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Effect of fonofos on soil dehydrogenase activity

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A b s t r a c t. The purpose of the present study was to provide information about the effect exerted by fonofos pesticide on dehydrogenase activity of Mollic Gleysol, Eutric Fluvisol and Eutric Histosol soil samples. The effect of fonofos on dehydrogenase activity was evaluated taking into consideration the pesticide dose and soil incubation time. It was found that soil dehydrogenase activity showed an inhibitory effect with increase of the pesticide doses. Dose on the level of 1μg g^{-1} of pesticide caused 5-21% inhibition of enzymatic activity, whilst ten times higher doses resulted in 17-44% drop of activity in comparison to the control. Increase of dehydrogenase activity was also connected with fall of redox potential (Eh). The results suggest a negative effect of fonofos on soil dehydrogenase activity in the first stage after application.

K e y w o r d s: pesticide, soil, dehydrogenase activity

INTRODUCTION

Pesticides are widely used to improve the yield and quality of agricultural produce and for controlling pests and diseases in crop production (Crum *et al*., 1999; McDonald *et al*., 1999). Pesticide application has increased dramatically over time and the potential negative effects on human health and the environment are now of concern (McDonald *et al.*, 1999; Perucci *et al*., 2000). Pesticides, where used correctly, can save up to 40% in crop losses, when pesticides are mis- or over-used the environmental and public health consequences can be very considerable (Richardson, 1998). The ideal outcome of pesticide use occurs when a pesticide accomplishes the purposes for which it was applied and then rapidly breaks down into harmless components, such as carbon dioxide and water. Fate process can be separated into three major types: adsorption, which binds pesticides, transfer processes, which move pesticides, and degradation processes, which break pesticides down. Pesticide degradation in soils is the result of a combination of chemical and biological events (Wu and Nofziger, 1999). The transformation of pesticides by soil microorganisms consists in essentially intracellular, enzyme-catalyzed reactions encountered in their regular metabolic activity (Wu and Nofziger, 1999). Soil conditions, such as moisture, temperature, aeration, pH, the amount of organic matter, affect the rate of microbial degradation because of their direct influence on microbial growth and activity. Wet soils tend to adsorb less pesticide than dry soils because water molecules compete with pesticide for the binding sites on soil particles. Pesticides applied to the soil at planting should persist during the development of plant roots. Therefore, a portion of the pesticide likely interacts with microorganisms in the soil and in the rizosphere (Perucci *et al*., 2000).

Soil enzyme activity is believed to be sensitive to pollution and has been proposed as an index of soil degradation (Gianfreda *et al*., 2005; Trasar-Cepeda *et al*., 2000). Dehydrogenase is thought to be an indicator of overall microbial activity, because it occurs intracellular in all living microbial cells and is linked with microbial oxydoreduction processes (Quilchano and Maranon, 2002; Stępniewska and Wolińska, 2005). It is a specific kind of enzyme which plays a significant role in the biological oxidation of soil organic matter by transferring protons and electrons from substrates to acceptors. Soil dehydrogenase activity is considered to be a valuable parameter for assessing the side effects of herbicide treatments on the soil microbial biomass.

The purpose of the present study was to provide information about the effect of the fonofos pesticide on dehydrogenase activity of selected soils samples. Fonofos is an insect exterminant applied for corn, fruit trees and decorative plants protection, usually at doses of 1-2.5 kg ha^{-1}.

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It is sold in the form of preparations named dyphonate, dyfonate 106, or difonaftol. Fonofos dose and its reflect time were taken into account when estimating dehydrogenase activity in three types of soils: Mollic Gleysol, Eutric Fluvisol and Eutric Histosol.

MATERIALS AND METHODS

The characteristics of the soil material are reported in Table 1. The soil samples from Ap level were taken as follows: Eutric Histosol from Hajdów, near Bystrzyca river; Eutric Fluvisol from Vistula Valley (Puławy); Mollic Gleysol from the Valley of Tyśmienica river (Kock), located in the south-east part of Poland. In the model experiment soil samples were incubated (at 20°C) for 42 days, amended with doses pesticide, equal to 1µg of fonofos 1 g^{-1} of soil and ten times higher (10 µg of fonofos 1 g^{-1} of soil). Moreover, soil samples without fonofos supplement were prepared as a control. Determination of soil dehydrogenase activity in soils is based on the use of soluble tetrazolium salts [2,3,5-triphenylotetrazolium chloride (TTC)], as artificial electron acceptors, which are reduced to red-coloured formazans, extracted and then determined calorimetrically (Casida *et al.*, 1964). Soil, in portions of 6 g, was placed in 50 ml glass flasks, where 1 ml of 3% aqueous solution TTC, 120 mg $CaCO_3$ and 4 ml distilled water were added. Thus prepared soil samples were incubated for 20 h in thermostatic chamber at 30°C. After incubation the soils were extracted with ethanol, and filtered. Absorbance was measured by means of HITACHI UV-VIS U-2001 spectrophotometer at λ=485 nm. All determinations of enzymatic activity were performed in triplicate (1, 3 and after one week, up to 42 days of incubation), and all values reported are averages. Redox potential (Eh) was measured by using a combined electrode placed in the soil with Eh meter Radiometer pIONeer 65".

RESULTS AND DISCUSSION

Realized laboratory experiments showed that there exists a relationship between soil enzymatic activity and fonofos concentration in the soil environment. Changes in the soil dehydrogenase activity in the presence of pesticide doses during incubation time are presented in Fig. 1. The most sensitive to pesticide contamination seemed to be the Eutric Histosol. In that case a 40% decrease of dehydrogenase activity as a result of 10 µg of fonofos supplement was estimated. The results suggest that dehydrogenase activity depends on the type of soil. In the literature, it is usually reported that high enzymatic activities are associated with rich organic matter contents (Davies and Graves, 1981; Lethbridge *et al*., 1981). However, much of the reported data contrast with this assertion, as it is apparent in the Eutric Histosol soil which presents a very high organic matter content, but low dehydrogenase activity (Ladd, 1985). On the first day of the laboratory experiment rather low values of enzymatic activity were noted, whilst the final days of incubation resulted in an extension of dehydrogenase activity, presumably because the process of fonofos decomposition was completely finished. The maximum values of dehydrogenase activity, in the range of 80-120 [µg TPF g^{-1} min^{-1}]10^{-6} were recorded in the Mollic Gleysol soil samples, whereas the lowest - at the level 12-22 [µgTPF $g^{-1}min^{-1}$]10^{-6} - in the Eutric Histosol soil, on the 42nd day of the incubation period. In the Eutric Fluvisol soil samples, the pesticide influence on inhibition of dehydrogenase activity became observable on the 14th day of incubation, when the one by one dose caused a 25% fall, whereas the ten time higher factor resulted in a 52% reduction of enzymatic activity. Till that time, dehydrogenase values were comparable with the control sample. The Eutric Histosol soil is the most sensitive to pesticide contamination. A negative effect of fonofos amendment was observed after 3 days of incubation, when 1 µg g^{-1} of fonofos brought about a 36% drop of dehydrogenase activity, while the 10 µg g^{-1} dose resulted in a 54.6% inhibition of soil enzymatic activity. Stronger reduction, up to 62.4%, as a result of the 10 µg g^{-1} dose, was noted after one and two weeks of incubation, but the highest spread between the control sample and the sample with pesticide addition was recorded after 28 days and caused a 68.2% fall of dehydrogenase activity. In the Mollic Gleysol samples the influence of the pesticide on soil enzymatic activity started to be observed after one week of incubation, but from the 14th day to the end of the experiment this effect was significant and noticeable. Generally, 1µg g^{-1} dose of fonofos was responsible for about 26% inhibition of soil dehydrogenase activity, whereas ten times higher factor reduced the activity by 46.6% on the 21st day of incubation time; later the fall of enzymatic activity ranged from 22.5% to 30 % in relation to the control samples.

T a b l e 1. Main features of the soils investigated

Type of soil	Granulometric composition (%, dia in mm)			$C_{org.}$ (%)	pH_{H_2O}
	1-0.1	0.1-0.02	<0.025		
Eutric Histosol	28	36	36	36.5	7.5
Eutric Fluvisol	15	46	39	2.47	6.3
Mollic Gleysol	38	25	37	2.12	7.5

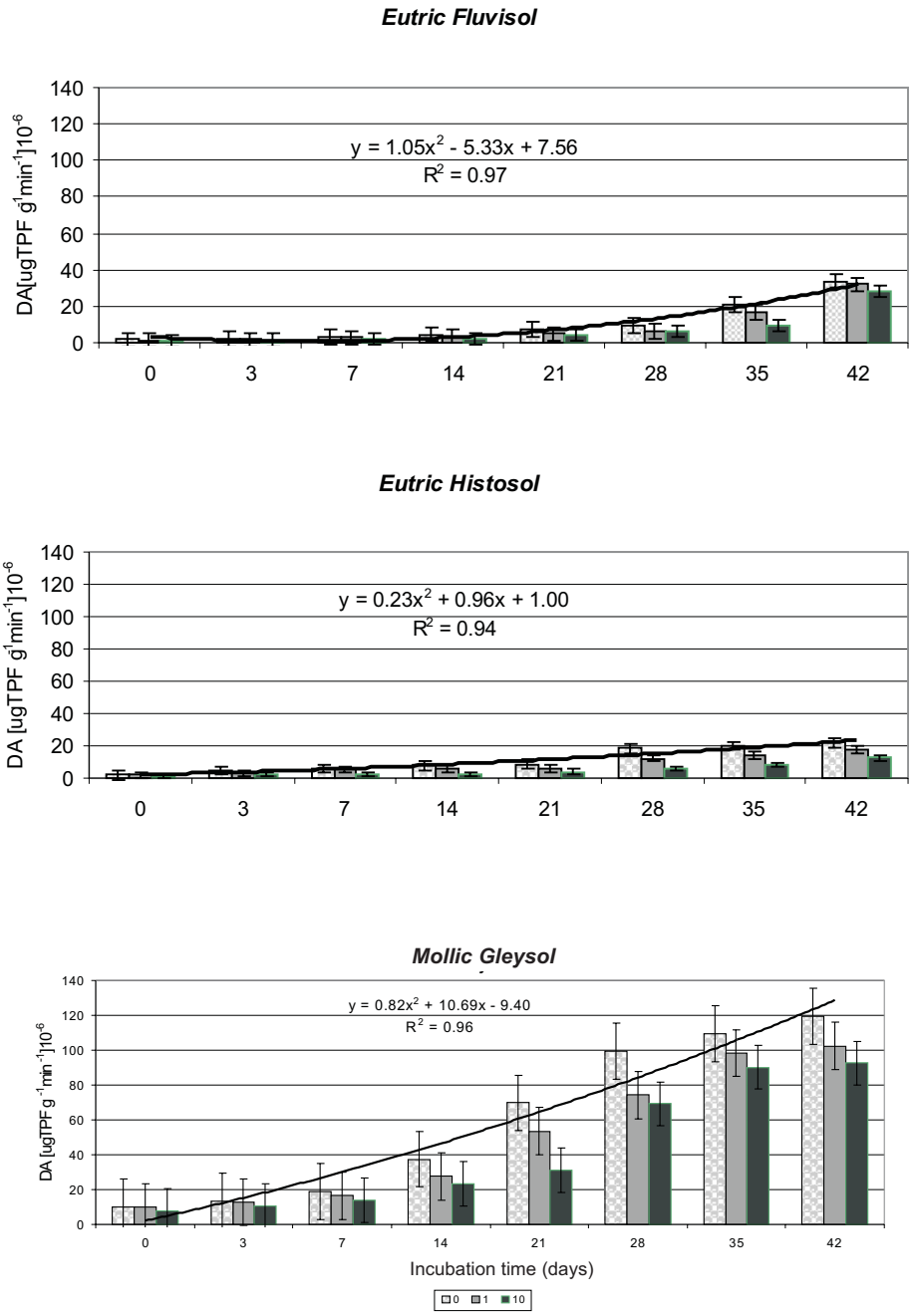

Fig. 1. Dynamic of dehydrogenase activity during incubation at 20ºC (0 – control; 1 – 1 μg g^{-1} fonofos supplement; 10 – 10 μg g^{-1} fonofos supplement).

Values obtained for redox potential (Eh) during 42 days of experiment are presented in Table 2. As a result of adding the pesticide to the soil samples a drop of Eh values was observed. This phenomenon is connected with reduction processes in the soil environment, as well as with decomposition of fonofos. However, it is also worth taking into account that the activity of dehydrogenase increases in anaerobic conditions. At the beginning of the experiment high values of redox potential were noted. The highest Eh values, on the level of 515 mV, were found for Eutric Fluvisol in the control sample, while for Eutric Histosol and Mollic Gleysol these values equalled 432 and 351 mV, respectively. However, the strongest influence of fonofos on Eh values was observed in the Mollic Gleysol samples. Redox potential in the control samples reached a higher level than in the samples with pesticide supplement. For each soil sample ΔEh between the first and the last day of experiment was calculated. The results are displayed in

T a b l e 2. Eh values (mV) of the soils investigated during incubation at 20°C

Soil	Fonofos dose (μg g^{-1})	Incubation time (days)								ΔEh (mV) (mean)
		1	3	7	14	21	28	35	42	
		Eh (mV)								
Eutric Fluvisol	0	515	485	364	261	204	264	242	193	322
	1	494	374	338	335	203	248	253	195	299
	10	482	341	345	286	215	210	220	184	298
Eutric Histosol	0	432	382	365	360	262	173	248	241	191
	1	402	401	391	358	271	253	303	289	113
	10	395	388	389	399	340	332	360	265	130
Mollic Gleysol	0	351	357	308	303	185	199	158	94	257
	1	357	351	359	285	162	143	121	68	289
	10	348	346	332	279	184	198	139	49	299

Table 2. Finally, with fall of redox potential an increase of soil dehydrogenase activity was observed. However, besides anaerobic conditions which were favourable for enzymatic activity, pesticide doses of 1 and 10 μg g^{-1} caused an inhibitory effect on soil dehydrogenases. On the last (42nd) day of incubation, the 1 μg g^{-1} dose of fonofos was responsible for a reduction of dehydrogenase activity by 5.6% in Eutric Fluvisol, 14.1% in Mollic Gleysol, and 20.7% in Eutric Histosol, in relation to the controls values. Whereas, the ten times higher dose (10 μg g^{-1} of fonofos) resulted in a drop of enzyme activity by 16.8, 22.5 and 43.5% in Eutric Fluvisol, Mollic Gleysol and Eutric Histosol, respectively. As a consequence of this, it is worth taking into consideration that not only the presence of fonofos affected soil enzymatic activity in that case, but also the influence of aeration conditions in the soil environment was significant and considerable. Vink and Sjoerd (1997) found that prevailing redox conditions have a strong impact on pesticide transformation rates. Some phenoxy-acetic compounds, which are considered improbable leachers based on their short aerobic half lives, appear to be persistent in low-oxygen conditions. The opposite effect was observed for aldicarb, in which chemical catalysis increased transformation rates when redox potential decreased.

In the literature, contrasting and opposite effects on several soil enzymes are reported (Gianfreda *et al.*, 1994, 2005; Quilchano and Maranon, 2002). Glyphosate was found to inhibit dehydrogenase, phosphatase and urease activities in a sandy loam soil (Dzantor and Felsot, 1991), or to activate, to varied degrees, urease in different types of soils (Gianfreda *et al*., 2005). No effects on soil dehydrogenase, phosphatase, urease, arylesterase and arylacylamidase were detected by Lethbridge *et al*. (1981), and Nakamura *et al*. (1990). Reduced enzymatic activities were also found by Perucci and Scarponi (1990) and by Dzantor and Felsot (1991) in studies on the interference of atrazine with the phosphatase, dehydrogenase and esterase activity of soil. Soil dehydrogenase activity is also sensitive to heavy metals contamination. Stępniewska and Wolińska (2005) found that the application of trivalent and hexavalent chromium compounds had a noticeable negative effect on soil dehydrogenase activity. The lowest values of dehydrogenase activities were the effect of increasing Cr(III) and Cr(VI) doses. Indeed, it is particularly difficult to explain a change of soil enzymatic activity in response to a certain factor or to establish cause-effect relationships between the applied disturbing factor and the soil enzyme activity variation (Gianfreda *et al*., 2005). More detailed knowledge of the nature and mechanisms involved in the pesticideenzyme interactions requires further studies and is a challenge for future research activities.

CONCLUSIONS

1. Fonofos introduction to Eutric Fluvisol, Eutric Histosol and Mollic Gleysol resulted in an inhibition of dehydrogenase activity.

2. Dose of the pesticide at the level of 1 μg g^{-1} was responsible for 5-36% reduction of dehydrogenase activity, whilst 10 μg g^{-1} fonofos supplement was the reason of 17-65% drop of enzymatic activity.

3. Changes in dehydrogenase activity in the presence of pesticide contamination depend on the type of soil. Eutric Histosol seemed to be the most sensitive to fonofos contamination.

4. The maximum values of dehydrogenase activity ranged from 80 to 120 [μg TPF g1 min^{-1}]10^{-6} in the Mollic

Gleysol samples, whereas the lowest values, at the level of 12-22 [μg TPF $g^{-1}min^{-1}]10^{-6}$, were observed in the Eutric Histosol at the end of the 42-day incubation period.

5. Increase of soil dehydrogenase activity is strongly connected with fall of Eh values.

REFERENCES

Casida L., Johnson J., and Klein D., 1964. Soil dehydrogenase activity. Soil Sci., 98, 371-376.

Crum S.H.J., Polman A.M.M., and Leistra M., 1999. Sorption of nine pesticides to three aquatic macrophytes. Arch. Environ. Contam. Toxicol., 37, 310-316.

Davies H.A. and Graves M.P., 1981. Effects of some pesticides on soil enzymatic activities. Weed Res., 21, 205-209.

Digrak M. and Ozcelik S., 1998. Effect of some pesticides on soil microorganisms. Bull. Environ. Contam. Toxicol., 60, 916-922.

Dzantor E.K. and Felsot A., 1991. Microbial responses to large concentrations of herbicides in soil. Environ. Toxicol. Chem., 10, 649-655.

Gianfreda L., Rao M.A., Piotrowska A., Palumbo G., and Colombo C., 2005. Soil enzyme activities as affected by anthropogenic alterations: intensive agricultural practices and organic pollution. Sci. Total Environ., 341, 265-279.

Gianfreda L., Sannino F., Ortega N., and Nannipieri P., 1994. Activity of free and immobilized urease in soil: effects of pesticides. Soil Biol. Biochem., 26, 777-784.

Ladd J.N., 1985. Soil enzymes. In: Soil Organic Matter and Biological Activity. Nijhoff, Dordecht.

Lethbridge G., Bull A.T., and Burns R.G., 1981. Effects of pesticides on 1,3-ß-glucanase and urease activities in soil in presence and absence of fertilizers, lime and organic materials. Pesticide Sci., 12, 147-155.

McDonald L., Jebellie S., Madramootoo C., and Dodds T., 1999. Pesticide mobility on hillside soil in St. Lucia. Agric. Ecosyst. Environ., 72, 181-188.

Nakamura T., Mochida K., Ozon Y., Ukawa S., Sakai M., and Mitsugi S., 1990. Enzymological properties of three soil hydrolases and effects of several pesticides on their activities. J. Pesticide Sci., 15, 593-598.

Perucci P., Dumontet S., Bufo S.A., Mazzatura A., and Casucci C., 2000. Effects of organic amendment and herbicide treatment on soil microbial biomass. Biol. Fert. Soils, 32, 17-23.

Perucci P. and Scarponi L., 1990. Atrazine, alachlor and metalachor: persistence and phopshatase activity in a clay-loam soil. Agrochimica, 34, 214-222.

Quilchano C. and Maranon T., 2002. Dehydrogenase activity in Mediterranean forest soils. Biol. Fert. Soils, 35, 102-107.

Richardson M., 1998. Pesticides-friend or foe? Water Sci. Techn., 37, 19-25.

Stępniewska Z. and Wolińska A., 2005. Soil dehydrogenase activity in the presence of chromium (III) and (VI). Int. Agrophysics, 19, 79-83.

Trasar-Cepeda C., Leiros M.C., Seoane S., and Gil-Sotres F., 2000. Limitations of soil enzymes as indicators of soil pollution. Soil Biol. Biochem., 32, 1867-1875.

Vink J.P.M. and Sjoerd E.A.T.M., 1997. Effect of oxygen status on pesticide transformation and sorption in undisturbed soil and lake sediment. Environ. Toxicol. Chem., 4, 608-616.

Wu J. and Nofziger D.L., 1999. Incorporating temperature effects on pesticide degradation into a management model. J. Environ. Qual., 28, 92-100.