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EFFECT OF PETROLEUM DERIVATIVES CONTAMINATED SOIL ON GERMINATION AND EARLY GROWTH OF CHOSEN PLANTS

Summary

Most of products of petroleum have a toxic effects to plants, animals and also to man. Overworked motor oils and mixtures of different petroleum substances are especially dangerous for environment. The aim of this work was to determine the effect of overworked motor oil, diesel and their mixtures on the growth and advancement of chosen species of plants. The effect of increasing concentration of the petroleum substances in the soil (0.5, 1.0, 2.5, 5.0, 10.0, 25.0 ml kg⁻¹) on: seed germination, the elongation of shoot and root of barley, maize and mustard were studied. On the basis of research results it was stated that limited germination and growth of plants was caused by used petroleum substances and their mixture. Increasing the dose of xenobiotic caused reduction in the quantity of seeds or inhibition of germination as well as considerably limited lengths of roots and shoot of all tested plants. The studied species showed different sensitivity to the concentration of overworked motor oil. The highest toxicity was observed for mixture of overworked motor oil with diesel. Among all tested plants, the mustard was the most sensitive to the presence of petrochemical impurities. **Key words**: diesel oil, overworked motor oil, seed germination, barley, maize, mustard

WPŁYW ZANIECZYSZCZENIA GLEBY POCHODNYMI ROPY NAFTOWEJ NA KIEŁKOWANIE I POCZĄTKOWY WZROST WYBRANYCH ROŚLIN

Streszczenie

Większość z produktów naftowych oddziałuje toksycznie na organizmy żywe, w tym na rośliny, zwierzęta a także na człowieka. Szczególnie groźne dla środowiska są przepracowane oleje silnikowe oraz mieszanin różnych substancji petrochemicznych, np. diesla i przepracowanego oleju silnikowego. Celem pracy było określenie wpływu przepracowanego oleju silnikowego, diesla oraz ich mieszaniny na wzrost i rozwój wybranych gatunków roślin. Badano wpływ substancji ropopochodnych w glebie w dawkach 0,5, 1,0, 2,5, 5,0, 10,0 i 25 ml kg⁻¹ gleby na kielkowanie, wzrost części nadziemnych i podziemnych jęczmienia, kukurydzy oraz gorczycy. Na podstawie wyników przeprowadzonych badań stwierdzono, iż obecność diesla oraz przepracowanego oleju silnikowego wpływa na ograniczenie kielkowania i wzrostu roślin. Zwiększenie dawki ksenobiotyku spowodowało zmniejszenie ilości lub całkowite zahamowanie kiełkowania nasion, a także wpływało na znaczne ograniczenie wzrostu części nadziemnych i podziemnych roślin użytych w doświadczeniu. Badane gatunki wykazały zróżnicowaną wrażliwość na zanieczyszczenie gleby substancjami ropopochodnymi. Największą toksyczność wykazuje mieszanina diesla i przepracowanego oleju silnikowego.

Słowa kluczowe: diesel, przepracowany olej silnikowy, kiełkowanie nasion, jęczmień, kukurydza, gorczyca

1. Introduction

The development of civilization, accompanied by industrialization, has caused severe contamination of natural environment over extensive parts of many countries. Pollution of the soil and water with petroleum compounds is often observed in area where petroleum is obtained and soils around industrial plants [1, 7]. The contamination of soils also occurs while processing and distributing petroleum hydrocarbons as well as using petroleum products [5].

The presence of petroleum compounds in soils reduces the growth of plants, among others, by the inhibition of germination and growth, photosynthesis, and respiration processes. The anatomical changes of roots, deformation of cells, reduction in the amount of root hair, vascular obstruction, and oil accumulation in tissues and their dehydration were also observed [16, 23]. The petroleum derivatives change soil properties and also can lead to water and oxygen deficits as well as to shortage of available form of nitrogen and phosphorous [21]. Diesel reduces soil fertility and soil microflora population [18, 22]. The soil contamination with diesel strongly inhibits nitrification process and also causes a significant reduction in organic carbon content. Authors described a reduction in seed germination and primary root length of peanut, cowpea, sorghum, maize, rape, bean [2, 39, 13, 19]. This reduction depends on both level of contamination and plant species. Gasoline on the other hand is a complex mixture of organic compounds and it also has been shown to be toxic to plants [2, 19].

The phytotoxicity tests could be a way of finding potential bioindicators of the presence of petroleum products in the environment and an assessment method of the effectiveness of the conducted remediation treatments [15].

The effect of the individual petroleum products on plants was evaluated by many studies [2, 3, 8, 15, 22], but limited information is available on the combined effects of different petroleum products on plant species. There is the need to study the combined effects of petroleum products on plant because most of them are present in an environment at the same time or on same environment at different times. The objective of the study was to determine the toxic influence of diesel and overworked motor oil and their mixtures on barley (*Hordeum vulgare*), maize (*Zea mays*) and mustard (*Sinapis*).

2. Materials and methods

The experiment was conducted with three common crops and included barley (Hordeum vulgare), maize (Zea mays), mustard (Sinapis) which was treated with a diesel, overworked motor oil and their mixture along with a noncontaminated soil (control). Culture was carried out in vertical plastic PhytotoxkitTM containers (Phytotoxkit, Tigret company, Belgium). Onto the plates 100 g of soil were applied, and then a quantity of the analyzed substance was added. The experiments consisted of following sets: (1) control - soil without analyzed compounds, (2) soil contaminated with diesel (at 0.5, 1, 2.5, 5, 10, 25 ml·kg⁻¹ d.w.s), (3) soil contaminated with overworked motor oil (at 0.5, 1, 2.5, 5, 10, 25 ml·kg⁻¹ d.w.s.), (4) soil contaminated with diesel + overworked motor oil (at 0.5 ml \cdot kg⁻¹ d.w.s diesel + 0.5 ml·kg⁻¹ d.w.s overworked motor oil, 1.25 ml·kg⁻¹ d.w.s. diesel + 1.25 ml·kg⁻¹ d.w.s. overworked motor oil, 2.5 ml·kg⁻¹ d.w.s. diesel + 2.5 ml·kg⁻¹ d.w.s. overworked motor oil, 5 ml·kg⁻¹ d.w.s.. diesel + 5 ml·kg⁻¹ d.w.s. overworked motor oil, 12.5 ml kg⁻¹ d.w.s. diesel + 12.5 ml·kg⁻¹ d.w.s. overworked motor oil). The experiment involved five repetitions and three series for each variant. Phytotoxkit plastic containers were placed in the dark, constant parameters such as temperature $(25 \pm 1^{\circ}C)$ and humidity (80%) were maintained. On thus prepared soil, 10 seeds of plants were put (5 plates for each concentration of the test compounds). After the end of the experiment counts were made of the number of germinated seeds and measurements were taken of the root and shoot length.

On the basis of the obtained results the germination index (GI) for each treatment was calculated using equation [11]:

$$GI = \left[\left(\frac{G_x}{G_c} \right) \left(\frac{L_x}{L_c} \right) \right] \cdot 100\%$$
⁽¹⁾

where G_x and G_c are the number of seeds germinated in the sample and control, respectively whereas L_x and L_c are the length of root in the sample and control, respectively.

For the experiment, the soil was used having the following elemental composition: 81 mg P·kg⁻¹ of soil, 88 mg K·kg⁻¹ of soil, 69 mg Mg·kg⁻¹ of soil, pH of 5.92 (in KCl), C organic content of 1.01% (10.1 g·kg⁻¹ of soil), 2.4 mg N-NH₄·kg⁻¹ of d.w.s. and 9.2 mg N-NO₃·kg⁻¹ of d.w.s. Grain size distribution, particle size: 1 - 0.1 mm = 95%, 0.1 - 0.02 mm = 5%, < 0.02 mm 0%. Soil granulation was homogenous, composed of well-homogenized loamy sand. The influence of diesel oil and overworked motor oil was investigated using the phytotoxicity test based on the ISO-11269-2:2012 International Standard [10].

Petroleum diesel (EN 590:2004) was purchased from PKN Orlen, Poland. Overworked motor oil was obtained from local auto repair facility.

Average values were calculated for each analysed group. All phytotoxicity tests were carried out in three series and five replicates. Average values were evaluated with their standard deviations (SD). Results were plotted with Microsoft Excel software.

3. Results

Figure 1 demonstrates the germination index 6 days after sowing for barley, maize and mustard exposed to petroleum derivatives. The ability of the tested plants to germinate in soil contaminated with diesel, overworked motor oil and their mixtures varied and largely depended on the plant species and type of pollution. Increasing overworked motor oil and mixture of diesel with overworked motor oil concentration in the soil below 1.0 ml·kg⁻¹ d.w.s. significantly decreased seed germination, and at 2.5 ml·kg⁻¹ and below of petroleum derivatives in soil germination of analyzed plants was completely inhibited.



Source: own work / Źródło: opracowanie własne

Fig. 1. Influence of petroleum derivatives on the germination index of plants after 6-day germination. (a - soil contaminated with diesel, b - soil contaminated with overworked motor oil, c - soil contaminated with mixture of diesel and overworked motor oil)

Rys. 1. Wpływ związków petrochemicznych na indeks kielkowania roślin po 6 dniach uprawy. (a - gleba zanieczyszczona dieslem, b - gleba zanieczyszczona przepracowanym olejem silnikowym, c - gleba zanieczyszczona mieszaniną przepracowanego oleju silnikowego z dieslem) Slightly different situation was observed for plants which were sown in soil contaminated with diesel. The reducing in seed germination was observed for highest diesel concentration in soil (10 and 25 ml·kg⁻¹ d.w.s.), and seed of barley and mustard was completely inhibited, but maize seed germination was reduced to 60%. Among all tested plants, the mustard was the most sensitive to the presence of petrochemical impurities

The combination of the petroleum compounds contributed to a drop of the GI in the case of all tested plants, which was the most significant at the highest concentration of diesel, overworked motor oil and their mixture. Effective concentration of diesel, which caused 50% inhibition of root growth was: 17, 25 and 5 ml·kg⁻¹ d.w.s., respectively for barley, maize and mustard. Soil contaminated with overworked motor oil was more toxic and EC50% was: 3.5, 4.0 and 1.0 ml·kg⁻¹ d.w.s., respectively for barley, maize and mustard. The significant decreasing in the GI was most notable for mixed xenobiotic (diesel with overworked motor oil) and EC50% was 0.5 + 0.5 ml·kg⁻¹ d.w.s. diesel + overworked motor oil for all tested plants.

Figure 2 indicates the shoot length of barley, maize and mustard exposed to diesel, overworked motor oil and their mixture. The concentration of xenobiotics in the soil influenced due to the shoot length. Significant inhibition of shoot growth was observed for plants which were sown in soil contaminated with overworked motor oil and mixed xenobiotics (diesel with overworked motor oil). The influence on shoot length also depended on the level of contamination. Increasing xenobiotics concentration in the soil caused the decreasing in shoot growth. The overworked motor oil inhibited shoot length with a decrease from 16% to 100% in barley, 50% to 100% in maize and 26% to 100% in mustard at range concentration 1.0 to 2.5 ml·kg⁻¹ d.w.s. The mixture of petroleum compounds (overworked motor oil with diesel) significantly inhibited shoot length of barley, maize and mustard with a reduction from 86% to 100%, from 78% to 100% and from 45% to 100% at concentration range 0.5+0,5 ml $kg^{\text{-1}}$ d.w.s. diesel + overworked motor oil to 12.5+12.5 ml·kg⁻¹ d.w.s. diesel + overworked motor oil, respectively.

Root length was also negatively affected by petroleum compounds, especially overworked motor oil and mixture of diesel with overworked motor oil (Figure 3). Diesel slightly inhibited growth of root barley and maize, but significantly reduced mustard root by 80% at 10 ml·kg⁻¹ d.w.s. The presence of overworked motor oil in soil was more toxic than diesel and at 2.5 ml·kg⁻¹ d.w.s. reduced root length by 83% and 100%, respectively for maize, mustard and barley. Mixed xenobiotics reduced root length of barley and maize from 70% to 100% and mustard from 36% to 100% at range concentration from 0.5 + 0.5 ml·kg⁻¹ d.w.s. diesel + overworked motor oil to 12.5 + 12.5 ml·kg⁻¹ d.w.s. diesel + overworked motor oil.

4. Discussion

Petroleum derived products are important pollutants because they influence the functioning ecosystems. Petroleum derivatives are phytotoxic to plants at relatively low concentration. Petroleum products toxicity is an important factor governing germination and seedling growth of plants. The effects of toxic substances are dependent on both level of contamination and plant species. Germination and seedling establishment are vulnerable stages in the plant life cycle [20]. Seedling growth is considered as an indicator of petroleum products stress on plant ability to survive [12]. Diesel, overworked motor oil and their mixture treatment decreased seed germination percentage and most of the seedling growth parameters such as shoot and root length. Reduction in seed germination and seedling growth parameters of all tested plants provided evidence that the petroleum products such as diesel and overworked motor oil if present in the soil – plant environment are responsible for producing toxic effects which reduced plant development.



Source: own work / Źródło: opracowanie własne

Fig. 2. Influences of petroleum compounds contamination on shoot length of barley, maize and mustard. (a - soil contaminated with diesel, b - soil contaminated with overworked motor oil, c - soil contaminated with mixture of diesel with overworked motor oil)

Rys. 2. Wpływ zanieczyszczenia związkami petrochemicznymi na długość pędów jęczmienia, kukurydzy i gorczycy. (a - gleba zanieczyszczona dieslem, b - gleba zanieczyszczona przepracowanym olejem silnikowym, c - gleba zanieczyszczona mieszaniną przepracowanego oleju silnikowego z dieslem)



Source: own work / Źródło: opracowanie własne

Fig. 3. Influences of overworked motor oil contamination on root length of barley, maize and mustard. (a - soil contaminated with diesel, b - soil contaminated with overworked motor oil, c - soil contaminated with mixture of diesel with overworked motor oil)

Rys. 3. Wpływ zanieczyszczenia związkami petrochemicznymi na długość korzeni jęczmienia, kukurydzy i gorczycy. (a - gleba zanieczyszczona dieslem, b - gleba zanieczyszczona przepracowanym olejem silnikowym, c - gleba zanieczyszczona mieszaniną przepracowanego oleju silnikowego z dieslem)

The decrease in seed germination and seedling growth due to petroleum – derived substances is in conformity with the findings of other researchers [4, 17, 22]. Houshmandfar and Asli [9] observed spillage of mixture of diesel and gasoline threats to germination and seedling growth of wheat, barley, alfalfa and cover plants. Njoku et al. [12] observed reduction in germination percentage, shoot and root length of cowpea treated with gasoline and diesel fuel mixture.

The work Adedokun and Ataga [2] reported that treatment of soils with crude oil, automotive gasoline oil and spent motor oil significantly affected the time of germination, germination percentage, plant height and leaf production of cowpea. The toxic effect on germination percentage and seedling growth in this work could be viewed as a function of lower water and nutrient uptake related to water and nutrient bioavailability. The degrading effect of petroleum derivative compounds on soil leads to serve nitrogen and phosphorus depletion, reduction in water balance and biological equilibrium [6]. According to Adam and Duncan [1], the degree of germination inhibition and plant growth depends not only on the plant species, its cultivar, time of exposure, and contamination concentration, but also on the volatile content of the fuel fraction. Conventional fuel has an adverse effect on the water-air relations in the soil, creating an impermeable oily film layer around the seeds or roots and interfering with proper germination and growth of plants [1, 23]. If oil is sprayed on vegetation, it penetrates into the plant tissues, through the sensitive stomata, the thin cuticle and also through the epidermis. These penetrations are possible by its transfer through the vascular system of the plant. This is due to insufficient aeration of the soil because of displacement of air from the pore space between the soil particles by crude oil [19]. Our early studies [14] also show that the soil contaminated with diesel, sodium chloride and their mix are potentially toxic to the natural vegetation. We observed toxicity to maize, which seems to be dependent on the type and level of pollutant in soil. The highest concentration of sodium chloride and diesel reduced seed germination and shoot and root development. But higher toxicity was observed for shoot than root. Furthermore, the presence of several impurities in the soil causes an increase in the toxicity manifested through partial or complete inhibition of seed germination and plant growth.

5. Conclusion

Petroleum products polluting environment are an increasingly serious ecological problem as contamination of water and soil with these chemicals has adverse effect on the biochemical properties of soil and plant production. General observation in this study can conclude that diesel, overworked motor oil and mixture of these petroleum products produced toxic impact on seed germination and seedling growth of plants. Mixture application into the soil brought up changes in all of the determined germination and early seedling growth parameters. The identification of the effect of individual and mixed petroleum products on different plant species would be helpful for the establishment of an environment quality standard.

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