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## EFFECT OF SOIL POLLUTION WITH OIL DERIVATIVES ON THE OCCURRENCE OF *Harpalus rufipes* DEG.

### ODDZIAŁYWANIE ZANIECZYSZCZENIA GLEBY ROPOPOCHODNYMI NA WYSTĘPOWANIE *Harpalus rufipes* DEG.

**Abstract:** The research aimed at investigating the effect of oil derivatives during the process of their bioremediation on dynamics of *Harpalus rufipes* Deg. (Coleoptera, Carabidae) occurrence. The following objects were established: control - unpolluted soil; soil polluted with petrol; soil polluted with diesel fuel and soil polluted with used engine oil (dose: 6,000 mg of fuel · kg<sup>-1</sup>d.m. of soil). Experiment was set up in two series: with natural and supported bioremediation. *H. rufipes* was trapped using Barber's traps, during the periods from June to October 2010, from May to October 2011 and 2012. Activity of *Harpalus rufipes* Deg. species representatives under conditions of soil polluted with oil derivatives depended on the kind of pollutant substance and on the time which passed from the moment of the soil pollution. Petrol had the least negative effect - it was visible only during the first four months after the pollution. Negative effect of diesel and engine oil was observed even 14 months from the moment of pollution. Application of supported bioremediation on the soil polluted with diesel oil contributed to increasing the number of trapped *H. rufipes* beetles after 14 months from the moment of its application and from the moment of the soil contamination, whereas after two years the same measure neutralized the effect of intensified activity of the above mentioned beetles under conditions of the soil polluted with diesel and engine oil.

**Keywords:** oil derivatives, soil, bioremediation, Carabidae, *Harpalus rufipes* Deg.

*Harpalus rufipes* Deg. beetle from the Carabidae family is often counted among the species dominating the epigeal fauna of arable fields [1-4]. It has been stated that it is both herbivorous species (particularly in spring) and may feed on animal food [5]. Both feeding systems may have positive aspects, because consumed plants and their seeds often include weeds, whereas aphids are among the eaten animals [6]. Great number of beetles from this species and their distribution on crops on a majority of soils may prove very important from the perspective of pest and weed control. Quality of the soil environment is of crucial importance for the epigeal fauna presence. Among many kinds of soil pollution, contamination with oil derivatives has been the least identified as to its effect on invertebrates connected with the soil environment. At the same time the occurrence of many Carabidae species is known to be connected both with the type of soil and soil management [7, 8].

The research aimed at investigating the effect of oil derivatives during the process of their bioremediation on dynamics of *Harpalus rufipes* Deg. (Coleoptera, Carabidae) occurrence.

### Materials and methods

The research was carried out in 2009-2012 at the Experimental Station of the University of Agriculture in Mydlniki near Krakow. In the experiment containers with

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a capacity of 1 m<sup>3</sup> were used. The containers were dug into the soil, so that their upper area was on the same level with the surrounding soil. In June 2010 the soil in containers was polluted with the following oil derivatives: petrol, diesel oil and used engine oil (dose 6,000 mg of fuel · kg<sup>-1</sup>d.m. of soil). A week later half of the containers was subjected to bioremediation with the use of ZB-01 biopreparation, specially prepared for this purpose. The experiment was performed in four replications. Four objects were created in two series (natural and supported bioremediation): 1. Control - unpolluted soil, 2. Soil contaminated with petrol, 3. Soil contaminated with diesel oil and 4. Soil contaminated with used engine oil. Epigeal fauna including *Harpalus rufipes* Deg. was trapped using Barber's traps (0.9 dm<sup>3</sup> jars dug even with the soil level and protected against atmospheric precipitation with a plastic roof) placed in the central point of each container. The traps were emptied once a week during the periods from June to October 2010, from May to October 2011 and 2012. Trapped carabid beetles were classified using appropriate keys [9]. Statistical computations were made using Statistica 10.0 PL computer programme. Means were diversified using NIR Fisher test at significance level  $\alpha = 0.05$ .

## Results and discussion

The dynamics of *H. rufipes* trappings points to its most numerous presence in the summer months (July, August, September) and generally at the same time a significant diversification in the trapped number was observed depending on the applied pollutant substance (Figs. 1 and 2, Table 1).

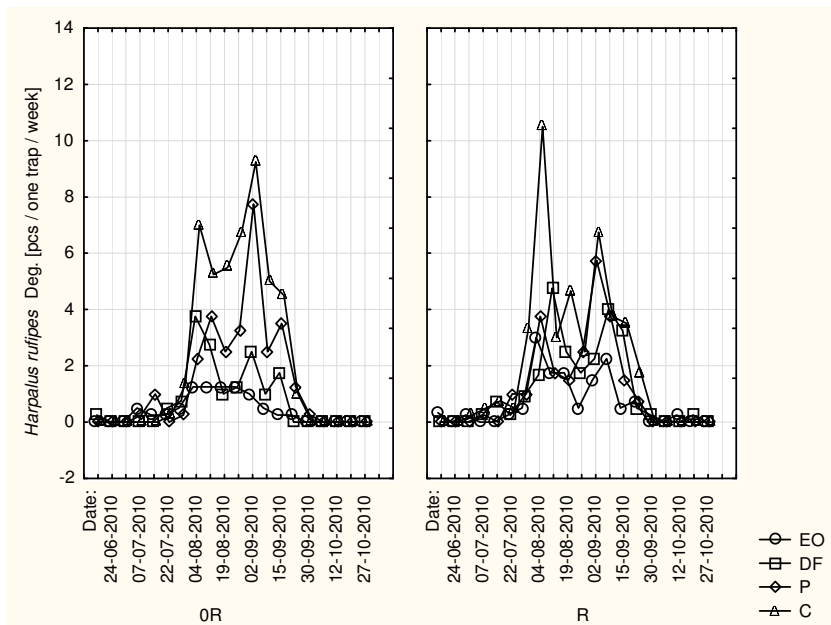


Fig. 1. Course of dynamics of *Harpalus rufipes* Deg. occurrence trapped using Barber's traps in 2010. EO - soil contaminated with used engine oil, DF - soil contaminated with diesel fuel, P - soil contaminated with petrol, C - unpolluted soil, OR - series without bioremediation, R - series with bioremediation

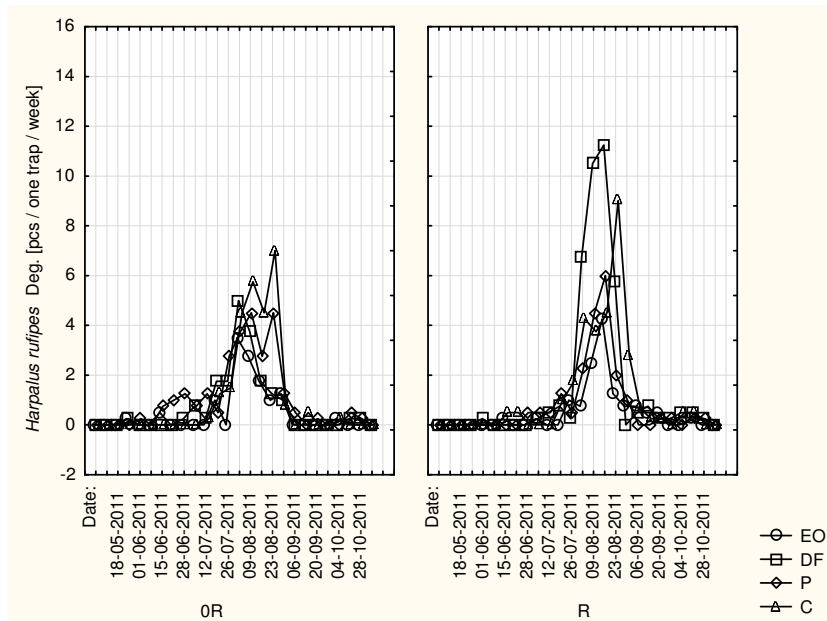


Fig. 2. Course of dynamics of *Harpalus rufipes* Deg. occurrence trapped using Barber's traps in 2011. The symbols as in Figure 1

Table 1  
Occurrence of *Harpalus rufipes* Deg. trapped using Barber's traps in individual months after soil contamination. The symbols as in Figure 1

Number of months from the moment of soil contamination	<i>Harpalus rufipes</i> Deg. [pcs/trap/month]							
	Control		Petrol		Diesel fuel		Engine oil	
	OR	R	OR	R	OR	R	OR	R
1	0.00 a*	0.75 a	0.25 a	0.25 a	0.26 a	0.25 a	0.50 a	0.58 a
2	7.17 b	15.08 c	3.50 ab	5.75 ab	5.00 ab	3.42 ab	2.25 a	3.75 ab
3	26.75 b	16.92 ab	17.25 ab	11.50 a	7.50 a	11.25 a	4.75 a	5.50 a
4	10.50 d	9.00 cd	7.50 bcd	6.00 a-d	2.75 ab	8.00 bcd	1.00 a	3.50 abc
5	0.00 a	0.00 a	0.00 a	0.00 a	0.00 a	0.25 a	0.00 a	0.25 a
11	0.00 a	0.00 a	0.25 a	0.00 a	0.25 a	0.25 a	0.25 a	0.00 a
12	0.00 a	1.00 a	3.00 b	0.50 a	0.25 a	0.00 a	0.50 a	0.25 a
13	3.00 a	2.75 a	5.25 a	2.75 a	4.50 a	1.75 a	1.00 a	1.25 a
14	22.50 ab	24.25 ab	16.75 ab	15.75 a	12.75 a	34.25 b	10.25 a	9.50 a
15	0.50 ab	1.17 ab	0.75 ab	0.50 ab	0.00 a	1.75 b	0.00 a	1.75 b
16	0.25 a	1.00 a	0.75 a	0.50 a	0.50 a	1.25 a	0.25 a	0.25 a
23	1.50 b	0.50 ab	0.75 ab	0.25 a	0.50 ab	0.00 a	0.75 ab	0.50 ab
24	2.50 a	1.25 a	2.50 a	1.25 a	1.50 a	2.25 a	1.25 a	2.00 a
25	5.00 ab	2.83 a	7.25 ab	3.50 a	18.83 d	10.50 bc	15.08 cd	5.83 ab
26	1.00 a	1.00 a	0.83 a	3.50 bc	1.83 ab	5.00 c	1.50 ab	3.42 bc
27	1.25 c	0.92 bc	0.00 a	0.25 ab	0.33 ab	0.08 ab	0.50 abc	0.00 a
28	0.00 a	0.00 a	0.00 a	0.00 a	0.00 a	0.00 a	0.00 a	0.00 a

\*Means in lines marked with the same letters do not differ significantly according to NIR test at  $\alpha = 0.05$ ; factors contamination x remediation

A similar seasonal course of the dynamics was noted by Huruk [1]. The author states that characteristic for *H. rufipes* is the model of dynamics in which the species activity intensifies with passing vegetation season and reaches the peak in August or September, then decreases. Only in few cases the peak activity were observed in other months. It results from the fact that it is a species of autumnal type of development, wintering as a larvae or pupa. Further development ensues in the subsequent year, so that the peak appearance of particularly the beetles which winter as larvae, fall in the summer months. During the first 5 months from the pollution (in the 2010 season) considerably limited number of trapped beetles from this species was registered under the influence of all applied substances (Fig. 1). Petrol reduced *H. rufipes* presence the least, whereas engine oil the most. In laboratory experiments on the effect of soil derivative polluted soil collected from the presented experiment 2 months after pouring the oil derivatives, no negative effect on the beetle viability or changes of their body weight was noted, except for the engine oil which decreased beetle survivability by about 30% after 4 weeks of culturing. Also inhibited activity of the enzymes connected with functioning of the defence system in these insects was noted, specific for the pollutant substance [10]. Statistical analysis of the results obtained in respective months passing from the moment of pollution revealed significantly less trapped specimens under conditions of soil polluted with diesel oil in months 3 and 4, while in polluted with engine oil in months from 2 to 4, in comparison with the unpolluted soil (Table 1). Application of biopreparation contributed to an increase in the number of trapped specimens in the control soil in the 2<sup>nd</sup> month of the experiment. In the objects with polluted soil greater number of *H. rufipes* were caught in the series with bioremediation, however the differences were not statistically proven. No statistically significant differences were registered either in the 1<sup>st</sup> or in the 5<sup>th</sup> month of the experiment, but it is due mainly to low activity of beetles in this period. There were June and October months when according to many authors, dynamics of *H. rufipes* trappings is characterized by a sharply outlined peak number preceded by a very low number before the peak and equally low after the maximum has been reached [1, 11, 12]. Analysis of results for the whole period from the 1<sup>st</sup> to the 5<sup>th</sup> month after contamination revealed a marked decrease in the number of trapped *H. rufipes* for all analysed kinds of pollution, the highest for the engine oil, the lowest for petrol. On the other hand, no significant effect of the applied bioremediation was noticed (Fig. 4).

During the subsequent research period (2011), *ie* after 11-16 months from the contamination, the course of *H. rufipes* trapping dynamics still indicated a negative effect of the engine oil (Fig. 2). Even 14 months after the contamination, half as many of these insects were trapped in the soil polluted with it than in the control soil. Similar as in the 2010 season, the peak number of the trapped specimen was "sharp" and fell in August, with very low number in May, June, September and October. Statistical analysis of the results from individual months indicated a significant effect of the applied bioremediation in case of the soil polluted with diesel oil. During the period of maximum presence of the insects, over twice more specimens were trapped in the soil contaminated with diesel oil and subjected to bioremediation than on the same object when the measure was not applied (Table 1). It was also reflected in the statistical analysis of results for the entire 2011 season (Fig. 4).

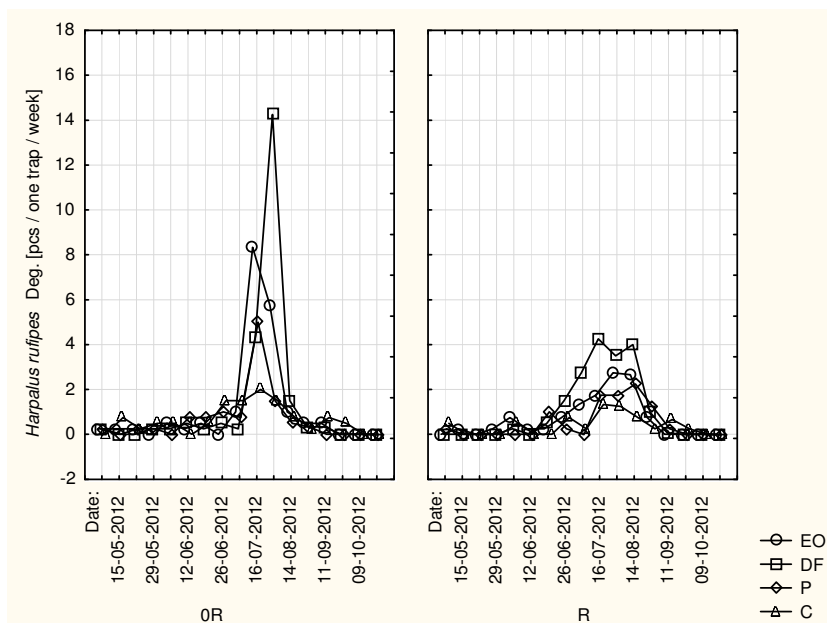


Fig. 3. Course of dynamics of *Harpalus rufipes* Deg. occurrence trapped using Barber's traps in 2012. The symbols as in Figure 1

In the 2012 season, *ie* after 23-28 months from the moment of the oil derivatives pouring, no negative effect of the pollutants on *H. rufipes* activity was registered. In the soil contaminated with diesel or engine oil at the peak number, even more of *H. rufipes* beetles were trapped than on the control (Fig. 3). In this case bioremediation was neutralizing, since the number of trapped insects in the objects polluted with diesel oil and engine oil after application of this measure was similar as in the unpolluted soil (Table 1). Similar relationships between the objects were obtained after subjecting the results obtained for the whole 2012 research period to statistical analysis.

*H. rufipes* Deg. is counted among the species which easily adapt to unfavourable conditions resulting from human activity. In the research on the effect of urbanization on the occurrence of Carabidae, the species was counted among the most numerous in the environment under the strongest human pressure [13]. *H. rufipes* was regarded also as one of the potential species which could be environmental indicator of Carabidae biodiversity. However, the research of Doring et al [2] revealed that the presence of none of the Carabidae species which they investigated were directly connected with this family species diversity. Huruk [1], who analysed the effect of soil chemical properties on the number of *H. rufipes* trappings, found a negative correlation between the number of captured specimens and C, K, Mg and N concentrations in the soil, whereas the correlation was positive for Ca. As results from his investigations *H. rufipes* is a species preferring soils with low content of organic matter, loose but warm. It may partially explain higher number of trappings in 2012 in the object with soil polluted with engine oil. It might result from a simultaneous decreasing of substances repellent for *H. rufipes* [14] and faster warming of

soil resulting from darker colouring of the polluted soil. On the other hand, Sadej et al [15], while analysing the influence of fertilization and soil tillage method on Carabidae occurrence, caught the greatest number of *H. rufipes* on the 12-year-old fallow. The other factors, such as N and C concentrations, soil pH or C:N ratio did not reveal any direct relation with this species occurrence.

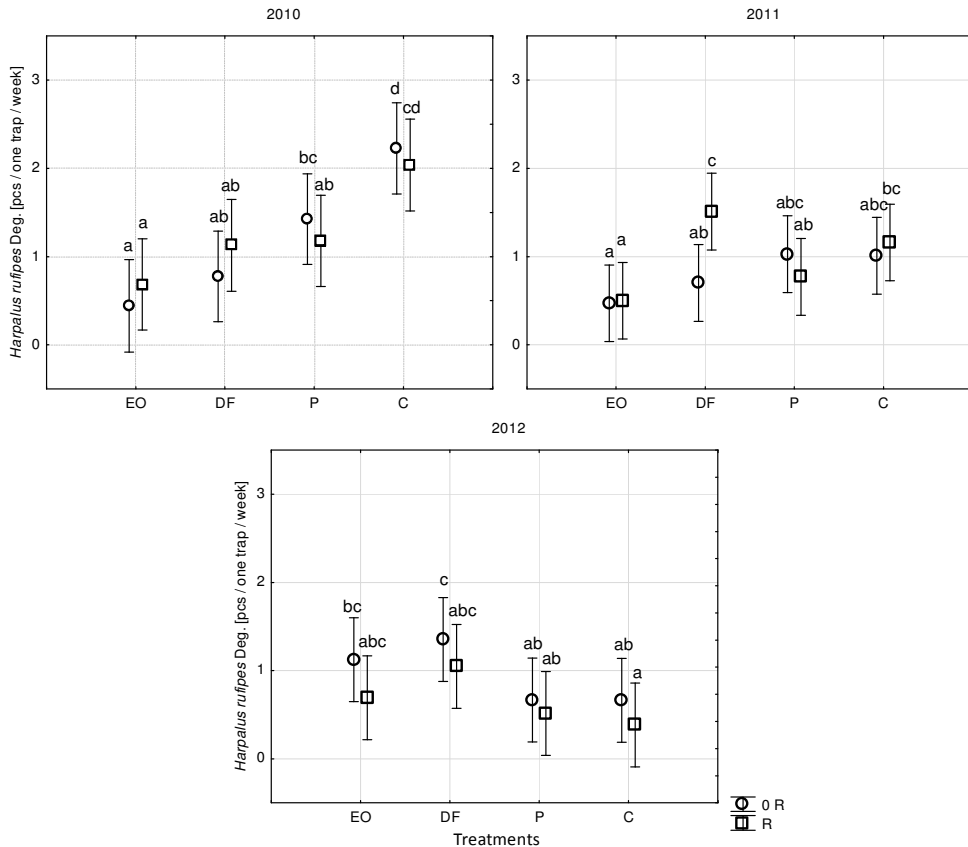


Fig. 4. Occurrence of *Harpalus rufipes* Deg. trapped using Barber's traps mean in the years 2010, 2011, 2012. The symbols as in Figure 1. Means marked with the same letters do not differ significantly according to NIR test at  $\alpha = 0.05$ ; factors contamination x remediation.  $\perp$  Mean  $\pm$  0.95 confidence interval

## Conclusions

1. Activity of *Harpalus rufipes* Deg. species representatives under conditions of soil polluted with oil derivatives depended on the kind of pollutant substance and on the time which passed from the moment of the soil pollution. Petrol had the least negative effect - it was visible only during the first four months after the pollution. Negative effect of diesel and engine oil was observed even 14 months from the moment of pollution.

2. Application of supported bioremediation on the soil polluted with diesel oil contributed to increasing the number of trapped *H. rufipes* beetles after 14 months from the moment of its application and from the moment of the soil contamination, whereas after two years the same measure neutralized the effect of intensified activity of the above mentioned beetles under conditions of the soil polluted with diesel and engine oil.

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## ODDZIAŁYWANIE ZANIECZYSZCZENIA GLEBY ROPOPOCHODNYMI NA WYSTĘPOWANIE *Harpalus rufipes* DEG.

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**Abstrakt:** Celem pracy było zbadanie oddziaływania substancji ropopochodnych w trakcie procesu bioremediacji gleby na przebieg dynamiki występowania *Harpalus rufipes* Deg. (Coleoptera, Carabidae). Utworzono następujące obiekty: kontrola - gleba niezanieczyszczona; gleba zanieczyszczona benzyną; gleba zanieczyszczona olejem napędowym oraz gleba zanieczyszczona zużytym olejem silnikowym (dawka: 6000 mg paliwa · kg<sup>-1</sup> s.m. gleby). Eksperyment został przeprowadzony w dwóch seriach: z naturalną i wspomaganą bioremediacją. *H. rufipes* był odławiany z użyciem pułapek Barbera w okresie od czerwca do października 2010 oraz od maja do października 2011 i 2012 roku. Aktywność przedstawicieli gatunku *Harpalus rufipes* Deg. w warunkach gleby zanieczyszczonej ropopochodnymi zależała od rodzaju substancji zanieczyszczającej oraz od czasu, jaki upłynął od momentu skażenia gleby. Najmniej negatywnie oddziaływała benzyna - jej wpływ widoczny był tylko przez początkowe 4 miesiące po zanieczyszczeniu. Negatywne działanie oleju napędowego i silnikowego obserwowano jeszcze po upływie 14 miesięcy od momentu skażenia. Zastosowanie bioremediacji wspomaganą na glebę zanieczyszczoną olejem napędowym przyczyniło się do zwiększenia liczby odławianych chrząszczy *H. rufipes* po upływie 14 miesięcy od momentu jej przeprowadzenia oraz od momentu skażenia gleby, natomiast po upływie dwóch lat zabieg ten neutralizował efekt zwiększenia aktywności wspomnianych chrząszczy w warunkach gleby poddanej zanieczyszczeniu olejem napędowym i silnikowym.

**Słowa kluczowe:** ropopochodne, gleba, bioremediacja, Carabidae, *Harpalus rufipes* Deg.