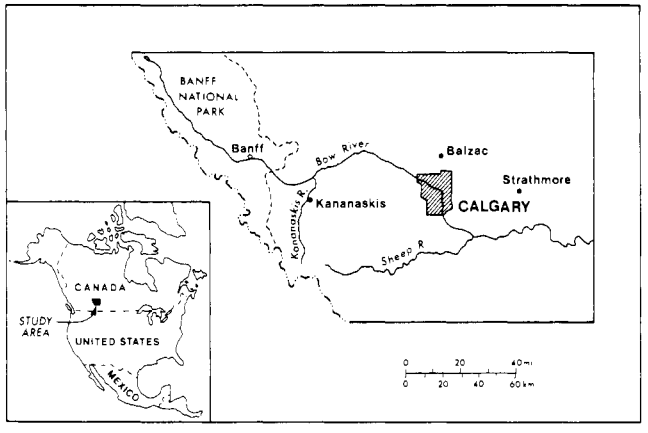

Figure 1. Key map showing the source of the samples of soil and the locale of the field experiments.

the identification scheme used by the Canada Department of Agriculture (1974). Figure 1 shows the geographic positions of the localities involved.

Experiments began on July 4, 1974 and continued until September 11, 1975 comprising two summers and one winter period. Disappearance rates were measured by means of a modified microplot method. Clay pots, 18 cm high with top and bottom diameters of 20 and 13 cm, respectively, and having a 2-cm drainage hole, were filled with a 2-cm layer of gravel followed by 15 cm of soil. These pots were placed in the ground so that the soil surface inside the pot was level with the surface of the ground. The experiment was conducted at two locations. Pots with DCP applied at the rates of 1.9 and 0.95 kg/ha (2.70 and 1.35 mg/pot) on the Kananaskis brunisol and Strathmore brown chernozem and 1.9 kg/ha on the Balzac black chernozem were placed at the Environmental Sciences Centre (Kananaskis) located 82 km west of Calgary. Pots with 0.95 kg/ha of DCP on the Balzac black chernozem were stationed at a farm at Balzac some 8 km north of Calgary. The pot method was used so that a comparison could be made between disappearance rates of DCP in different soils subject to the same climatic conditions (Kananaskis) and two locations were used so that the effect of differing climatic conditions (Kananaskis vs. Balzac) might be observed.

On July 4th, 1974, after allowing the soils to stabilize in the pots for 2 weeks, DCP was applied as a 1-mL aqueous solution of the formulation. The 1-mL application was spotted over the central 13-cm diameter area of the soil in the pot by means of a pipet and incorporated into the top 2 cm of soil by careful manipulation with a fork. Only the central area was so treated (the area equivalent to the bottom diameter of the pot) so that downward movement of the herbicide would not be inhibited by contact with the sloping walls of the pot where adsorption and/or channelling could occur.

Pots were taken out of the ground to be analyzed after intervals of 0, 2, 4, 8, 12, and 16 weeks at which point winter set in, freezing the pots into the ground. Sampling of the black chernozem soil pots continued after the winter (from May 1, 1975 until September 11, 1975). The herbicide was no longer present in the brown chernozem or brunisol soils and their sampling was discontinued. All samples were stored at −20 °C while awaiting analysis (up to 6 months).

To prepare samples for analysis, the soil from each pot was thawed and spread out to dry at room temperature. Thorough mixing of the soil was performed by crushing it to pass through a no. 7 mesh sieve (2.83 mm), filtering it through a stemless metal funnel, and passing it through the sieve once more. The amounts of soil averaged at 2920 g for the black chernozem, 4050 g for brown chernozem, and 2780 g for brunisol. Samples of approximately 100 mg were taken from the mixed soil to be crushed and analyzed by the electron-capture gas chromatographic method described in a previous publication (Pik and Hodgson, 1976).

Leaching Study. DCP has a pK_a of 2.33 (Dow Chemical Co., 1974) and hence exists in the anion form in most soils. The mobility of the herbicide was therefore expected to be considerable. The extent of leaching of the herbicide was determined in each of the three soils under the same climatic conditions. Thus, three metal cylinders, 92 cm high by 19 cm in diameter and partitioned by metal sieves 54 cm from the top, were filled with 2 cm of gravel, followed by 50 cm of soil (one type per cylinder). To collect the drainage water, a funnel leading into a 1.9-L jar was sealed in below the sieve and the whole cylinder was loosely capped at the bottom. These cylinders were placed in the ground at Kananaskis to a depth such that the soil in the cylinder was level with the surface of the ground.

On July 8, 1975, after the soils had stabilized in the cylinders for 2 weeks, 1.35 mg of DCP was applied as a 1-mL aqueous solution of the formulation to give a rate of 0.95 kg/ha. The cylinders were left in the ground for a period of 9 weeks after which the soil in each cylinder was sectioned into four 12.5-cm segments for analysis in duplicate. Water that had collected in the drainage traps (1, 5, and 150 mL for black chernozem, brown chernozem, and brunisol, respectively) and the gravel layer were also analyzed. Rainfall and temperature data collected by the weather station at Kananaskis were recorded for the test period.

Adsorption Study. Adsorption coefficients, also known as distribution coefficients (K_d), were determined for DCP on each of the three soils by two different methods. Batch-type equilibration is the most commonly used method; the second method, column leaching, was introduced by Swoboda and Thomas (1968).

(a) Column Leaching. Three glass columns, 1.1 cm^2 in cross sectional area by 31 cm in length and fitted with a Teflon stopcock, were filled with each of the three air-dried

soils to a depth of 15 cm. The soils, originally passed through a 2-mm sieve, were packed to give bulk densities (ρ) of 0.9, 1.4, and 1.1 g/cm^3 for the black chernozem, brown chernozem, and brunisol, respectively. The soils were slowly saturated with water from bottom to top, expelling all air bubbles and thus allowing for the void volume (V_v) to be calculated as 8.1, 6.0, and 7.2 cm^3 for the three soils, respectively. One milliliter of a 25-ppm aqueous DCP solution of the formulation was added to duplicate columns of each of the soils giving a 1.9 kg/ha application rate, and a 2-mL aliquot of leachate was immediately collected. The column was then leached with water, under a varying 10- to 8-cm head, and the leachate was collected in 2-mL aliquots until all the herbicide had passed through the column. Flow rates averaged 0.5 mL/min.

The aliquots collected were acidified with 8.5% (v/v) phosphoric acid (0.5 mL) and extracted with 5% (v/v) ethanol/chloroform (4 mL). The organic phase was shaken with sodium sulfate (1 g) for drying and removal of emulsions, after which the DCP was transferred into 0.1 N NaOH (4 mL). The amount of DCP in the eluate was subsequently determined by measuring the absorbance at 280 nm using the Varian Techtron (Model 635) UV-VIS spectrophotometer with 1-cm cells. The concentration slope for DCP absorption was established by use of standard solutions which had been subjected to the same extraction procedure. This approach eliminated the need for applying percent extraction corrections to the read-out values, although extraction efficiency was determined at a value of 88.4 ± 0.7%. The absorbance of DCP was linear in the tested range of 0–4 ppm.

(b) Batch-Type. Batch-type adsorption studies were performed by equilibrating 4 g of soil with 4 mL of an aqueous 1.19 ppm solution of DCP formulation for a period of 10 h at room temperature (21 °C). After centrifuging, the supernatant liquid was drawn off and recentrifuged. One milliliter of this supernatant fraction was diluted to 4 mL with 1 N NaCl, and the solution was analyzed in the same way as the soil extracts. Duplicate equilibrations were performed for each soil and the amount of DCP adsorbed by the soil was taken to be equivalent to the decrease in the solution concentration.

Degradation Study. This study observed the degradation of DCP in natural and sterilized soils under constant moisture content conditions. An enriched soil culture experiment was also performed to monitor the degradation pathway of the herbicide.

(a) Degradation in Natural Soil. Degradation rates of DCP were determined for each of the three soils by filling 1.9-L jars with 1 kg of soil, bringing each to 70% field capacity with water, and applying the herbicide at the rate of 1.9 kg/ha (1.47 mg/jar). The jars were loosely capped and placed in the ground on July 8, 1975 at Kananaskis so that the soil inside the jar was level with the ground surface. The air in the jars was periodically renewed by removing the lid for several minutes each week. After 9 weeks the jars were removed and the soils analyzed in duplicate for DCP residues. These experimental procedures were deemed appropriate although they may have tended to promote more anaerobic conditions than expected in the natural soils profile, especially in the case of the sterilized samples.

(b) Degradation in Sterilized Soil. In the summer of 1974, two series of jars containing sterilized loam soil (sterilization by autoclaving at 121 °C (20 psi steam) for 12 h) were brought to 70 and 100% field capacity for water and treated with 1.9 kg/ha of DCP. The jars were sealed with canning lids to keep the soil sterile and placed in the ground at Kananaskis. A previous test had shown that soil treated in this way would remain sterile for at least 2 weeks. Jars from each of the two series were removed periodically over a period of 14 months for analysis in duplicate.

(c) Degradation in a Soil Enriched Culture Medium. To examine the degradation pathway of DCP, a series of culture tubes containing 0.5 g of black chernozem soil and 4 mL of a culture medium were set up. The culture medium, as first used by Naik et al. (1972), contained 20 ppm each of Difco yeast extract, Oxoid peptone, and glucose; 4 ppm $(NH_4)_2SO_4$; and 180 ppm KH_2PO_4. The medium was adjusted to pH 7.0 with KOH.

Two milligrams of DCP in 1 mL of a neutral aqueous solution were added to each of the tubes as well as to four control tubes containing sterilized medium but no soil. The tubes were slowly shaken in a water bath maintained at 25 °C. Tubes were examined periodically by diluting 1 mL of the centrifuged solution to 50 mL with water and scanning the absorbance from 350 to 200 nm in 1-cm cells. Such a scan allowed for determination of the concentration of the herbicide and detection of any UV-absorbing metabolites as well.

The sample from day 72 of the experiment was also analyzed for both neutral and acidic metabolites by electron-capture GC and by GC–MS. One milliliter of the centrifuged solution was extracted directly with 5% ethanol/chloroform, methylated with CH_2N_2, and analyzed. A further 1-mL aliquot was first acidified with 8.5% phosphoric acid, and the acidified solution was then extracted with 5% ethanol/chloroform, methylated, and analyzed as before. The electron-capture GC analyses were performed under the same conditions as the DCP analyses, except that the column temperature in several runs was lowered to 170 °C in case any metabolites more volatile than DCP were present. Mass scans were recorded from 50 to 350 amu. Total respiration rates in both a control tube and the 96-day sample were measured on a Gilson differential respirometer in order to determine the extent of microbial activity in the experiment to that time.

(d) Synthesis of a Possible Metabolite. One of the possible metabolites of DCP, 3-chloro-6-hydroxypicolinic acid, was synthesized by refluxing DCP with 10% NaOH for 15 h. The cooled solution was acidified with sulfuric acid to pH 1, filtered, and the product redissolved in 2% NaOH. The product was then precipitated from solution by adjusting the pH to 1 with hydrochloric acid. The precipitate was collected and dried overnight in a dessicator. Total yield of the white, flaky product was 98.1 mg or 22.9%. A solution of 1 mg of this material was subsequently methylated with CH_2N_2 and subjected to both electron-capture GC and GC–MS analysis.

RESULTS

Overall Disappearance Study. The disappearance rates of DCP which were obtained for each of the soils at the two different locations, together with the cumulative rainfall data for the test period are graphed in Figures 2 and 3. The graphs clearly show that disappearance of the herbicide at the Kananaskis location was rapid in the brown chernozem and brunisol (8–12 weeks) and slower in the black chernozem (55 weeks). Disappearance rates in all samples at the one location were independent of the amount of herbicide applied (1.9 vs. 0.95 kg/ha). However, disappearance rates were dependent on soil type: the disappearance rate in the brown chernozem was slightly faster than in the brunisol, but much faster than in the black chernozem at either location (more than 61 weeks).

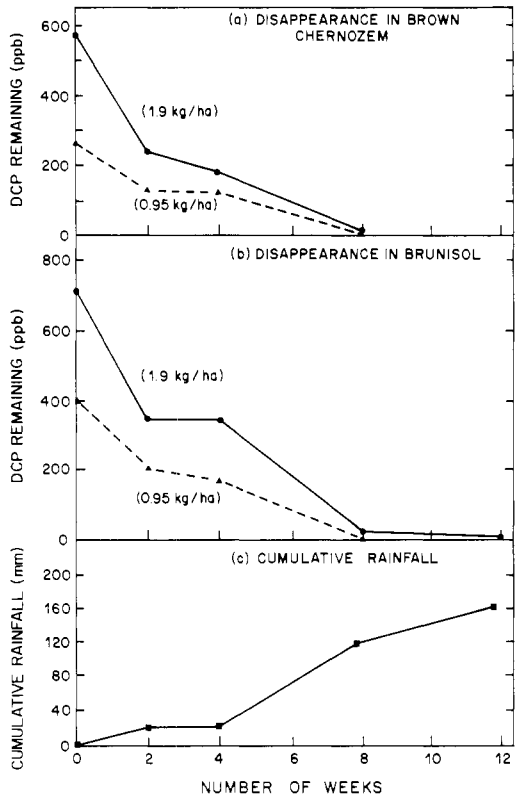

Figure 2. Disappearance rates of DCP in soils and cumulative rainfall at Kananaskis from July 4 to September 26, 1974. Initial application rates were at 1.9 and 0.95 kg/ha on both brown chernozem and brunisol as shown.

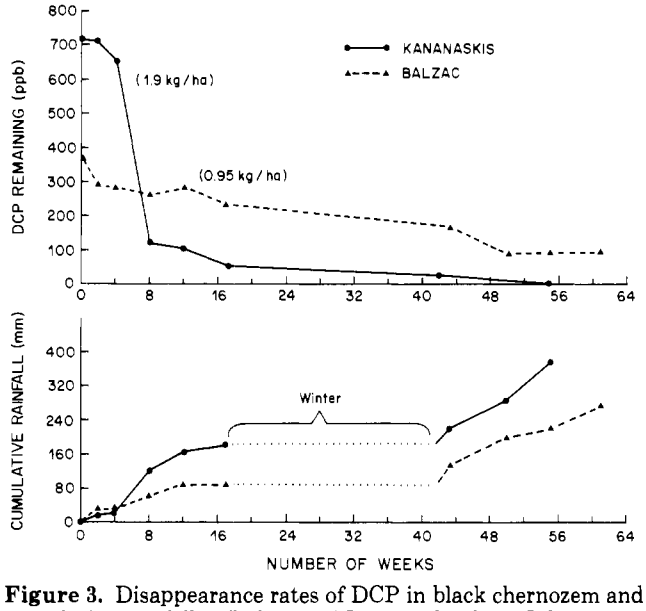

Figure 3. Disappearance rates of DCP in black chernozem and cumulative rainfall at Balzac and Kananaskis from July 4, 1974 to September 11, 1975. Initial application rates were 1.9 kg/ha at Kananaskis and 0.95 kg/ha at Balzac.

Differing climatic conditions had an even greater effect on disappearance rates. Thus, although the difference in DCP dosages of 1.9 and 0.95 kg/ha had no effect on disappearance rates in the brown chernozem and brunisol stationed at the same location (Kananaskis), the difference in dosages did affect disappearance rates in the black chernozem series at the two different locations. At Balzac, 25.8% of the DCP applied at only 0.95 kg/ha on black chernozem had still not disappeared after 61 weeks, while at Kananaskis a 1.9 kg/ha dosage of DCP on black

Table II. Climatological Data for Kananaskis from July 8 to September 11, 1975, during which the Field Leaching Study Was Performed

Week	Rainfall, mm	Temperature, °C	
		Mean max.	Mean min.
1	24.1	25.0	10.5
2	2.3	20.0	9.7
3	37.6	25.0	9.7
4	5.1	16.8	6.5
5	2.5	18.9	4.1
6	32.0	17.9	6.0
7	6.6	16.8	4.7
8	6.6	16.2	4.8
9	2.5	18.9	1.7
Total	119.3	19.5 (Av)	6.4 (Av)

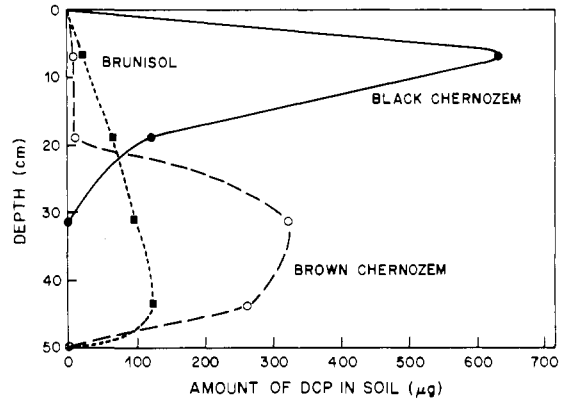

Figure 4. Concentration profiles of DCP remaining in soils after a period of 9 weeks during which 119 mm of rainfall occurred. The herbicide was applied on July 8, 1975 at the rate of 0.95 kg/ha. The three curves are arbitrarily extended to the zero concentration level at zero depth.

chernozem had completely disappeared after only 55 weeks.

It is also evident that during the winter the rate of loss was slowed. Furthermore, as particularly noticeable at Balzac, the herbicide disappeared more slowly after having been in the soil over a winter than it did immediately following the time of its original application. Thus, despite the 1975 spring snow run-off and a total rainfall of 185 mm during the second summer, 39.9% of the DCP present when the winter set in still remained in the soil at the end of the second summer (September, 1975). Overall then, in the Balzac area with its low rainfall, DCP remained in the black chernozem to the extent of 25.8% over a period of some 14 months; at Kananaskis, where rainfall was higher than at Balzac, the herbicide had effectively disappeared from the top soil by the end of the first summer season.

An interesting side result was obtained at the Balzac site during the second summer period (1975). Although DCP was still present in the black chernozem at a concentration of 110 ppb (0.36 kg/ha), various weed species were found to be actively growing in the soil. The species all belonged to the general class of herbaceous annual dicots and were identified as shepardspurse (*Capsell bursa-pastoris* (L.) Medic), lambsquarter (*Chenopodium album* L.), *Atriplex* sp., and tansymustard (*Descurainia sophia* (L.) Prantl). Although there was some evidence of stem elongation (indicating some uptake of DCP), the plants grew healthily and remained alive over the whole summer period.

Leaching Study. Weekly temperature and rainfall data are recorded in Table II. Mean maximum and minimum temperatures for the 9-week period during which the study

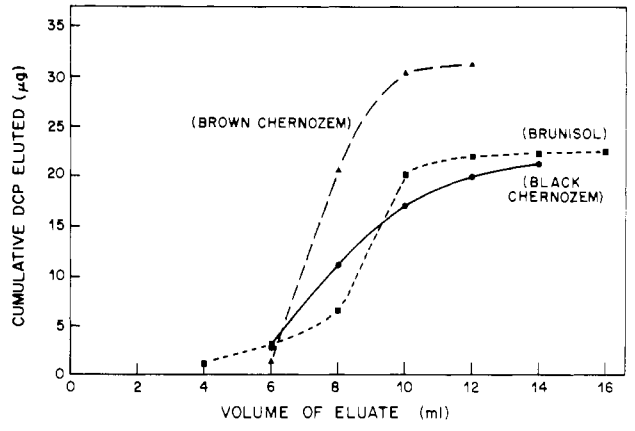

Figure 5. Elution curves of DCP from soil columns, 15 cm high and 0.7 cm in diameter. Application at 1.9 kg/ha. Flow rates averaged 0.5 mL/min.

was conducted were 19.5 and 6.4 °C, respectively. Total rainfall for the same period amounted to 119.3 mm.

The results of the leaching experiment are collected in Figure 4 which depicts the concentration profiles of DCP remaining in the soil columns 9 weeks after application of the herbicide at 0.95 kg/ha. The graphs clearly show that penetration of DCP was greatest in the brown chernozem and brunisol (approximately 45 cm) and least in the black chernozem (approximately 20 cm). In both the brown and black chernozem the herbicide had moved downward in a fairly distinct band. In the brunisol, however, the herbicide had been partially retained as it spread down through the soil profile. The drainage traps were all free of DCP, indicating that no herbicide had completely leached through any column. Therefore, the disappearance rates as measured are a true indication of the rate of degradation. Thus over a 9-week summer period at Kananaskis, with 119.3 mm of rainfall and mean maximum and minimum temperatures of 19.5 and 6.4 °C, respectively, the amounts of DCP degraded in various soils were: black chernozem, 45%; brown chernozem, 56%; and brunisol (with reservations as noted below), 77%. These rates were determined in samples with an initial DCP application of 0.95 kg/ha. Unfortunately, considerable activity of ants occurred in the brunisol column, so that some displacement of herbicide-bearing soil may have occurred both within and out of that column.

Adsorption Study. The results of the adsorption study in terms of the column leaching experiments are presented in Figure 5 which shows the cumulative amounts of DCP eluted from the column as a function of the eluate volume. Plotted in this way the graph allows for calculation of the volume (V_p) of effluent required to elute one-half of the solute (DCP) through the column. Recoveries of DCP eluted were generally good except for the leaching of the brown chernozem column which indicated that an interfering soil constituent was apparently also eluted. A somewhat higher value thus resulted for this column. After V_p determinations were made, K_d values were calculated according to the equation of Swoboda and Thomas (1968):

$$K_d = [(V_p/V_v) - 1](V_p/W) \quad (1)$$

where W is the weight of the soil in the column. The calculated K_d values from the column leaching study show that adsorption of DCP was greatest on the black chernozem (0.38 mL/g), least on the brown chernozem (0.06 mL/g), and intermediate on the brunisol (0.11 mL/g). (Larger values indicate larger volume of water required to leach the herbicide through the soil and hence indicate stronger adsorption.)

Table III. Adsorption Equilibrium of DCP Expressed in Various Ways[a]

Soil	Org. C content, %	Concentration on solid, μg/g	K_d, mL/g	Percent adsorp.
Black chernozem	4.8	0.12	0.10	9.6
Brown chernozem	0.68	0.01	0.01	0.8
Ortho eutric brunisol	1.3	0.05	0.04	4.1

[a] Determined by batch-type equilibrations at 21 °C using a 1:1 soil/solution ratio at an original concentration of 1.19 ppm.

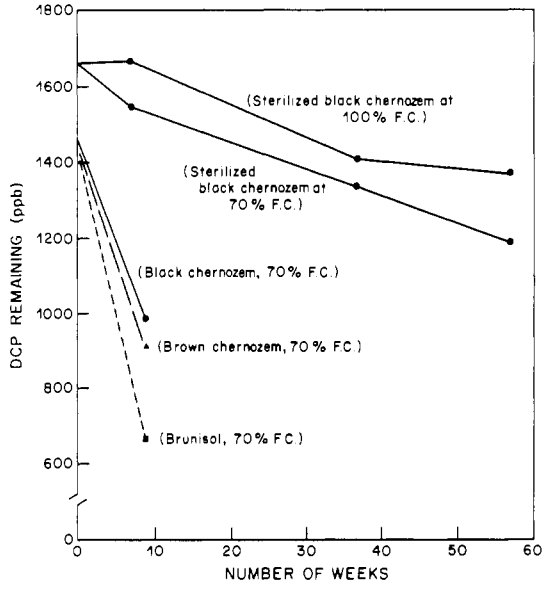

Figure 6. Degradation rates of DCP in natural and sterilized soils at Kananaskis. Initial application of the herbicide was 1.9 kg/ha.

Results from the batch-type adsorption experiment are shown in Table III. Again they show that adsorption of DCP on the soils was low. Adsorption values are quoted in three ways for easy comparison: in their usual way as μg of DCP/g of soil (μg/g), as a percentage of DCP adsorbed (%), and in terms of the units used in the column leaching experiment, i.e., mL of effluent/g of soil (mL/g). K_d in units of μg/g were 0.12, 0.01, and 0.05 for adsorption of DCP on the black chernozem, brown chernozem, and brunisol, respectively. The inherent error in analysis of ±1% does of course mean that the low value of K_d for adsorption on the brown chernozem may be of limited significance at best since the value was obtained by difference.

Degradation Study. (*a*) *Degradation in Natural Soils.* As illustrated in Figure 6, degradation of DCP was fairly rapid in the moist (70% field capacity) soils. The amounts that were degraded after 9 weeks amounted to 30, 38, and 55% for the black chernozem, brown chernozem, and brunisol, respectively, again indicating that degradation occurs most rapidly in the brunisol soil.

(*b*) *Degradation in Sterilized Soil.* In the sterilized black chernozem degradation rates were greatly retarded (Figure 5). Moisture content of the soil seemed to play a role. Thus, over a 57-week period, 28% of DCP was

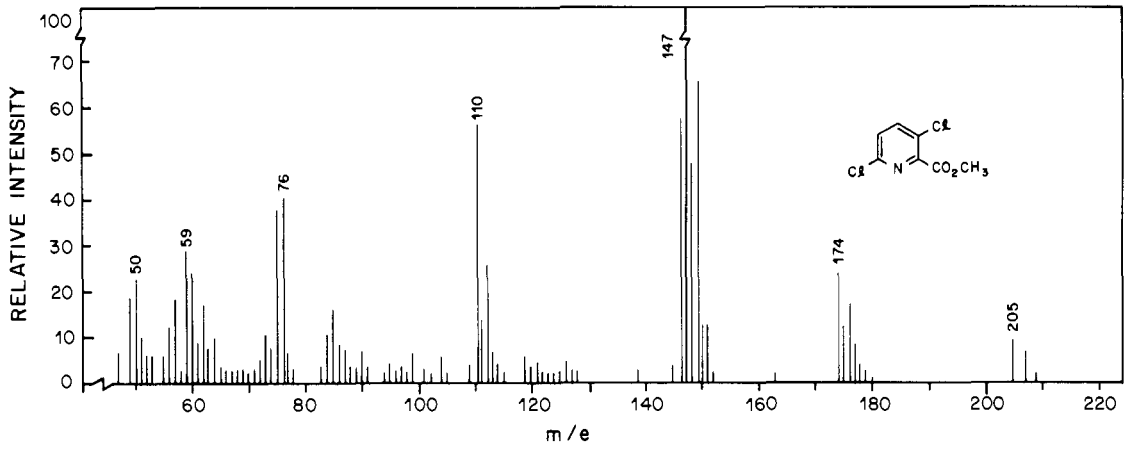

Figure 7. Mass spectrum of DCP methyl ester at an ionizing energy of 70 eV.

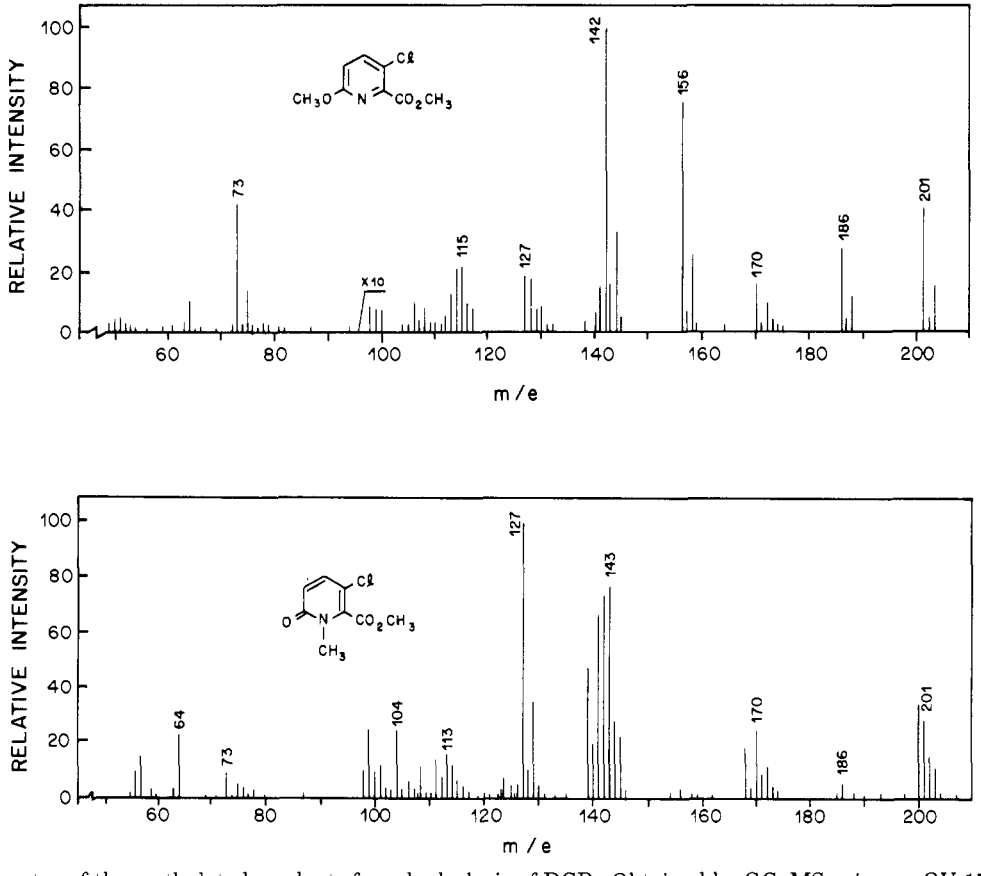

Figure 8. Mass spectra of the methylated products from hydrolysis of DCP. Obtained by GC–MS using an OV-17, 100-ft capillary column held at 170 °C and an ionizing energy of 70 eV.

degraded in the series held at 70% field capacity while only 18% was degraded in the 100% field capacity series. Nevertheless, both rates were substantially lower than that occuring in the natural black chernozem.

(*c*) *Degradation in a Soil Enriched Culture Medium.* The degradation in the culture medium was also slow, with only 15% of the DCP degrading over a 96-day period. A somewhat lower amount (11%) had degraded in the control tubes during the same time period. Although biweekly analyses were performed, no metabolites were detected by UV, electron-capture GC, or GC–MS. Figure 7 depicts the mass spectrum of DCP (methylated) that was left in the sample after 96 days. The spectrum shows the molecular ion at *m/e* 205 with the typical dichloro isotopic distribution patterns at *m/e* 205, 207, 209; *m/e* 174, 176, 178 (loss of OCH_3), and *m/e* 147, 149, 151 (loss of CO_2CH_2). The base peak occurs at *m/e* 147 and a loss of 95 amu is evident at *m/e* 110 (loss of HCl from *m/e* 146). That only one chlorine atom was left on the fragment at *m/e* 110 was confirmed by the isotope peak at *m/e* 112. The occurrence of the base peak at *m/e* 147 instead of at *m/e* 146 may be attributed to loss of CO_2CH_2 following a McLafferty type rearrangement of a hydrogen/atom from the methyl of the leaving group to the nitrogen of the pyridine ring. Such a loss was also observed for picloram methyl ester (National Research Council, 1974).

The total respiration rates in control and natural samples were found to be 3.0 and 9.0 μL of O_2 taken up (mL of solution)$^{-1}$ h^{-1} at 25 °C, respectively. These results indicate that the control was not completely sterile and that a low measure of microbial activity occurred in the soil-amended culture medium.

(*d*) *Synthesis of a Possible Metabolite.* The hydrolysis of DCP produced two products as revealed by electron-

Table IV. Prediced and Experimental Depths of Leaching of DCP in Three Soils Occurring as the Result of a Total Rainfall of 119 mm in the Field at Kananaskis

Soil	Bulk density ρ, g/cm^3	Void[a] fraction, θ	Density of[b] soil solids, d_s, g/cm^3	Experimental K_d		Observed[c] depth of leaching, cm	Calculated[d] depth of leaching, cm		Calculated[e] depth of leaching, cm	
				Column	Batch		Column	Batch	Column	Batch
Black chernozem	0.9	0.491	1.77	0.38	0.10	7	14.3	20.5	13.6	17.3
Brown chernozem	1.4	0.364	2.20	0.06	0.01	32	26.6	31.6	22.1	22.9
Ortho eutric brunisol	1.1	0.436	1.95	0.11	0.04	43	21.4	24.9	17.9	19.6

[a] Void fraction, θ = void volume/soil volume. [b] Density of soil solids, $d_s = \rho/1\text{-}\theta$. [c] Determined as the depth to maximum concentration, from the curves of Figure 4. [d] Depth of leaching, cm = $R/\rho K_d + \theta$, where R is total rainfall, 11.9 cm. [e] Depth of leaching, cm = $R/[\theta^{2/3}(1 - K_d d_s) + K_d d_s]$.

capture GC and GC–MS. Both products had a molecular weight of 201 amu and characteristic monochloro isotope clusters as shown in Figure 8. A small amount (0.01%) of unreacted DCP also remained. The products were tentatively identified by their mass spectrum as the 6-keto-*N*-methyl and the 6-methoxy derivatives of DCP. The methylated compounds had retention times of 1.9 and 5.4 min, respectively, compared with that for DCP methyl ester of 2.8 min. These retention times apply to separation on the 6-ft 3% OV-17 column.

DISCUSSION

The anticipated usage pattern for DCP allows for crop planting 1 year after application of the herbicide (Dow Chemical Co., 1974). Based on the disappearance rates observed in this study such a usage pattern would be appropriate for application on brown chernozems and brunisol soils but not necessarily for application on black chernozems, especially in drier areas such as Balzac. In Balzac the herbicide degraded slowly and apparently became strongly adsorbed over a winter period, making it unavailable for microbial degradation. The subsequent growth of weed species observed in the black soil at Balzac suggests that the herbicide was also no longer available for plant uptake. Nevertheless, the fact that the herbicide was still present after 14 months suggests that DCP could pose a persistence problem in loam type agricultural soils under dry conditions and relatively low average yearly temperatures.

The rapid leaching of the herbicide in the brown chernozem and brunisol soils makes DCP appropriate for control of deep-rooted weed species. Some of these species have roots to a depth of 1 or 2 ft so that movement of the herbicide to these depths is desirable. On the other hand, the decrease in microbial populations with increasing depth (Burgess and Raw, 1967) will limit biodegradation. Hence the herbicide may pose problems in groundwater contamination. In farm areas where irrigation water is pumped from the ground the problem could be serious depending on whether or not the groundwater is channelled into localized areas. Predictions on the extent of such leaching under varying rainfall conditions are difficult to make. The results from the column leaching adsorption study did not accurately predict the depth of movement of DCP in the field. Rearranging the equation formulated by Swoboda and Thomas (1968), the depth of movement (L) that occurs under an amount of rainfall (R) is given by:

$$L = R/(K_d\rho + \frac{V_v}{V}) \quad (2)$$

where ρ is the bulk density of the soil and V_v/V is the porosity or void fraction, θ (θ = void volume/soil volume). Alternatively, soil density, d_s, rather than bulk density, ρ, can be utilized as shown by Hamaker (1975). Thus:

$$L = R/\theta^{2/3}(1 - K_d d_s) + K_d d_s \quad (3)$$

where $d_s = \rho/(1 - \theta)$. The depths, L, thus predicted relate to the movement of the maximum concentration of the herbicide as opposed to the front of movement. Values of K_d obtained by both methods (column and batch) were used to solve the equations as shown in Table IV. On comparing these predicted depths to those actually observed in the field leaching experiment (Table IV), it is evident that the predicted values are in reasonable agreement for leaching in the black and brown chernozems but much too low in the brunisol. However, the unusually large amount of water that drained through the brunisol column (150 mL) may indicate that channelling occurred in this column. This could explain the highly skewed nature of leaching observed (Figure 4) which in turn undoubtedly contributes to the poor prediction for this soil. It should also be remembered that the field leaching values represent the average concentration of the herbicide in a 12.5-cm segment represented at its midpoint. Lastly, the simpler equation (2), is seen to provide the closest agreement with experimental results.

The K_d constants obtained by the two methods for adsorption of DCP on the three soils indicate that adsorption of the herbicide was directly related to the organic carbon content of the soil. Both methods yielded the same relative order of adsorption (black chernozem > brunisol > brown chernozem) but the column leaching values were in all cases greater than those obtained by batch-type equilibration by a factor of about 4. These differences are not unexpected, however, since the methods for calculating the values were derived independently of each other. They are therefore not an accurate measure of the same values, especially since such K_d values are extremely dependent on concentration. The concentration of herbicide in a solution passing through a column is continually changing (Swoboda and Thomas, 1968) and even a change in flow rate in the column leaching experiment could have altered the K_d values obtained. Nevertheless, adsorption of DCP is very low and the herbicide may easily fall into mobility class 5 (very mobile, according to the scheme of Helling et al., 1971), along with dicamba and 2,3,6-TBA. According to the Helling scheme, picloram falls in class 4 (mobile). The ready leachability of DCP also indicated that the measured loss of herbicide in the overall disappearance study was due not only to degradation but also to concomitant leaching of the herbicide through the top 15 cm of soil.

The results from the degradation study clearly show that degradation of DCP occurred mainly by microbial means,

as was also shown to be the case for picloram degradation (National Research Council, 1974). The lack of metabolite accumulation (particularly the 6-hydroxy derivative) also confirms the hypothesis of Meikle et al. (1974), that the rate-limiting step in the degradation sequence of picloram is a ring cleavage and a prior hydroxylation at the 6 position is not involved. The slow degradation rate in the soil-amended culture medium over a period of 96 days was unexpected as was the lack of significant microbial activity. The results seem to indicate that DCP itself was not a good energy source for the microbial population which was present, and that the medium was perhaps not enriched enough for cometabolism to be effective. Naik et al. (1972), using the same medium with a fertile garden soil instead of the black chernozem from the present study, found that decomposition of DCP took longer than 160 days. On this basis, the apparent lack of degradation may be explained by the relatively short time period used (96 compared to 160 days). The 96-day period may well have been part of lag period during which an effective microbial population was being built up. Such lag periods are well known for 2,4-D degradation (Hamaker and Thompson, 1972) as well as for other compounds. In fact, Sheets et al. (1968), commenting on the lack of degradation of 2,3,6-TBA, fenac, and methoxy fenac over a period of 80 days in an enrichment system, postulated that the length of the study period was insufficient for adaptation of effective microbial populations. The much slower degradation rate in the soil-enriched culture medium as compared to that in the natural soils can be readily explained by the fact that both the microbial populations and the food sources available to them are much greater in the natural soils than in the culture medium. Also, the concentration of DCP in the natural soils was extremely low (1.5 ppm) compared to the concentration in the culture medium (400 ppm).

The formation of the two methylated products from the initial hydrolysis of DCP can be readily explained by noting that a keto-enol tautomerism of the 6-hydroxy derivative, and 2-pyridinols in general, takes place in solution. Upon subsequent methylation in 10% methanol/chloroform solution of the acidified hydrolysis product, the equilibrium mixture would become effectively fixed into its two isomeric forms giving the 6-methoxy and the *N*-methyl, 6-keto derivatives of DCP. Thus, even though it may be possible to favor the isolation of a 6-hydroxy derivative by careful choice of work-up conditions as was done for picloram by Meikle et al. (1974), it would appear that any subsequent redissolving and methylation of the product would lead to a mixture of isomeric forms. Hence care must be taken to look for both possible products in any methylation/GLC method of determining degradation products of picloram type herbicides. The relative amounts of each isomer formed would depend on both the condition of methylation as well as the nature and position of the other ring substituents.

CONCLUSION

The major conclusion to be drawn from this research is that DCP can be relatively persistent in alkaline soils in Alberta (Canada) and that its mobility through soil is pronounced. It appears that no metabolites accumulate during the degradative process so that no additional contamination of the environment occurs. The degradation that occurs is mainly microbial and the rates are positively correlated with soil moisture content and temperature but inversely with organic matter content of the soil.

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LITERATURE CITED

Burgess, A., Raw, F., Ed., "Soil Biology", Chapter 2, Academic Press, New York, N.Y., 1967.

Canada Department of Agriculture, "The System of Soil Classification for Canada", pub. 1455, 1974.

Dow Chemical Co., "Technical Information on Dowco 290", Midland Mich., 1974.

Hamaker, J. W. in "Environmental Dynamics of Pesticides", Hague, R., Freed, V. H., Ed., Plenum Press, New York, N.Y., 1975, pp 115–133.

Hamaker, J. W., Thompson, J. M., in "Organic Chemicals in the Soil Environment", Goring, C. A. I., Hamaker, J. W., Ed., New York, N.Y., 1972, pp 49–143.

Helling, C. S., Kearney, P. C., Alexander, M., *Adv. Agron.* **23**, 147 (1971).

Meikle, R. W., Youngson, C. R., Hedlund, R. T., Goring, C. A. I., Addinton, W. W., *Weed Sci.* **22**, 263 (1974).

Naik, M. N., Jackson, R. B., Stokes, J., Swaby, R. J., *Soil Biol. Biochem.* **4**, 313 (1972).

National Research Council, Ottawa, Canada, "Picloram: The Effect of Its Use as a Herbicide on Environmental Quality", NRC Publication No. 13684, 1974.

Pik, A. J., Hodgson, G. W., *J. Assoc. Off. Anal. Chem.* **59**, 264 (1976).

Sheets, T. J., Smith, J. W., Kaufman, D. D., *Weed Sci.* **16**, 217 (1968).

Swoboda, A. R., Thomas, G. W., *J. Agric. Food Chem.* **16**, 923 (1968).

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