Impact of Selected Plant Species on Enzymatic Activity of Soil Substratum on Post-Mining Heaps

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ABSTRACT

The natural mineral resources (hard coal, sands, dolomites, lead and zinc ores) found in the Silesia and the excavation of them led to significant transformation or even degradation of the environment. The landscape of Upper Silesia was dominated by heaps created as a result of the accumulation of post-mining coal waste. These post-industrial sites are characterised by difficult conditions for the development of plant communities. Nevertheless, the heaps are spontaneously overgrowing and over time, a separate ecosystem can be observed (for heaps). The article analyzes the enzymatic activity of the substrate in relation to the selected dominant grass (*Monocots*) and herbaceous (*Dicots*) plant species. The aim of this study was to compare the activity of particular enzymes in soil substratum of the vegetation patches dominated by grass and herbaceous plants.

Keywords: carboniferous waste rock dumping grounds, mining heaps, enzymatic activities, herbaceous plants, grasses.

INTRODUCTION

The landscape of the Upper Silesia has been under strong impact of a man for many years. As a result of the exploitation of mineral deposits and the storage of waste coming from extraction and processing works, a lot of anthropogenic forms were created, such as spoil heaps or sedimentation pools. The spoil heaps are distinguished by specific harsh conditions (Bradshaw, 1993), such as: lack of water, high temperature, sometimes high salinity, low nutrient availability, small amount or lack of organic carbon (organic matter), small amount of nitrogen and macroelements (P, Mg, K) [Bradshaw, 2000; Rostański, Woźniak 2007; Woźniak et al., 2003, Woźniak, 2010; Markowicz et al., 2015]. Since in many places the process of vegetation development is very slow, such heaps are susceptible to water or wind erosion [Bradshaw,

2000; Rostański, Woźniak 2007; Woźniak et al., 2003, Woźniak, 2010; Chmura et al., 2013, Molenda et al., 2013, Woźniak et al., 2015]. Moreover, some of them are undergoing – thermal and chemical activities [Bradshaw, 2000; Rostański, Woźniak 2007; Woźniak et al., 2003, Woźniak, 2010, Piekarska-Stachowiak et al., 2014].

Due to these unfavorable physical and chemical conditions, the biological activity of the subsoil is very low [Snajdr et al., 2013] and ecological processes proceed slowly [Nicia et al., 2014, Urbanova et al., 2011]. Some of the heaps were subjected to mainly technical and biological reclamation, but many of them were left without management. Over time, spoil heaps are colonized by plants and animals as a result of natural succession [Cohn et al. 2001, Chmura et al. 2011, Rahmonov et al. 2011, Błońska et al., 2013, Kompała-Bąba and Bąba 2013, Chmura et al. 2013]. The majority of the spontaneously

developed vegetation is dominated by grasses. Grasses are frequently considered as being better adopted to post-industrial sites harsh site conditions [Prach 2003, Woźniak et al. 2003, Rostański and Woźniak 2007, Frouz et al. 2008, Rahmonov 2009]

The soil enzymes produced by microorganisms and fungi seem to be one of the most important indicator of the substratum quality of the soil.

They are responsible for many processes that take place in the substratum, such as nitrogen fixation, release and availability of mineral substances to plants, catalysis of processes related to energy and organic matter decomposition [Russel and Wyczółkowski 2005]. The question arises what (if any) is the influence of some dominant species on the enzymatic activity of over 30 years of heaps depending on the dominant species (*Calamagrostis epigejos, Poa compressa, Daucus carota, Tussilago farfara*). The grasses are considered to be pioneer vegetation, preparing the soil through a rich root system and giving the opportunity to develop a new habitat [Rostański and Woźniak, 2007; Baryła, 2005]. It was assumed that the activity of particular enzymes may be higher in the vegetation patches dominated by grasses species than in the vegetation patches where herbaceous vegetation prevails. The aim of this study was to compare the activity of particular enzymes in the soil substratum of the vegetation patches dominated by grass and herbaceous plants.

MATERIAL AND METHODS

Site description

Coal deposits have been exploited in Europe since the 18th century. As the result of this activity, the environment was seriously transformed and many anthropogenic forms appeared which resulted in the appearance of visible effects in the environment, such as the coal-mine spoil heaps [Różkowski and Różkowski, 2011]. It is estimated that the post-mining waste has been deposited in around 380 heaps, which cover over 2,000 ha [Szczepańska and Twardowska, 1999]. Over 6000 km^2 of the Upper Silesian Coal Basin (GZW) requires reclamation [Probierz et al., 2017]. This landscape forms are built with carboniferous sediments on the Pre-Cambrian crystalline massif [Cabała et al., 2004]. These objects are characterized by extreme abiotic conditions that change both in time and space and regulate the colonisation of heaps by plants [Woźniak 2010]. Most plants appeared on heaps using volatile predisposition of seeds (domination of anemochoric plants) and then were spread further. Three heaps „Sośnica" Heap (Gliwice) (50°16'22"N, 18°44'43"E), „Wesoła" Heap (Mysłowice) (50°10'28"N, 19°5'44"E) and „Kostuchna" Heap (Katowice) (50°11'4"N, 19°0'33"E) were selected for further investigations. The location of the considered heaps is shown in Figure 1. According to Kondracki's classification, all of them are situated in the central part of the Silesian Upland macroregion, in the Katowice Upland mesoregions [Kondracki, 2002].

The study area is located in the transition climate zone between the temperate continental climate in the east and temperate oceanic climate in the west. The average annual temperature varies between 7 and 9°C and the precipitation between 700 and 900 mm. The vegetation season lasts about 210–220 days [Kondracki, 2002].

The examined heaps are the landfills of carboniferous wastes, mainly made of claystone, siltstone, sandstone, conglomerate, coal shale and small quantities of coal [Rożkowski et al., 1993, Woźniak, 2010].

Methods

The vegetation samples were taken from heaps of the same age classes (over 30 years, according to the date of the end of material deposition), on which herbaceous vegetation appeared spontaneously [Woźniak, 2010].

In total, 75 research plots were established in the field in patches with the dominance of wood small-reed/bushgrass (*Calamagrostis epigejos*), flattened meadow-grass (*Poa compressa*), wild carrot (*Daucus carota*), coltsfoot (*Tussilago farfara*) and without vegetation (control).

For each of the plots, the list of species and species abundance (the coverage of individual species) were determined. Moreover, soil samples were taken from each plot at the depths between 0 and 10 cm. In the laboratory, the soil samples were sieved through a 2 mm mesh sieve and cooled at 4°C degree until chemical analyses were carried out.

Acid and alkaline phosphatase were determined by means of spectrophotometric methods with 115 mM of p-nitrophenylphosphate solution
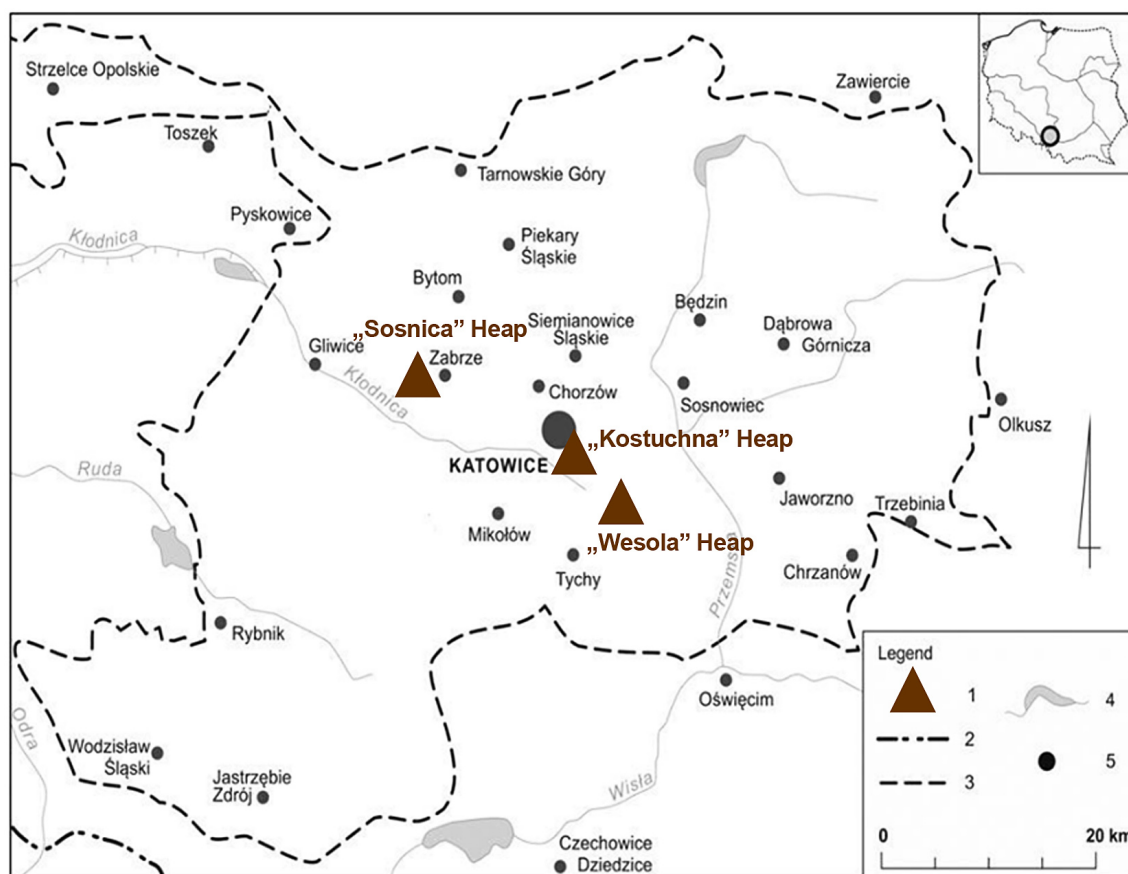
Figure 1. Location of investigated coal mine spoil heaps (marked in colour). Explanations: 1 – investigated heaps, 2 – State border, 3 – Area of Silesian Upland macroregion, 4 – water reservoirs, 5 – cities

as a substrate [Schinner et al., 1996]. Similarly, the urease activity was determined using urea as a substrate [Hoffmann, 1968]. The dehydrogenase activity was determined by using TTC, i.e. 2,3,5-triphenyltetrazolium chloride as a free radical acceptor [Schinner et al., 1996]. The activity of dehydrogenases was expressed in µg TPF g^{-1} d.m. h^{-1}, urease in µg N-NH$_4$ g^{-1} d.m. h^{-1} and acid phosphatase and alkaline phosphatase in µg NP (nitrophenol) g^{-1} d.m. h^{-1}. All enzymes were determined with a UV-VIS DR 5000 HACH-Lange spectrophotometer. The activity of dehydrogenases was measured at the wavelength of $\lambda = 546$ nm, urease at $\lambda = 580$ nm and that of acid and alkaline phosphatase at $\lambda = 410$ nm.

Statystical analyses

In order to obtain the normal distribution of data we used the Box-Cox transformation. The differences between investigated soil enzymes activity were tested using ANOVA with Welsh correction, and wherever significant differences were identified, a *post hoc* Tukey test was performed.

RESULTS AND DISCUSSION

The soils from the control and plots covered by the studied plant species differ significantly in terms of dehydrogenase ($F_w = 7.358$; $p = 0.000$) (Figure. 2) as well as acid ($F_w = 21.770$; $p = 0.000$) (Figure. 3) and alkaline ($F_w = 35.831$; $p = 0.000$) (Figure. 4) phosphatase activity. Only in the case of urease ($F_w = 3.293$; $p = 0.150$) (Figure. 5), no statistically significant differences in the activity of this enzyme were found in the studied soils. Higher activity for alkaline and acid phosphatases was recorded for the grasses, then herbs herbaceous vegetation plots. A similar situation was observed for the dehydrogenase activity.

Plots dominated by monocotyledonous species were characterized by a higher dominant coverage (*C. epigejos* – 48%, *P.compressa* – 33%), in comparison with plots dominated by the dicotyledonous species (*T. farfara* – 29%, *D. carota* – 24%) (Figure 6).

The well-developed root system has a positive effect on macroaggregates formation, soil organic matter (SOM) accumulation and soil stabilization
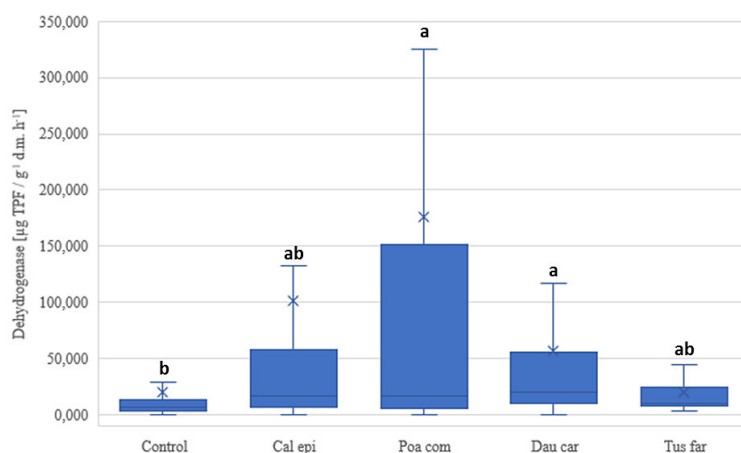
Figure 2. Comparison of the dehydrogenase activity in substrate samples from plots dominated by *Calamagrostis epigejos (Cal epi)*, *Poa compressa (Poa com)*, *Daucus carota (Dau car)* and *Tussilago farfara (Tus far)* and controls. Results marked with the same letter do not differ statistically significantly
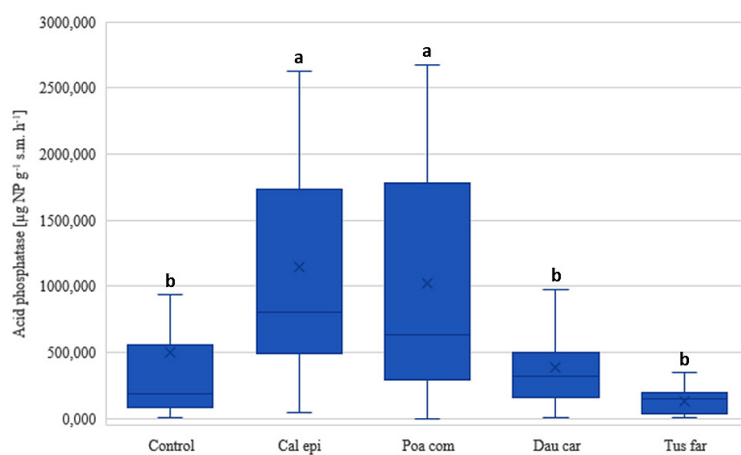

Figure 3. Comparison of the acid phosphatase activity in substrate samples from plots dominated by *Calamagrostis epigejos (Cal epi)*, *Poa compressa (Poa com)*, *Daucus carota (Dau car)* and *Tussilago farfara (Tus far)* and controls. Results marked with the same letter do not differ statistically significantly
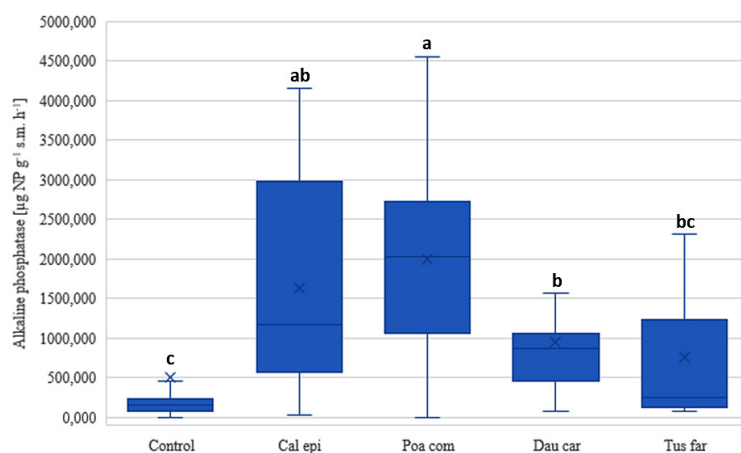

Figure 4. Comparison of the alkaline phosphatase activity in substrate samples from plots dominated by *Calamagrostis epigejos (Cal epi)*, *Poa compressa (Poa com)*, *Daucus carota (Dau car)* and *Tussilago farfara (Tus far)* and controls. Results marked with the same letter do not differ statistically significantly
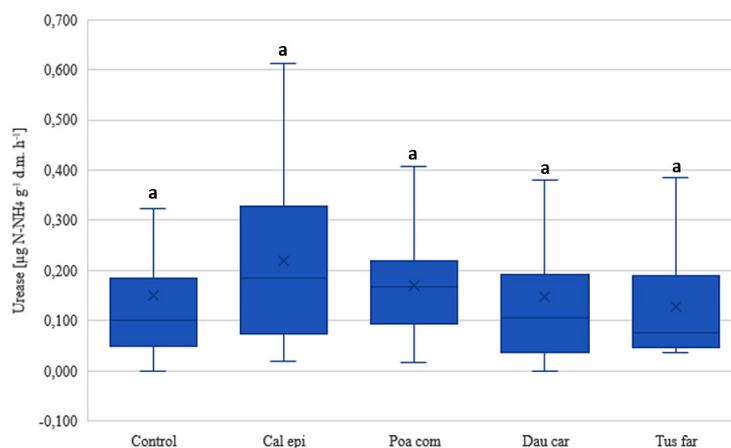
Figure 5. Comparison of the urease activity in substrate samples from plots dominated by *Calamagrostis epigejos (Cal epi)*, *Poa compressa (Poa com)*, *Daucus carota (Dau car)* and *Tussilago farfara (Tus far)* and controls. Results marked with the same letter do not differ statistically significantly
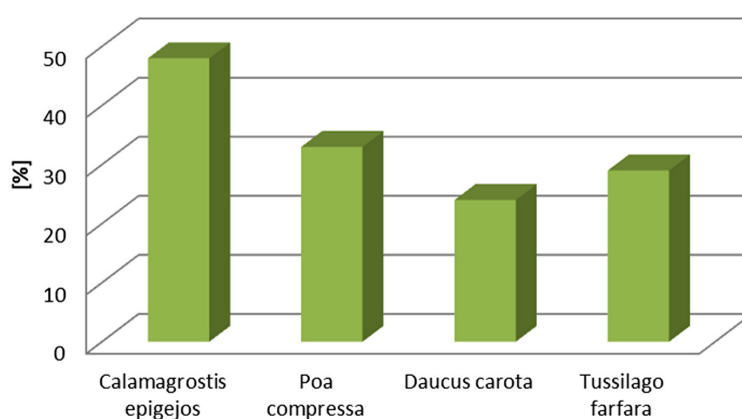

Figure 6. Comparison of the dominant species coverage

[Oades, 1984]. Moreover, the high abundance of plants can stimulate activity and biodiversity of microorganisms [Eisenhauer, 2013; Eisenhauer et al., 2017]. Therefore, the higher enzyme activity in grass dominated vegetation plots may be caused by high coverage and more developed grass root system, which generates the root exudates stimulating fungi and other microorganisms communities in the rhizosphere [Mommer et al., 2010; Mueller et al., 2013; Ravenek et al., 2014, Eisenhauer et al, 2017]. The studies conducted by Haynes [1999] have shown that the presence of grass increases the amount of SOM and improves the activities of the enzymes involved in N, P and S cycling.

The dehydrogenase activity is an indicator of the redox system and a measure of respiratory activity of microorganisms [Kieliszewska-Rokicka, 2001; Bierza et al. 2012]. Dehydrogenase is active only within living organisms, and after a cell death,

their degradation follows quickly. Therefore, the dehydrogenase activity indicates the presence of physiologically active microorganisms [Dick, 1997]. Higher dehydrogenase activity in soils from plots dominated by grassy species may be associated with a large amount of mycelium in the soil [Caravaca et al. 2004], because as mentioned above, the extensive root system of grasses stimulates the development of arbuscular fungi.

In soil ecosystems, phosphatases play a key role in the phosphorus cycle by catalysing the hydrolysis of organic phosphorus compounds to its inorganic forms, directly accessible to plants and soil organisms [Das and Varma, 2010]. The research conducted by Waldrop et al. [2000] and Ushio et al. [2010] indicate a positive correlation between the amount of mycelium in the soil and the activity of acid phosphatase. These authors suggest that fungi, compared to other microorganisms, can produce vastly more acid phosphatase.

CONCLUSIONS

Higher enzymatic activity was observed in the sites with the dominance of grasses, compared to the sites with the plants of the *Dicots* families or the control sample. This may be due to the stimulating effect of the extensive root system of grasses, and thus a large amount of root exudates stimulating the development and activity of microorganisms. Lower enzyme activity on the plots of *Daucus carota* and *Tussilago farfara* may also be associated with lower abundance (biomass) of these species, in comparison to grasses.

On the basis of research and literature, it can be concluded that the enzymatic activity depends on the presence of vegetation and affects the formation of soil properties [Myśków, 1981, Kobus, 1995, Koper and Piotrowska, 1996, Myśków, 1996]. This is an example of a natural spontaneous revegetation, i.e. revival of a degraded post-industrial area.

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