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BIOREMEDIATION OF CRUDE OIL DERIVATIVES IN SOILS NATURALLY AND ARTIFICIALLY POLLUTED WITH THE USE OF MAIZE AS THE TEST PLANT PART II. CROP YIELD

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Abstract. Contamination of soils artificially polluted with PAHs and aged polluted with crude oil in the phytoremediation process were compared. The plant used in the tests was maize (*Zea mays* L.). In chamber tests with soils artificially polluted (chernozem, calcareous rendzina, and lessives), selected three PAHs (anthracene, phenanthrene, and pyrene) were used at the doses of 100, 500, and 1000 $mg \cdot kg^{-1}$ d.m. of soil and diesel fuel at the doses of 0.1%, 0.5%, and 1% (v/v). Brown soil from crude oil refinery was chosen as the soil naturally, aged polluted with crude oil. Additionally, plant inoculation with the mixture of bacteria strains *Azospirillum* and *Pseudomonas stutzeri* was applied in the bioremediation process in the amount of 1 $ml \cdot 500\ g^{-1}$ of soil. Chamber pot-tests were carried out in controlled conditions (temperature, humidity) during a four-week-long plant growth period. Physical properties of soils and the dry mass of above- and underground parts of plants were analyzed in the work. The obtained results showed a statistically significant increase in the dry mass of the above- and underground parts of plants (both artificially and aged polluted) inoculated with *Azospirillum* and *Pseudomonas stutzeri* after maize growth.

Key words: above-ground and underground maize parts, anthracene, diazotrophic bacteria, phenanthrene, polycyclic aromatic hydrocarbons (PAHs), pyrene

INTRODUCTION

Soil pollution with crude oil derivatives (including polycyclic aromatic hydrocarbons; PAHs) has been a serious environmental problem for many years. Most difficulties are posed by the proper choice of remediation method due to cumulative properties of the soil. Remediation of soils polluted with PAHs most frequently is

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connected with two major problems: long biodegradation time of the above compounds and insufficient purification effect, as many intermediate products of PAH degradation do not undergo further degradation [Cerniglia 1992, Gogoi et al. 2003].

Crude oil derivatives pose a threat to the existence of living organisms and ecosystem functioning in the polluted areas. Many methods are used for the removal of pollutants from the soils or for limiting their negative effect on the environment. Accumulation degree of toxic compounds is faster than human actions taken in order to liquidate them, and therefore more and more attention is paid to the effective use of natural resources in order to reduce nature pollution. Thanks to scientific discoveries in the fields of physiology and biochemistry, fundamentals of environmental biotechnology were established, generally known as bioremediation and phytoremediation [Andreoni et al. 2007].

Bioremediation is a technology that uses microorganisms for environment decontamination, which thanks to great abilities to degrade chemical compounds and to transformation possibilities effectively decontaminate soil. Use of plant defensive mechanisms and the ability to metabolize toxic compounds is a basis for phytoremediation. It this technology, plants may be treated as a complex mechanism capable of soil, water, and air purification from pollutants which are heavy metals, organic compounds, and gases. Organic pollutant removal is based mainly on biodegradation, phytodegradation, and phytovolatization, where the major problem is the uptake of xenobiotics frequently lipophylic and bounded with soil components [Gűnther et al. 1996, Chauhan et al. 2008].

One of the conditions for well-conducted phytoremediation process is proper plant choice. For several years, trials have been conducted of the application of maize as the bioremediation plant in the decontamination processes of soils polluted with crude oil derivatives and heavy metals [Marchenko et al. 2001, Parales et al. 2002, Muratova et al. 2003, Parrish et al. 2005].

Many authors' works demonstrate the reasonableness of the application of bacteria strains capable of producing biosurfactants in PAH degradation processes [Parales et al. 2002, Zhou and Zhu 2007]. Production of surfactants by strain *Pseudomonas alcaligenes* significantly accelerated the bioremediation process of, for example, fluoranthene [Hickey et al. 2007]. Alasan obtained from *Acinetobacter radioresistens* strain KA53, in the dose of 500 $\mu g \cdot cm^{-3}$, increased the solubility in water of several hydrocarbons and thanks to this fact also caused twice faster mineralisation of fluoranthene and phenanthrene by *Sphingomonas paucimobolis* strain EPA505 [Barkovski et al. 1995]. It results from the studies by Kurek et al. [2001] and Król [1997, 2006] that such properties are displayed also by bacteria strains *Azospirillum* spp. and strains *Pseudomonas stutzeri* used in the experiment.

The aim of the experiment was the determination of the effect of the pollution of soils with crude oil derivatives in the conditions of their natural and artificial pollution on the yield of maize as a bioremediation plant.

MATERIAL AND METHODS

Effect of pollution of soils artificially polluted with polycyclic aromatic hydrocarbons (PAHs) and diesel fuel (ON) was studied, as well as of soil naturally polluted with crude oil in the phytoremediation process on plant yield expressed as the dry mass of their above- and underground parts. Detailed description of the experiment methodology is presented in Part I of the work [Gałązka et al. 2010]. Maize was chosen as the test plant. For artificial soil pollution, three model PAHs were applied: anthracene, phenanthrene, and pyrene (100, 500, and 1000 $mg \cdot kg^{-1}$ d.m. of soil) and diesel fuel (Multi Motor Oil Jasol 12 SG/CE 5W/4 originating from Jasło Refinery, JSC) at the concentration of: 0.1; 0.5; and 1% (v/v) d.m. of soil. Additionally, plant inoculation with bacteria mixtures *Azospirillum* and *Pseudomonas stutzeri* was applied in the amount of 1 ml per 500 g of soil. Bacteria strains originated from the collection of the Department of Agricultural Microbiology, Institute of Soil Science and Plant Cultivation – National Research Institute in Puławy. Pot tests (500 g of soil) were carried out in controlled conditions in a climatic chamber during a 4-week long plant growth period. Hydrocarbons were added in the form of solution in dichloromethane, and the control was soil with no PAHs with the addition of dichloromethane. After solvent evaporation, the soil was thoroughly stirred and moistened (to 60% of full water capacity). In the pots, pre-sprouted maize seeds were sown (4 plants per pot). After the completion of the 4-week plant growth period in the particular experiment combinations, basic physical properties of the soils were marked, as well as plant yield.

For the statistical assessment of the results, programme packet STATISTICA.PL (7) was used (Stat. Soft. Inc., 95% significance level). Regressions were calculated with the method of forward stepwise regression analysis at the condition of $F < 2$ (for the variable to enter the model).

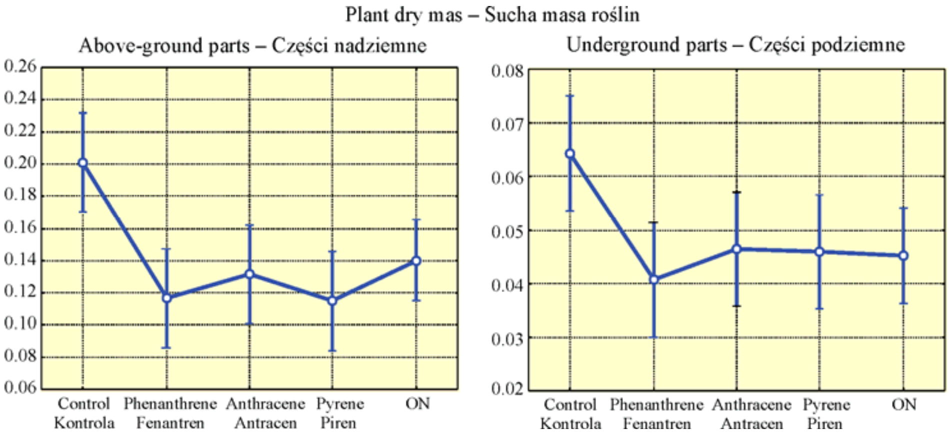
RESULTS

Effect of inoculation with *Azospirillum* spp. and *Pseudomonas stutzeri* on plant growth in the conditions of soil pollution with PAHs is presented as the analysis of two parameters: the dry mass of the above-ground parts and the dry mass of the underground plant parts.

On the basis of the analysis of variance (ANOVA, Table 1) average variables (of the above- and underground plant parts) in the polluted soils group, it can be stated that the mean values did not differ significantly statistically. It was found that in the case of the above-ground parts, the average value for the control differed significantly from all the average values except the one for diesel fuel. The remaining average values did not differ significantly between one another. Soil pollution with both PAHs and diesel fuel affected significantly plant yield expressed as the dry mass of the above- and underground plant parts (Fig. 1).

Table 1. One-way ANOVA ($P \leq 0.05$)
Tabela 1. Analiza wariancji jednoczynnikowej ($P \leq 0{,}05$)

Variable Zmienna	Soil – Gleba		Pollution – Skażenie		Inoculation – Szczepienie	
	F value wartość F	α	F value wartość F	α	F value wartość F	α
Dry mass – Sucha masa, mg						
Above-ground parts Części nadziemne	2.992	0.05093	5.147	0.00044	24.5118	0.00000
Underground parts Części podziemne	13.139	0.00000	2.829	0.02412	10.9128	0.00101



ON – diesel oil – ropa naftowa
expected edge means; Wilks' Lambda = .06325 – oczekiwane średnie brzegowe; Lambda Wilksa = ,06325
vertical columns signify 95% confidence intervals – pionowe słupki oznaczają 95% przedziały ufności

Fig. 1. Dry mass of the above- and underground plant parts (n = 70)
Rys. 1. Sucha masa części nadziemnych i podziemnych roślin (n = 70)

Table 2 presents Pearson's product-moment correlation coefficient of the physical properties of the soils with maize yield expressed as the sum of the above- and underground plant parts in the phytoremediation process. Strong correlations were found between the particular physical parameters of the soils in the case of maize inoculated with *Azospirillum* spp. and *Pseudomonas stutzeri*. Also, regression models were determined for the dependencies of maize yield variables (dry mass of the above- and underground parts) from the physical properties of the soils (Table 3). The obtained results made it possible to write the estimated regression model in the form of equations. In the presented dependencies, the models explained 54 to 98% of variability ($\check{y}_i$).

The above-ground plant parts were more sensitive to pollution than the underground parts. After soil pollution with the highest dose of anthracene, phenanthrene, and pyrene (1000 $mg \cdot kg^{-1}$) and 1% diesel fuel, statistically significant differences were found between the dry mass of the above-ground plant parts non-inoculated with *Azospirillum* spp. and *Pseudomonas stutzeri* and the inoculated plants. Similar observations concerned brown soil naturally polluted with crude oil (Fig. 2). In the case of the inoculated plants, the obtained results counted in percent in relation to the control showed a significant increase in the dry mass of the above-ground parts of the inoculated maize, particularly visible in the case of plant growth in calcareous rendzina (increase by circa 20-35%).

Table 2. Correlation coefficient values (r) between physical properties of soils and the dry mass of the above-ground plant parts in soils polluted with PAHs (1000 mg·kg^{-1}) and diesel fuel (1%)
Tabela 2. Wartości współczynników korelacji (r) pomiędzy właściwościami fizycznymi gleb a suchą masą części nadziemnych kukurydzy w glebach skażonych WWA (1000 mg·kg^{-1}) i olejem napędowym (1%)

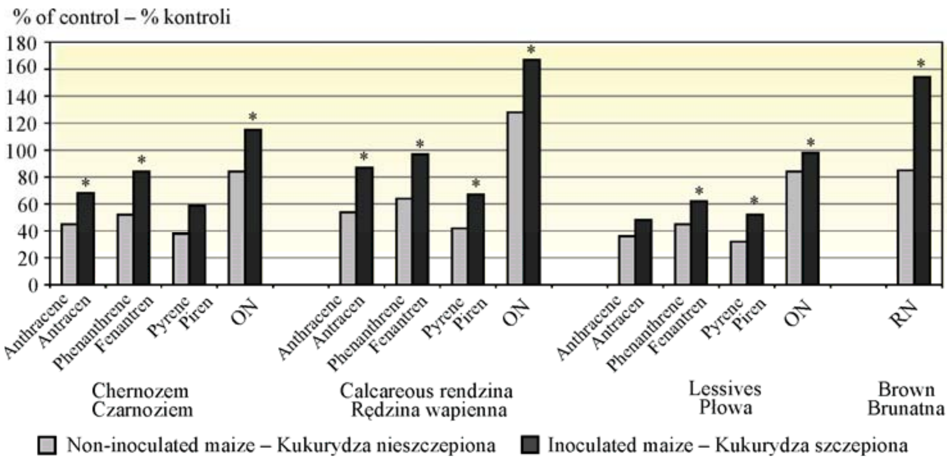
PAHs/diesel fuel WWA/ON	pH	C_{total} – $C_{og.}$ %	N_{total} – N_{og} %	P_2O_5	K_2O	Mg	N-NH_4	N-NO_3
Chernozem – Czarnoziem								
Non-inoculated maize – Kukurydza nieszczepiona								
Anthracene – Antracen	0.391	0.421	0.321	0.451	0.532	0.361	0.391	0.421
Phenanthrene – Fenantren	0.815*	0.831*	0.861*	0.831*	0.781*	0.891*	0.761*	0.691*
Pyrene – Piren	0.215	0.412	0.384	0.284	0.187	0.361	0.421	0.451
Diesel fuel – Olej napędowy	0.987*	0.854*	0.729*	0.742*	0.761*	0.712*	0.723*	0.781*
Maize inoculated with *Azospirillum* spp. and *Pseudomonas stutzeri* Kukurydza szczepiona *Azospirillum* spp. i *Pseudomonas stutzeri*								
Anthracene – Antracen	0.815*	0.784*	0.786*	0.452	0.567	0.394	0.875*	0.951*
Phenanthrene – Fenantren	0.942*	0.697*	0.859*	0.875*	0.964*	0.984*	0.978*	0.964*
Pyrene – Piren	0.961*	0.791*	0.915*	0.421	0.367	0.481	0.964*	0.987*
Diesel fuel – Olej napędowy	0.873*	0.894*	0.742*	0.789*	0.895*	0.874*	0.815*	0.891*
Calcareous rendzina – Rędzina wapienna								
Non-inoculated maize – Kukurydza nieszczepiona								
Anthracene – Antracen	0.875*	0.742*	0.421	0.461	0.317	0.512	0.748*	0.796*
Phenanthrene – Fenantren	0.945*	0.814*	0.871*	0.315	0.471	0.876*	0.948*	0.967*
Pyrene – Piren	0.475	0.347	0.367	0.486	0.477	0.475	0.615	0.518
Diesel fuel – Olej napędowy	0.875*	0.786*	0.791*	0.856*	0.915*	0.967*	0.894*	0.916*
Maize inoculated with *Azospirillum* spp. and *Pseudomonas stutzeri* Kukurydza szczepiona *Azospirillum* spp. i *Pseudomonas stutzeri*								
Anthracene – Antracen	6.78*	2.12*	0.134*	9.2*	18.2*	5.2*	0.99	0.945*
Phenanthrene – Fenantren	6.92*	2.16*	0.1448	9.4*	19.2*	4.9*	1.21*	0.912*
Pyrene – Piren	6.95*	2.23*	0.138*	9.6*	19.4*	5.4*	1.23*	0.872*
Diesel fuel – Olej napędowy	6.89*	2.32*	0.146*	8.9*	19.4*	5.6*	1.24*	0.961*
Lessives – Gleba płowa								
Non-inoculated maize – Kukurydza nieszczepiona								
Anthracene – Antracen	0.927*	0.877	0.785	0.324	0.452	0.361	0.215	0.451
Phenanthrene – Fenantren	0.924*	0.854*	0.791	0.521	0.526	0.589	0.785*	0.812*
Pyrene – Piren	0.696	0.921*	0.656	0.624	0.418	0.384	0.397	0.458
Diesel fuel – Olej napędowy	0.574	0.962*	0.914*	0.891*	0.781*	0.951*	0.966*	0.943*
Maize inoculated with *Azospirillum* spp. and *Pseudomonas stutzeri* Kukurydza szczepiona *Azospirillum* spp. i *Pseudomonas stutzeri*								
Anthracene – Antracen	0.992*	0.912*	0.845*	0.689*	0.748*	0.815*	0.748*	0.815*
Phenanthrene – Fenantren	0.987*	0.891*	0.869*	0.728*	0.915*	0.874*	0.815*	0.912*
Pyrene – Piren	0.981*	0.951*	0.967*	0.948*	0.877*	0.809*	0.943*	0.972*
Diesel fuel – Olej napędowy	0.784	0.934*	0.988*	0.924*	0.918*	0.843*	0.891*	0.967*
Brown soil – Gleba brunatna								
Crude oil – Ropa naftowa	0.944*	0.875*	0.877*	0.321	0.471	0.281	0.787	0.814*
Maize inoculated with *Azospirillum* spp. and *Pseudomonas stutzeri* Kukurydza szczepiona *Azospirillum* spp. i *Pseudomonas stutzeri*								
Crude oil – Ropa naftowa	0.987*	0.937*	0.988*	0.411	0.512	0.341	0.894*	0.912*

* statistically significant decrease in the content ($P \leq 0.05$) in comparison with the control in the particular soils – statystycznie istotny spadek zawartości ($P \leq 0,05$) w porównaniu z kontrolą w poszczególnych glebach
data is an arithmetic mean (n = 6) – dane są średnią arytmetyczną (n = 6)
control – kontrola – soil non-polluted with PAHs and diesel fuel – gleba nieskażona WWA i olejem napędowym

Table 3. Regression equations for the relationship between maize yield and chemical properties of soils ($P \leq 0.05$)
Tabela 3. Równania regresji dla zależności plonów kukurydzy od chemicznych właściwości gleb ($P \leq 0,05$)

Variable – Zmienna	Regression equation – Równanie regresji	R^2
Maize yield (dry mass of the above- and underground parts) Plon kukurydzy (sucha masa części nadziemnych i podziemnych)		
Non-inoculated maize – Kukurydza nieszczepiona		
N_{total} – $N_{og.}$, %	$\breve{y}_i = -622.622 + 1155.19\,x_i$	0.70
N-NH_4, mg N·kg^{-1} fresh m. of soil N-NH_4, mg N·kg^{-1} św.m. gleby	$\breve{y}_i = -622.622 - 0.436\,x_i$	0.78
N-NO_3, mg N·kg^{-1} fresh m. of soil N-NO_3, mg N·kg^{-1} św.m. gleby	$\breve{y}_i = -622.622 + 0.272\,x_i$	0.61
Inoculated maize – Kukurydza szczepiona		
N_{total} – $N_{og.}$, %	$\breve{y}_i = 517.05 - 2098.22\,x_i$	0.78
N-NO_3, mg N·kg^{-1} fresh m. of soil N-NO_3, mg N·kg^{-1} św.m. gleby	$\breve{y}_i = 517.05 - 3.89\,x_i$	0.90
Assimilable Mg, mg in 100 g d.m. of soil Mg przyswajalny, mg w 100 g s.m. gleby	$\breve{y}_i = 517.05 + 18.30\,x_i$	0.54
Assimilable K, mg in 100 g d.m. of soil K przyswajalny, mg w 100 g s.m. gleby	$\breve{y}_i = 517.05 + 0.80\,x_i$	0.94

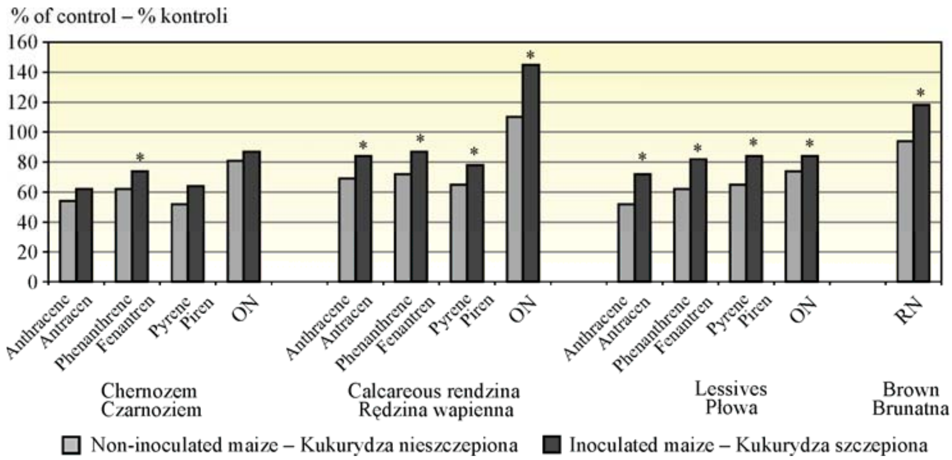
$\breve{y}_i$ – dependent variable values – wartości zmiennej zależnej
x_i – independent variable values – wartości zmiennej niezależnej
R^2 – determination coefficient – współczynnik determinacji



values statistically significantly different ($P < 0.05$) – wartości istotnie różne statystycznie ($P < 0,05$)
PAH dose (1000 mg·kg^{-1}), diesel oil dose (1% v/v) – dawka WWA (1000 mg·kg^{-1}), dawka oleju napędowego (1% v/w)
% in relation to dry mass yield from the control – % w stosunku do plonów suchej masy z obiektów kontrolnych

Fig. 2. Dry mass of the above-ground plant parts in soils artificially polluted with anthracene, phenanthrene, pyrene, and diesel oil (ON) and in brown soil aged polluted with crude oil (RN)
Rys. 2. Sucha masa części nadziemnych roślin w glebach sztucznie skażonych antracenem, fenantrenem, pirenem, olejem napędowym (ON) oraz w glebie brunatnej naturalnie skażonej ropą naftową (RN)

The underground plant parts to a smaller degree than the above-ground ones reacted to soil pollution. Statistically significant increase was found in the yield of the dry mass of the underground parts of the plants that grew on soils polluted with PAHs and diesel fuel in the case of their inoculation with bacteria suspention *Azspirillum* spp. and *Pseudomnas stutzeri*. Figure 3 compared the yield of the dry mass of the underground parts of non-inoculated plants and inoculated ones in the conditions of caclareous rendzina pollution with PAHs in the dose of 1000 $mg \cdot kg^{-1}$. Statistically significant ($P \leq 0.05$) increase (by circa 25-35%) in the underground plant parts in inoculation conditions was noted.

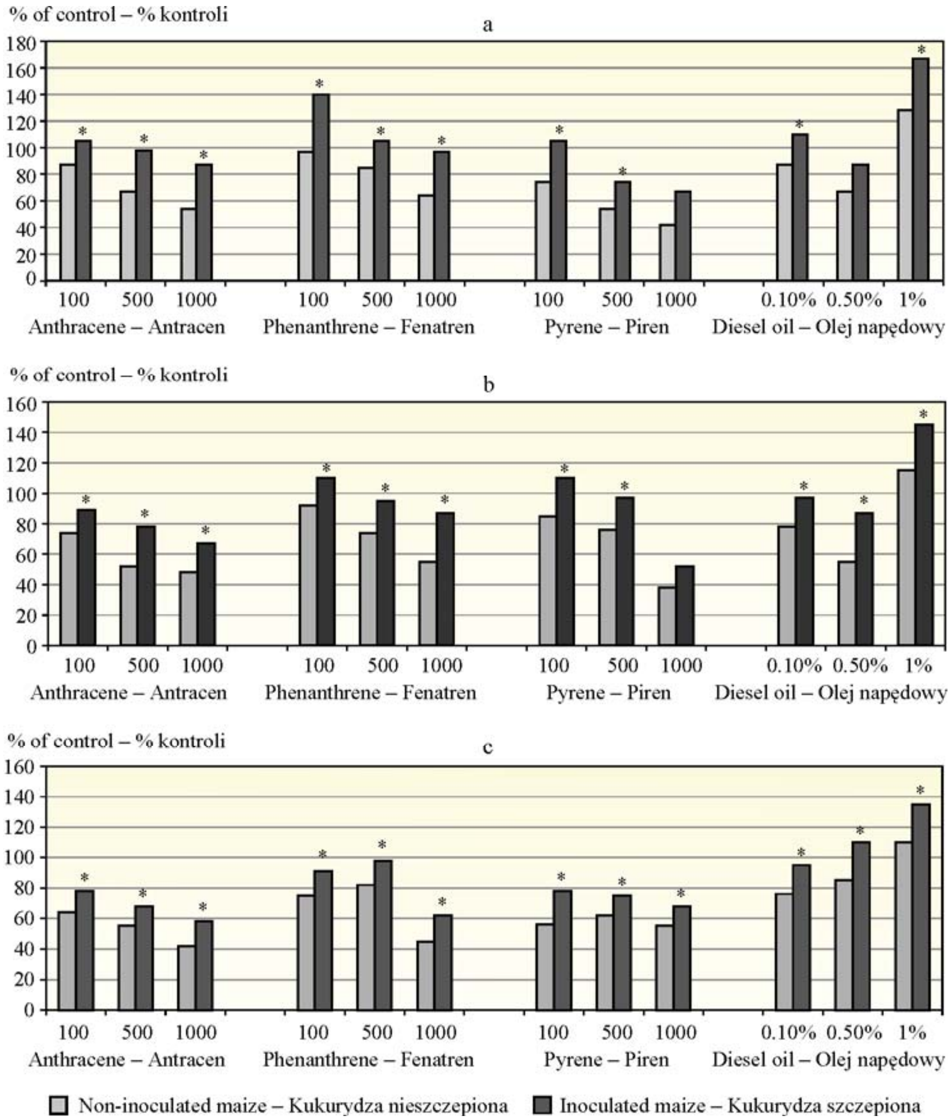


for explanations, see Fig. 2 – objaśnienia pod rys. 2

Fig. 3. Dry mass of the underground plant parts in soils artificially polluted with anthracene, phenanthrene, pyrene, and diesel oil (ON) and in brown soil aged polluted with crude oil (RN)

Rys. 3. Sucha masa części podziemnych roślin w glebach sztucznie skażonych antracenem, fenantrenem, pirenem, olejem napędowym (ON) oraz w glebie brunatnej naturalnie skażonej ropą naftową (RN)

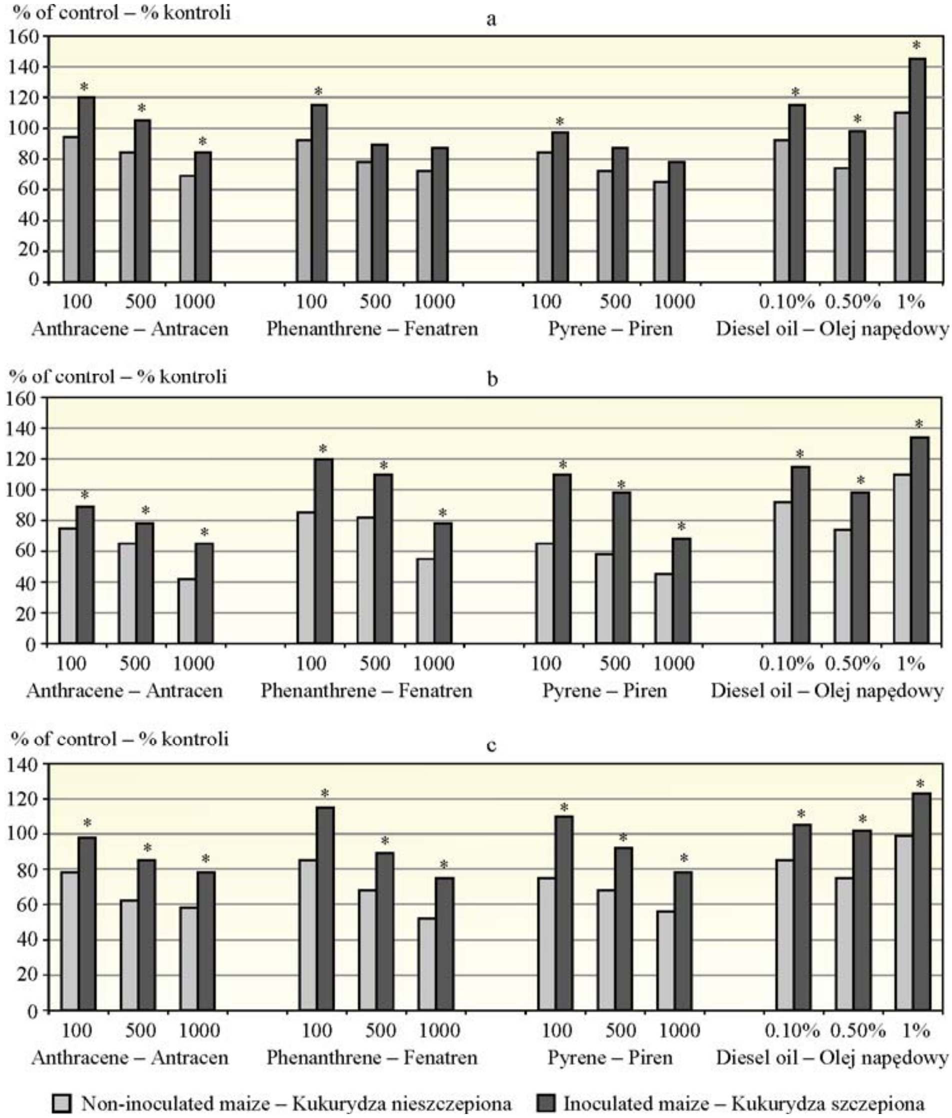
Statistically significant increase was stated in both the yield of the dry mass of the above-ground (Fig. 4) and the underground plant parts (Fig. 5) after the application of all the doses of soil pollution with PAHs and diesel oil. Moreover, statistically significant maize yield stimulation was noted in the case of the above- and underground plant parts after soil pollution with 1% diesel fuel (Figs 4 and 5).



for explanations, see Fig. 2 – objaśnienia pod rys. 2

Fig. 4. Dry mass of the above-ground plant parts in soils artificially polluted with anthracene, phenanthrene, pyrene, and diesel oil:
a) in calcareous rendzina artificially polluted with PAHs ($mg \cdot kg^{-1}$) and diesel oil (% v/v)
b) in chernozem artificially polluted with PAHs ($mg \cdot kg^{-1}$) and diesel oil (% v/v)
c) in lessives artificially polluted with PAHs ($mg \cdot kg^{-1}$) and diesel oil (% v/v)

Rys. 4. Sucha masa części nadziemnych roślin w glebach sztucznie skażonych antracenem, fenantrenem, pirenem i olejem napędowym:
a) w rędzinie wapiennej sztucznie skażonej WWA ($mg \cdot kg^{-1}$) oraz olejem napędowym (% v/w)
b) w czarnoziemie sztucznie skażonym WWA ($mg \cdot kg^{-1}$) oraz olejem napędowym (% v/w)
c) w glebie płowej sztucznie skażonej WWA ($mg \cdot kg^{-1}$) oraz olejem napędowym (% v/w)



values statistically significantly different ($P < 0.05$) – wartości istotnie różne statystycznie ($P < 0,05$)

Fig. 5. Dry mass of the underground plant parts in soils artificially polluted with anthracene, phenanthrene, pyrene, and diesel oil:
a) in calcareous rendzina artificially polluted with PAHs ($mg \cdot kg^{-1}$) and diesel oil (% v/v)
b) in chernozem artificially polluted with PAHs ($mg \cdot kg^{-1}$) and diesel oil (% v/v)
c) in lessives artificially polluted with PAHs ($mg \cdot kg^{-1}$) and diesel oil (% v/v)

Rys. 5. Sucha masa części podziemnych roślin w glebach sztucznie skażonych antracenem, fenantrenem, pirenem i olejem napędowym:
a) w rędzinie wapiennej sztucznie skażonej WWA ($mg \cdot kg^{-1}$) oraz olejem napędowym (% v/w)
b) w czarnoziemie sztucznie skażonym WWA ($mg \cdot kg^{-1}$) oraz olejem napędowym (% v/w)
c) w glebie płowej sztucznie skażonej WWA ($mg \cdot kg^{-1}$) oraz olejem napędowym (% v/w)

DISCUSSION

Soil and plants are inseparably connected with each other, and the relations that occur between them are characterised by many-sidedness and reciprocity [Barabasz and Vořišck 2002, Badura 2004]. Changes in soil composition affect plants directly, and indirectly also humans and animals. Searching for relations between the physicochemical properties of organic xenobiotics and their persistence in the soil environment was the topic of many studies [Maliszewska-Kordybach 1993, Bogan and Sullivan 2003, Smreczak and Maliszewska-Kordybach 2003]. In the present study, already after a short time (4-week-long experiment) statistically significant changes in the physical properties of the soils were found. They depended on both hydrocarbon properties and soil type and composition. Such relations in the course of PAH degradation in soils newly polluted with these compounds were also paid attention to in the works of other authors. Maliszewska-Kordybach [1993] showed that PAH degradation rate, particularly at the first stage after the introduction of these compounds into the soil (30-60 days) depends not only on hydrocarbon properties, but also on some soil properties: organic substance content, soil texture, and hydrolytic acidity, which during the course of the experiment often undergo changes.

Small solubility of hydrocarbons in water and their absorption on soil particles cause a considerable decrease in their availability, which significantly limits the bioremediation process [Field et al. 1995]. Low degradation effectiveness of these compounds does not result from too small metabolic activity of microorganisms that populate given soil but from small substrate availability, that is pollution components [Semple et al. 2003]. Application of detergents or other surfactants causes desorption and increases solubility of hydrophobic compounds in the water phase, and consequently increases biodegradation rate [Zhou and Zhu 2007]. Introduction of synthetic detergents may turn out to be toxic for the environment. Non-toxic biosurfactants produced by endogenous microflora that populate the polluted soil are characterised by better properties.

Maize, thanks to well-developed and dense root system, became proper habitat for the applied in the inoculation endophitic bacteria capable of using PAHs as the only source of carbon and energy.

In the root area of plants, an increased bioremediation rate of organic pollutants is observed in comparison with non-rhizospheric soil [Andreoni and Gianfreda 2007]. This is related first of all to the metabolic activity of microflora, which populates rhizosphere in great numbers. It turns out that of significant importance are also microorganisms directly connected with the plant that live inside root, stem, and leaf tissues. Examples of such microorganisms are strains *Azospirillum* spp. and *Pseudomonas stutzeri*. Rhizobacteria from genus *Azospirillum*, which fix free nitrogen and which are classified as optional endophytes capable of colonising both the external surface of the root and the inside intracellular space, favourably affect plant growth and development [Król 2006].

The conducted studies make it possible to preliminarily evaluate the effect of plant inoculation with *Azospirillum* spp. and *Pseudomonas stutzeri* as protective for plant yield in the conditions of soil pollution with crude oil derivatives. The effect was observed in all the studied soils newly polluted with PAHs, particularly in chernozem and calcareous rendzina, but also in lessives, slightly acid, with poor physical properties, as well as in brown soil aged polluted with crude oil.

CONCLUSIONS

1. Plant inoculation with *Azospirillum* spp. and *Pseudomonas stutzeri* stimulated maize growth (growth of the mass of the above- and underground parts) in the conditions of natural and artificial soil pollution with crude oil derivatives.
2. Diesel fuel used for soil pollution affected significantly the growth of dry mass yield of the above- and underground maize parts.

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BIOREMEDIACJA SUBSTANCJI ROPOPOCHODNYCH W GLEBACH NATURALNIE I SZTUCZNIE SKAŻONYCH Z WYKORZYSTANIEM KUKURYDZY JAKO ROŚLINY TESTOWEJ CZ. II. PLON ROŚLIN

Streszczenie. Badano wpływ skażenia gleb naturalnie i sztucznie skażonych wielopierścieniowymi węglowodorami aromatycznymi (WWA) i olejem napędowym (ON) na plon suchej masy nadziemnych i podziemnych części roślin kukurydzy (*Zea mays* L.). Do badań w przypadku sztucznego skażenia gleb wybrano: antracen, fenantren i piren, które zastosowano w dawkach: 100, 500 oraz 1000 $mg \cdot kg^{-1}$s.m. gleby oraz olej napędowy w stężeniu: 0,1; 0,5; 1% (v/w) s.m. gleby. W procesie bioremediacji zastosowano dodatkowo szczepienie roślin mieszaniną bakterii *Azospirillum* i *Pseudomonas stutzeri* w ilości 1 ml na 500 g gleby. Doświadczenia doniczkowe prowadzono w kontrolowanych warunkach w komorze klimatyzacyjnej podczas 4-tygodniowego okresu wegetacji roślin. Oznaczono podstawowe właściwości fizyczne gleb oraz plon roślin wyrażony suchą masą części nad-ziemnych i podziemnych. Stwierdzono statystycznie istotny wzrost plonu roślin w warunkach skażenia po zastosowanym szczepieniu roślin bakteriami. Stwierdzono statystycznie istotny przyrost plonu części nadziemnych i podziemnych w warunkach skażenia gleb 1% olejem napędowym. Szczepienie roślin oddziaływało ochronnie na plon roślin – zarówno części nadziemnych, jak i podziemnych w glebach sztucznie skażonych i w glebie naturalnie skażonej ropą naftową.

Słowa kluczowe: antracen, bakterie diazotroficzne, części nadziemne i podziemne kukurydzy, fenantren, piren, WWA

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