Spatial variability in the biodegradation rate of the herbicide bentazone with soil depth assessed using contrasting incubation methods

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Abstract

Vertical and horizontal spatial variability in the biodegradation of the herbicide bentazone was compared in sandy-loam soil from an agricultural field using sieved soil, which represents the method most widely used to investigate biodegradation rates, and intact soil cores. An initial experiment compared degradation at 5 depths between 0 and 80 cm using sieved soil. Degradation was shown to follow first order kinetics, and DT50 declined progressively with soil depth from 56 days at 0-10 cm to 520 days at 70-80 cm. DT50 was significantly correlated with organic matter, pH and dehydrogenase. In a subsequent experiment, degradation rate was compared after 127 days in sieved soil and intact cores from 0-10 and 50-60 cm depth from 10 locations across a 160 x 90 m portion of the field. Method of incubation significantly affected mean dissipation rate, although there were relatively small differences in the amount of pesticide remaining in intact cores and sieved soil, ranging between 4.6 and 10.6 %. Spatial variability in degradation rate was higher in soil from 0-10 cm depth relative

to that from 50-60 cm depth in both sieved soil and intact core assessments. Patterns of spatial variability measured using cores and sieved soil were similar at 50-60 cm, but not at 0-10 cm depth. This could reflect loss of environmental context following processing of sieved soil. In particular, moisture content, which was controlled in sieved soil, was found to be variable in cores, and was significantly correlated with degradation rate in intact topsoil cores from 0-10 cm depth.

Keywords: Biodegradation, spatial variability, bentazone, intact core, sieved soil, soil depth

1. Introduction

Management of envivonmental contamination by pesticides requires understanding of pesticide sorption and degradation processes, which determine pesticide motility through the soil profile. However, studies on pesticide biodegradation have typically been conducted in topsoils and much less information is available on pesticide transformation processes in subsoils. Generally, pesticide degradation rates decline with soil depth (Stenrod et al., 2006; Rodriguez Cruz et al., 2006), although exceptions have been reported (Di et al., 1998; Karpouzas et al., 2001). Within the topsoil, there can be significant spatial variability in the biodegradation rates of pesticides, even within fields where the soil appears to be uniform (Bending et al., 2006). Similarly, there can be significant spatial variability of degradation rates within the subsoil, with some reports suggesting that spatial variability in degradation processes may increase with depth, reflecting greater spatial variability in the distribution of microbial communities (Rodriguez Cruz et al., 2006).

Studies of pesticide fate in soil are usually conducted using samples which have been air-dried, sieved and homogenised following collection from the field, and which are maintained under controlled moisture and temperature conditions during experimentation in the laboratory (Beulke et al., 2005). However, the sampling and handling of soil in this way may irreversibly alter the physical, chemical and biological properties of the soil relative to field conditions (Topp et al., 1994; Beulke et al., 2005) and may influence both pesticide availability and microbial activity, thereby affecting pesticide degradation (Beulke et al., 2005). This is especially important to consider for the subsoil, which is never disturbed by tillage (Fomsgaard et al., 1998).

The sieving and air-drying of soil could lead to increased pesticide-soil contact which could potentially have a range of counteracting affects on biodegradation. One possible effect of sieving could be to enhance biodegradation by increasing homogeneity of distribution of pesticide through the soil, and thereby increasing contact between the pesticide and degrader microorganisms (Parkin et al., 1991; Topp et al., 1994; Beulke et al., 2005). On the other hand, enhanced distribution of pesticide within the soil matrix has the potential to increase contact of pesticide with sites at which sorption can occur, thereby decreasing pesticide availability and increasing the formation of bound residues (Lechon et al., 1997; Beulke et al., 2005). Furthermore, the sieving process and subsequent rewetting of dried soil typically result in short term enhanced microbial activity, while drying can release inorganic ions, such as manganese which may be toxic to microorganisms (Angle et al., 1995).

Soil cores have the advantage of maintaining undisturbed the soil macrostructure (Parkin et al., 1991; Angle et al., 1995). It has been suggested that degradation of pesticides in soil cores is more representative of field conditions (Parkin et al., 1991;

Lechon et al., 1997). However, in undisturbed cores, the distribution of pesticides could be localised and therefore restricted, and this coupled with uneven distribution of microbes within cores, particularly in sub soil (Nunan et al., 2001) could reduce pesticide bioavailability (Parkin et al., 1991; Genod et al., 2003). Several studies have investigated the influence of laboratory incubation method on pesticide degradation rate in soil, with some indicating differences in degradation rate between intact cores and sieved soil (McDonald et al., 2006; Bending et al., 2007) but others finding no difference (Topp et al., 1994).

Bentazone is a commonly used herbicide which poses environmental concerns because of its high mobility, persistence and susceptibility to leaching from soil to groundwater (Thorstensen and Lode, 2001; Boesten and Van der Pas, 2000; Li et al., 2003). It is applied at a high rate in agriculture and frequently occurs as contaminant of groundwater and surface water (Helweg et al., 2002; Lagana et al., 2002 Dousset et al., 2004). In general, bentazone degradation in soil follows first order kinetics (Thorstensen and Lode, 2001), suggesting cometabolic degradation, without growth of organisms involved (Piutti et al., 2002). Soil organic matter content and pH are the soil properties that have been identified as having the greatest influence on the degradation of bentazone (Boivin et al., 2004). Contact time and soil type have a significant effect on bentazone availability (Boivin et al., 2004). It has been observed that the rate constant for bentazone dissipation was highest in the topsoil and decreased with depth (Leistra et al., 2001).

The objective of this study was to compare spatial variability in the biodegradation of bentazone both vertically and horizontally within an agricultural field. In particular, the spatial variability in degradation rate between locations was determined using both sieved soil and intact cores, to determine how representative

spatial variability measurements made using sieved soil were relative to samples in which the physical and microbiological characteristics of the soil were retained.

2. Material and methods

2.1. Pesticide and pesticide treatment history

Bentazone (3-isopropyl-1H-2,1,3-benzothiadiazin-4(3H)-one-2,2-dioxide) is a moderately mobile compound, which shows low sorption to soil (Koc<100) and high solubility in water (570 mg l^{-1}). Commercial formulation of bentazone, Basagran (87% w/w), was supplied by BASF plc (Ludwigshafen, Germany). Analytical grade bentazone was supplied by Chem Service Inc. (West Chester, USA). Sampling occurred in Long Close field on the farm at Warwick HRI, Wellesbourne, Warwickshire, UK. The soil is a sandy loam of the Wick series (Whitfield, 1974). There had been no application of bentazone over at least the previous 10 years.

2.2. Soil collection

2.2.1 Vertical variability in degradation rate using sieved soil

Soil was collected from five depths at three sampling locations in the field. Three pits separated by 60 m were excavated to 1 m depth using a mechanical digger, in February 2003. One side of each pit was further excavated using a surface sterilised trowel, so that the face was free of loose soil. Soil was collected from 0-10, 20-30, 40-50, 60-70 and 70-80 cm depth using two methods. From each depth soil was collected using a trowel and placed into a polythene bag. The trowel was surface sterilised with ethanol between the collection of each soil sample. Soil was spread onto clean polythene bags and left on the bench overnight to reduce moisture content, before being passed through surface sterilised sieves (<3 mm).

2.2.2 Vertical and horizontal spatial variability in degradation using intact cores and sieved soil

Soil was collected from two depths at ten different sampling locations in Long Close field, in April 2003. Ten pits were excavated in the field at intervals of 80 m (N-S) and 60 m (E-W). Samples of disturbed soil and undisturbed soil cores were collected from 0-15 cm depth and 50-60 cm depth. Sieved soil was collected and processed by methods described above. In order to maintain the physical and microbiological integrity of the soil, further samples were taken using 10 x 5 cm pre-sterilised stainless steel cores. Two cores were obtained at 0-15 and 50-60 cm depth from each of the 10 sampling locations by hammering the core horizontally into soil. Following removal, the top and bottom of each core was sealed with parafilm.

2.3. Analysis of soil characteristics

In the pre-sieved soil, total organic matter, microbial biomass-N, dehydrogenase activity, pH, clay, sand and silt content were measured, as described in Bending et al. (2006).

2.4. Pesticide application

For pre-sieved soil, commercial bentazone formulation was dissolved in distilled water and added to 300 g fw portions of soil from each location to provide 5 mg pesticide kg^{-1} soil, and further water was added to bring the water holding capacity to 40% (-33 kPa). Each soil was mixed thoroughly by hand, and then further mixed by passing through a <3 mm sieve five times. Each soil was transferred to a sterile polypropylene container which was loosely capped and incubated at $15^{o}C$ in the dark. Moisture content was maintained by the addition of sterile distilled water as necessary (usually once each week).

In the case of the intact cores, four 250 µl aliquots of the commercial formulation of bentazone in water were injected centrally at distances of 2 cm apart down the core to give a final concentration within the core equivalent to 5 mg kg^{-1} soil. The soil cores were sealed base and top with parafilm and incubated vertically at 15^{o}C in the dark.

2.5. Pesticide extraction and analysis

The pre-sieved soils were sampled at regular intervals over a 3-month period, with extraction and HPLC as described by Rodriguez-Cruz et al. (2006). For the second experiment, the cores were sampled after 127 days incubation. Soil was pushed from the cores, mixed by hand and sieved (<3 mm) five times. Sub-samples (10 g) of each soil were dried in an oven at 110^{o}C overnight to determine soil moisture content. Pesticide was extracted from each soil sample and analysed using the procedures described above.

Using pre-sieved soil, sorption of bentazone was determined using a batch mixing method, and adsorption distribution coefficients (Kd) measured as described by Rodríguez-Cruz et al. (2006).

2.6. Statistical analysis

All statistical analyses were performed using GenStat software (7th edition, VSN International Ltd.) The exponential and linear models were found to provide best fit to the degradation kinetics, and were used to obtain time to 50% degradation (DT50) values. Analysis of variance was used to determine the significance of differences in pesticide degradation between soil depth and method (sieved soils or soil cores). For the study of vertical and horizontal spatial variability in degradation using intact cores and sieved soil, variability within samples was estimated by replicating the

measurement of the cores for each treatment combination measured. Due to the fact that the design was unbalanced a general linear model was used for analysis. An accumulated analysis of variance was calculated with the replication of cores giving an estimate of the underlying (within sample) variability for both the core and sieved samples. Predictions were then made for the treatment combinations to give the estimated means and standard errors for each of the treatment combinations tested. % coefficient of variation in the study of vertical and horizontal spatial variability in degradation using intact cores and sieved soil was determined following angular transformation to normalise the variance.

3. Results and discussion

3.1 Vertical variability in degradation rate using sieved soil

In pre-sieved top-soil, bentazone degradation showed an exponential decrease in concentration over time (Fig 1). This is typical of co-metabolic activity, where pesticide degradation does not result in proliferation of degrader organisms. In sub-soil, there was a very slow degradation of bentazone with time and degradation was fitted to a linear model. Degradation rate declined progressively down the soil profile. In topsoil, DT_{50} was 56 and 65 days at 0-10 and 20-30 cm depth, respectively, while in sub-soil DT_{50} value increased from 178 days at 40-50 cm depth to 515 days at 70-80 cm depth (Table 1). Similarly, DT_{50} values for bentazone degradation in topsoils range from 2 to 15 weeks in previous studies (Huber and Otto, 1994), and degradation rate has been shown to decline with depth (Leistra et al. 2001). In the current study the proportional decline in degradation rate was greater than that of biomass or dehydrogenase, so that the ratio of biomass or dehydrogenase to DT50 increased significantly ($p<0.001$) with soil depth, in the case of biomass, ranging from 0.85 at 0-

10 cm depth, to 27.0 at 70-80 cm (data not shown). This suggests that the specific degraders responsible for bentazone transformation formed a decreasing proportion of the biomass, or reduced their relative activity, as depth increases. Bentazone showed limited sorption, and there was no significant change in sorption with soil depth (Table 1).

Bentazone DT_{50} was significantly correlated with soil biomass ($r = -0.701$, $p < 0.01$), dehydrogenase activity ($r = -0.595$, $p < 0.05$), pH ($r = 0.597$, $p < 0.05$), OM content ($r = -0.744$, $p < 0.01$) and Kd (-0.676, $p < 0.05$). Similarly, von Götz and Richter (1999) observed that soil biomass, organic carbon and pH value had the greatest influence on bentazone degradation behaviour in soil. Bentazone Kd was significantly correlated with OM content ($r = 0.561$, $p<0.05$) and pH ($r = -0.704$, $p<0.01$) confirming work by Li et al. (2003) and Thorstensen et al. (2001).

3.2. Vertical and horizontal spatial variability in degradation using intact cores and sieved soil

In topsoil, significantly less ($p<0.05$) bentazone remained in intact cores relative to sieved soil, with 6.6 and 17. 2 % remaining respectively (Table 2), suggesting more rapid degradation in intact cores relative to sieved soil. In the subsoil, the percentages of extractable bentazone from the soil cores were not significantly different between sieved soil and intact cores, with a mean of 64.7% and 60.1% remaining respectively after 127 days (Table 2).

Several studies in which pesticide degradation was associated with growth-linked catabolism have found slower degradation rates in intact cores relative to sieved soil (Parkin et al., 1991, Bending and Rodriguez-Cruz, 2007), while in common with our data, McDonald et al. (2006) investigating a cometabolised compound, found faster

degradation in cores relative to sieved soil. A number of factors could account for differences in degradation rate between intact cores and sieved soil. In particular, distribution of pesticide within the soil will differ, reflecting a less homogenous distribution in cores, with concentrations higher at points of injection (Parkin et al., 1991). Such differences could have different effects on communites involved in growth-linked and cometabolic degradation, particularly if distribution of pesticide is associated with the speed at which growth-linked catabolising communities adapt the potential to use the pesticide as an energy source (Bending and Rodriguez-Cruz, 2007). Furthermore the capacity of degraders to spread through soil is likely to be more rapid in sieved soil relative to intact cores (Angle et al., 1995).

Moisture was maintained at a constant level (40 % WHC, equivalent to 15 %) in the sieved soil, but in the cores mean moisture content was variable, and averaged 13.3 % (coefficient of variation 13.1) in topsoil and 16.7 (coefficient of variation 17.6) in sub soil (data not shown). Degradation rate was significantly correlated with moisture content in the top soil cores ($r = 0.718$, $p<0.001$), but not in subsoil cores or in the sieved soil, suggesting that moisture content was an important determinant of degradation rate, and since the cores are likely to closely mimic field conditions (Parkin et al., 1991; Angle et al., 1995), the spatial variability of degradation rate within the field.

When top- and sub-soil samples were combined, degradation of bentazone in cores and sieved soil after 127 days was significantly correlated ($r=0.87$, $p<0.001$). However, within the top-soil samples amounts of bentazone remaining in sieved soil and cores were not significantly correlated ($r= 0.23$). This was in contrast to the sub-soil samples, in which degradation in cores and sieved soil were significantly correlated ($r=0.65$, $P<0.05$). In sub-soil, % CV of sieved soil and cores were similar,

at 7.2 and 6.8 respectively. However in the top-soil, there was substantially higher variability within cores relative to sieved soil, with % CV of 29.5 and 15.1 respectively. These differences could be attributed to differences in variability between the cores and sieved soil, particularly reflecting moisture content differences, as discussed above. The data suggests that moisture content was variable within the field, and as shown by analysis of degradation rates in intact cores, was a key factor likely to control degradation *in situ*. Processing soil prior to conducting degradation assessment resulted in loss of moisture differences between locations, so that variability in degradation rates determined using sieved soil underestimated likely variability in degradation rates *in situ*

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References

Angle, J.S., Levin, M.A., Gagliardi, J.V., McIntosh, M.S., 1995. Validation of microcosms for examining the survival of Pseudomonas aureofaciens (lacZY) in soil. Appl. Environ. Microb. 61, 2835-2839.

Bending G.D., Rodriguez-Cruz M.S., 2007. Microbial aspects of the interaction between soil depth and biodegradation of the herbicide isoproturon. Chemosphere 66, 664-671.

Bending G.D., Lincoln, S.D., Edmondson, R.N., 2006. Spatial variation in the degradation rate of the pesticides isoproturon, azoxystrobin and diflufenican in soil and its relationship with chemical and microbial properties. Environ. Pollut. 139, 279-287.

Beulke, S., van Beinum, W., Brown, C.D., Mitchell, M., Walker, A., 2005. Evaluation of simplifying assumptions on pesticide degradation in soil. J. Environ. Qual. 34, 1933-1943.

Boesten, J.J.T.I., van der Pas, L.J.T., 2000. Movement of water, bromide and the pesticides ethoprophos and bentazone in a sandy soil: The Vredepeel data set. Agricultural Water Management 44, 21-42.

Boivin, A., Cherrier, R., Perrin-Ganier, C., Schiavon, M., 2004. Time effect on bentazone sorption and degradation in soil. Pest Manag. Sci. 60, 809-814.

Boivin, A., Cherrier, R., Schiavon, M., 2005. Bentazone adsorption and desorption on agricultural soils. Agronomy for sustainable development 25, 309-315.

Day, P.R., 1965. Particle fractionation and particle size analysis. In: Black, C.A., Evans, D.D., White, J.L., Ensminger, L.E., Clark, F.E. (Eds.), Methods of soil Analysis, Part 1, Agron. Monogr., vol. 9. ASA, Madison, WI, USA, pp. 545-566.

Dousset, S., Babut, M., Andreux, F., Schiavon, M., 2004. Alachlor and bentazone losses from subsurface drainage of two soils. J. Environ. Qual. 33, 294-301.

Fomsgaard, I.S., 1997. Modelling the mineralization kinetics for low concentrations of pesticides in surface and subsurface soil. Ecological Modelling 102, 175-208.

Fomsgaard, I.S., Felding, G., Schjonning, P., 1998. Sampling and substrate application methods for pesticide mineralization experiments in undisturbed soil samples. Int. J. Environ. An. Ch. 70, 121-132.

Genod, L.V., Chenu, C., Soulas, G., 2003. Spatial variability of 2,4-dichlorophenoxyacetic acid (2,4-D) mineralization potential at a mllimetre scale in soil. Soil Biol. Biochem. 35, 373-382.

Helweg, A., Bay, H., Hansen, H.P.B., Rabolle, M., Sonnenborg, A., Stenvang, L., 2002. Pollution at and below sites used for mixing and loading of pesticides. Int. J. Environ. Anal. Chem. 82, 583-590.

Huber, R., Otto, S., 1994. Environmental behaviour of bentazone herbicide. Rev. Environ. Contamin. Toxicol. 137, 111-134.

Lagana, A., Bacaloni, A., De Leva, I., Faberi, A., Fago, G., Marino, A., 2002. Occurrence and determination of herbicides and their major transformation products in environmental waters. Anal. Chim. Acta 462, 187-198.

Lechon, Y., Sanchez-Brunete, C., Tadeo, J.L., 1997. Influence of the laboratory incubation method on chlorotoluron and terbutryn degradation in soil. J. Agric. Food Chem. 45, 951-954.

Leistra, M., Smelt, J.H., Matser, A.M., Boyte, J.J., van der Pas, L.J.T., 2001. Rate of bentazone transformation in four layers of a humic sandy soil profile with fluctuating water table. Pest Manag. Sci. 57, 1023-1032.

Li, K., Liu, W., Xu, D., Lee, S., 2003. Influence of organic matter and pH on bentazone sorption in soils. J. Agric. Food Chem. 51, 5362-5366.

McDonald, J.A., Gaston, L.A., Jackson, S.H., Locke, M.A., Zablotowicz, R.M. 2006. Degradation kinetics assessment for the fungicide BAS 505 in intact soil cores versus batch soils. Soil Sci. 171, 239-248.

Parkin, T.B., Shelton, D.R., Robinson, J.A., 1991. Evaluation of methods for characterizing carbofuran hydrolisis in soil. J. Environ. Qual. 20, 763-769.

Piutti, S., Marchand, A.L., Lagacherie, B., Martin-Laurent, F., Soulas, G., 2002. Effect of cropping cycles and repeated herbicide applications on the degradation of dichlofopmethyl, bentazone, diuron, isoproturon and pendimethalin in soil. Pest Manag. Sci. 58, 303-312.

Rodriguez-Cruz M.S., Jones, J.E., Bending, G.D., 2006. Field-scale study of the variability in pesticide biodegradation with soil depth and its relationship with soil characteristics. Soil Biol. Biochem. 38, 2910-2918.

Thorstensen, W., Lode, O., 2001. Laboratory degradation studies of bentazone, dichlorprop, MCPA, and propiconazole in Norwegian soils. J. Environ. Qual. 30, 947-953.

Thorstensen, C. W., Lode, O., Eklo, O.M., Christiansen, A. 2001. Sorption of bentazone, dichlorprop, MCPA and propiconazole in reference soils from Norway. J. Environ. Qual. 30, 2046-2052.

Topp, E., Smith, W.N., Reynolds, W.D., Khan, S.U., 1994. Atrazine and metolachlor dissipation in soils incubated in undisturbed cores, repacked cores, and flasks. J. Environ. Qual. 23, 693-700.

Tuxen, N., Tuchsen, P.L., Rugge, K., Albrechtsen, H-J., Bjerg, P.L. 2000. Fate of seven pesticides in an aerobic aquifer studied in column experiments. Chemosphere 41, 1485-1494.

van der Pas, L.J.T., Leistra, M., Boesten, J.J.T.I., 1998. Rate of transformation of atrazine and bentazone in water-saturated sandy subsoils. Pest. Sci. 53, 223-232.

Vischetti, C., Scarponi, L., Perniola, M., Tarantino, E., 1998. Field and lysimeter study on the leaching of bromide ion and the herbicides imazethapyr and bentazone in a clay loam soil in southern Italy. Fresenius Environ. Bull. 7, 641-648.

von Götz, N., Richter, O. 1999. Simulation of herbicide degradation in different soils by use of pedo-transfer functions (PTF) and nonlinear kinetics. Chemosphere 38, 1401-1407.

Whitfield, W.A.D., 1974. The soils of the National Vegetable Research Station, Wellesbourne. Report of the National Vegetable Research Station for 1973, pp. 21-30.

Table 1

Change in key soil properties and bentzone degradation and sorption with soil depth. DT50 and Kd measurements represent data obtained using sieved soil. % bentazone remaining in sieved soil and cores reflects 20 day time point following application. Data represents mean of 3 sampling locations at each depth.

Soil depth (cm)	**OM (%)[a]**	**pH [a]**	**Biomass (mg C kg^{-1} soil) [a]**	**Dehydrogenase (µg TPF[b] g^{-1} soil)[a, b]**	**DT_{50} (days)**	**Kd (ml g^{-1})**
0 - 10	2.62	7.06	65.5	43.1	56	0.15
20 - 30	2.33	7.13	64.5	22.9	65	0.11
40 - 50	1.98	7.79	36.4	10.7	178	0.10
60 - 70	1.66	7.98	21.5	7.48	306	0.09
70 - 80	1.35	8.09	19.1	6.60	515	0.06
LSD (P>0.05)	0.29	0.45	16.0	14.7	178	0.07
Significance of effect of depth[c]	***	**	***	***	***	NS

[a] From Bending and Rodriguez Cruz (2007)

[b] Triphenyl formazan

[c] NS, not significant; ** significant, P<0.01; *** significant, P<0.001

Table 2 Key soil characteristics and % bentazone remaining after 127 days incubation in sieved soil and intact cores. Data represents mean of 10 sampling locations at each soil depth over a 160 x 90 m sampling area.

Soil depth (cm)	**OM (%)[a]**	**pH [a]**	**Biomass (mg C kg^{-1} soil)[a]**	**Dehydrogenase (µg TPF[b] g^{-1} soil)**	**% Bentazone remaining in sieved soil [b]**	**% Bentazone remaining in cores [b]**
0 - 10	2.71	6.71	44.5	43.2	17.2 (23.7)	6.63 (12.5)
50 - 60	1.43	7.30	23.4	3.87	60.1 (51.4)	64.7 (54.21)
LSD P>0.05	0.47	0.53	16.3	11.9	3.0	
Significance of effect of depth[c]	***	***	***	***	***	***

[a] Triphenyl formazan

[b]data in brackets gives angular transformed data, to which LSD relates, for comparison between sieved soil and cores, and for the effect of depth.

[c] *** significant, P<0.001

Figure 1 Degradation of bentazone in sieved soil from different depths

Data represent average of three replicate sampling locations at each depth

Bars represent +/- standard error of the mean