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Evaluation of the effect of toxicity of biochar used for soil fertilization and its water extract on plants

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ABSTRACT

There are many publications indicating that biochar can stimulate germination, growth, and plant yield. However, the mechanism of biochar activity in the soil remains unclear. Harmful compounds that may be present in biochar (improperly prepared or obtained from contaminated biomass) can have a toxic effect on the soil environment and cultivated plants. The aim of this study was to show whether the analyzed biochar obtained from an energy plant (*Sida hermaphrodita*) can be phytotoxic after introduction into the soil and what dose can effective. Plant toxicological tests (using *Lepidium sativum*) were carried out on water extracts from the biochar alone to assess its properties. The aim of further research was to analyze the effect of the use of biochar for soil amendment on key stages of plant development such as germination, growth, and yielding of selected species. The following were used for vegetation tests: garden cress (*L. sativum* L.), Virginia fanpetals (*S. hermaphrodita*), maize (*Zea mays*), white mustard (*Sinapis alba*), and common sunflower (*Helianthus annuus*). Biochar was used to enrich sandy soil in doses of 0.5%, 1%, and 2%. After the end of plant vegetation, plant yield, and basic physical and chemical properties of soils were evaluated. The results of phytotoxicity tests of water extracts from biochar and sandy soil fertilized with the biochar studied showed that 0.5% and 1% were optimal doses of biochar (which did not cause noticeable germination disorders).

Keywords: Sida hermaphrodita; Biomass; Biochar; Toxicity; Soil; Fertilization

1. Introduction

The author of the paper proposed an innovative method of the analysis of phytotoxicity changes caused by the content of undesirable components in biochar used for fertilizing purposes. The effect of water extracts from biochar and soil enriched with biochar on germination and growth of selected plant species was analyzed.

Biochar production can be compared with charcoal production. The properties of both materials are very similar. The preconditions for biochar production are high temperature (350°C–700°C) and limited oxygen supply. The substrate is organic matter, for example, agricultural biomass, energy crops (e.g., *Salix viminalis* and *Miscanthus* sp.), wood waste (sawdust, bark), and waste from the agrifood industry. The cultivation of *Helianthus annuus* and *Sida hermaphrodita* (analyzed in this paper) is also very prospective for the purpose of biochar production. These energy plants are characterized by fast growth and a large amount of biomass produced.

Process conditions also allow for the use of raw materials such as sewage sludge and the organic fraction of municipal waste. Pyrolysis of organic materials generates solid, liquid, and gaseous products (all with energy value), that is, biochar, oil, and synthetic gas, respectively. The proportions between these products and their properties depend on the

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process conditions (temperature and its growth rate, interaction time, and pressure), and substrate parameters [1].

Biochar may become one of the most important products of the present civilization for sustainable development. Its properties can be used for the improvement of soil quality, in the broadly understood sectors of agriculture, energy production, construction, and other types of industry. According to scientific research, most agricultural soils have lost an average of 30%–75% of their organic carbon compared to soils before the era of fertilizers. In order to restore the primary contents in the soil, efforts should be made to apply 30 to 40 t/ha biochar [1,2].

Scientific research has shown that the addition of biochar to soil most often improves its fertility and crop yielding. Soil amendment with this material increases the water capacity of the soil and soil reaction [3,4]. Currently, many studies are being conducted on the use of biochar to limit the effect of soil degradation [1].

Depending on the properties and suitability for use, biochar can be considered an organic fertilizer or soil conditioner. The procedure for the approval of biochar in Poland as a fertilizer or soil improver is the same as for fertilizers and conditioners. It requires a marketing authorization in accordance with the applicable legal acts, that is, the Act of 10 July 2007 on fertilizers and fertilization [5] and the Ordinance of the Minister of Agriculture and Rural Development of 18 June 2008 on the implementation of certain provisions of the Act on fertilizers and fertilization [6].

The Ordinance does not take into account pollutants such as polycyclic aromatic hydrocarbons (PAHs), furans, dioxins, or polychlorinated biphenyls, with their quantity defined by US standards. PAHs, which can be formed during the pyrolysis process, are considered to be particularly hazardous. These compounds can, for example, cause a decrease in yielding and be accumulated in plant biomass. Therefore, in addition to monitoring of properties of biochar used as a fertilizer, it is advisable to control the changes occurring in the soil environment of the fertilized biochar by means of biotests. Different plants are used for the purpose. One of the most frequently used plant indices (biological tests) is garden cress (*Lepidium sativum* L.). This plant is often used as a bioindicator to determine the effect of various chemical compounds on germination and growth of plants [7].

In the present study, the problems of the evaluation of physical and chemical properties of biochar obtained from the energy plant (Virginia fanpetals *S. hermaphrodita*) and its usefulness for soil fertilization purposes were discussed. This is a very promising species which can be used for energy and industrial purposes. So far, there have been few studies on such a use of this plant [8].

The aim of the study was to assess the effect of the application of biochar produced at the combustion temperature of 350°C and 450°C on the yielding and quality of biomass of selected plants. Biochar formed at two different temperatures during the pyrolysis process was examined (as part of the research project) to determine the quality and fertilizing suitability of the obtained material. The likelihood of phytotoxicity of both biochars was analyzed. These products may contain various undesirable components such as heavy metals, PAHs, polychlorinated biphenyls, and various tarry compounds, which can migrate into soil water and adversely affect germination, growth, and plant yielding.

The effect of toxic properties of biochar and its water extracts on selected test plants (garden cress *L. sativum* L.), Virginia fanpetals (*S. hermaphrodita*), maize (*Zea mays*), white mustard (*Sinapis alba*), and sunflower (*H. annuus*) was analyzed.

2. Material and methods

2.1. Examinations of biochar and fertilized soil

Before the experiment was started, the physical and chemical composition of biochar formed after the pyrolysis process of *S. hermaphrodita* were examined at 350°C and 450°C. The results were compared with the recommendations for materials used for fertilization purposes in Poland. They were also compared with the standards used in other European countries and the USA [4].

Biochar samples were pre-dried at room temperature and sieved through a sieve with a 2 mm mesh. Next, they were dried at 105°C to constant weight, ground in a mortar and sieved again through a sieve with a mesh diameter of 0.6 mm. Three samples were prepared for analysis.

The following determinations were made [9]:

- pH in water: potentiometric method,
- pH in KCl: potentiometric method,
- total carbon (TC): determined by means of a Multi N/C 2100, Analytik Jena AG, Jena (Germany) carbon analyzer,
- Kjeldahl nitrogen – determined by distillation after prior mineralization of samples in Buchi K-435 mineralizer,
- content of organic matter: weighing method,
- content of available forms of phosphorus: the Egner–Riehm method,
- total phosphorus: determined by means of the spectrophotometric method with ammonium molybdate (samples were previously mineralized in a Berghof high-pressure microwave mineralizer).

The total contents of heavy metals, that is, mercury (Hg), copper (Cu), lead (Pb), cadmium (Cd), zinc (Zn), nickel (Ni), chromium (Cr), arsenic (As), and molybdenum (Mo) were also evaluated. Aqua regia (a mixture of concentrated hydrochloric and nitric acids at a volumetric ratio of 3:1) was used for extraction of metals. Mineralization was conducted at 180°C for 30 min, using a high-pressure microwave mineralizer (Berghof, Germany).

The contents of heavy metals were evaluated by means of an inductively coupled plasma optical emission spectrometers SPECTRO ARCOS FHX22.

Soil samples were similarly prepared and studied.

Furthermore, the content of PAHs was also determined (despite the lack of national requirements).

However, PAHs were also determined due to the presence of tarry compounds (which can be toxic for example, plants and soil organisms) in water extracts of the tested biochar.

The initial stage before chromatographic determination of PAHs in biochar was extraction with organic solvents of different polarity. The separation of the organic matrix was carried out by means of sonolysis using a mixture of cyclohexane and dichloromethane solvents (5:1 v/v). Separation of solvent extracts from the sample was carried out with a high-speed centrifuge. Silica gel was used to isolate the analyzed components from the extracts from other simultaneously extracted organic substances, with purification conducted under vacuum conditions. The treated extracts were concentrated in the nitrogen stream. The determination was performed using a gas chromatograph coupled to a mass spectrometer (Fisons GC800/MS800). The study determined 16 PAHs indicated on the EPA list for environmental analyses [10]. These were naphthalene, acenaphthylene, acenaphthene, fluorene, phenanthrene, anthracene, fluoranthene, pyrene, benz[a]anthracene, chrysene, benzo[b] fluoranthene, benzo[k]fluoranthene, benzo(a)pyrene, dibenz [a,h]anthracene, benzo[ghi]perylene, and indeno[1,2,3-cd] pyrene.

2.2. Examinations of the potential toxicity of biochar in relation to selected plants

The effect of biochar on the key stages of plant development, such as germination, root growth, or shoot weight was analyzed.

Toxicological studies of biochar were performed in two variants. The first type of plant biotest was to determine the toxicity of this product in relation to plants. Toxic properties of biochar (its water extract) before its introduction into the soil were examined. In the second stage, the effect of properties of soil water extracts after the introduction of different doses of biochar on germination and growth of plants was investigated.

In the first stage of toxicological tests, the plant test was performed in accordance with the procedure [7] modified for biochar. It allowed for the analysis of the effect of compounds contained in biochar and soluble in water on germination and root growth of *L. sativum*. In order to perform the test, 20 g of biochar was placed in six replicates in Petri dishes for each combination. The weighed amounts were then saturated with distilled water exceeding their total water capacity by 2 mL and stirred thoroughly. After the balance in the solution was established, the biochar was covered with a paper disk. Ninety *L. sativum* seeds were sown from each combination on three plates prepared in this way. On the remaining three, 10 seeds were sown, and they were incubated at 22°C. After 2 d, the percentage of germinated seeds on all plates was calculated. On plates containing 10 seeds each, the length of shoot roots was also measured after 2 and 4 d of growth. The control test was obtained by sowing the same amounts of seeds on Petri dishes with paper discs moistened with distilled water. Furthermore, the same tests were used in this experimental variant to check the species reaction for mustard seeds (*S. alba*), which is also used as a test organism.

A plant test developed by Masciandaro [11] was performed in the second stage of toxicological tests. The aim of this test was to determine the effect of all the conditions created after the introduction of biochar into the soil water on the development of garden cress (*L. sativum*). The 50 g weighed amounts of soil placed on Petri dishes (with the addition of biochar in the amount corresponding to 0.5%, 1%, and 2%) with the moisture content of 60% of the total water capacity were sown in three repetitions with 100 garden cress seeds each. After 4 d, the number of germinated seeds was calculated and the weight of fresh seedlings was evaluated. Based on these parameters, the growth index was calculated according to the formula:

$$\text{GI\%} = P\,(T{:}C) \tag{1}$$

where P is the mean percentage of germinated seeds in soil with biochar addition relative to the mean value of the control soil, T is the mean weight of *L. sativum* shoots grown on the soil with biochar, C is the mean weight of *L. sativum* shoots grown on the control soil.

2.3. Vegetation tests of plant yielding on substrates fertilized with biochar

Sandy soil was applied for biochar fertilization in the vegetation tests. Biochar was introduced with the amounts of 0.5%, 1%, and 2% (by weight). The control material was non-fertilized soil. The following materials were used in the vegetation tests: Virginia fanpetals (*S. hermaphrodita*), maize (*Z. mays*), white mustard (*S. alba*), and sunflower (*H. annuus*). The study was conducted under conditions of pot experiment in a foil tunnel. The pots used for the study had a capacity of 2 L. Toxicological tests (described in Chapter 2.2) of fertilized substrates were previously performed using test plants (on Petri dishes).

Maize, mustard, and sunflower were sown to each test object at the recommended doses. *S. hermaphrodita* was planted with four seedlings per pot. Seedlings from seed sowing were used for planting. The research used commercial cuttings with the length of the overground shoot length of 10 cm. They were characterized by balanced growth. During the experiment, the constant moisture content in the substrates (60%) was maintained in the pots (evaluated by weight method). The experiments were conducted for a period of 6 months of vegetation (May–October 2018) with three repetitions. All the results are represented by the means from these repetitions. After the end of the experiment, the amount of biomass (by weight) of individual plant species from each fertilization combination was examined. Analysis of the results was performed by means of a statistical software package STATISTICA 9.0. The significance of statistical differences was examined compared to control samples. The test probability of $p < 0.05$ was assumed to be significant and the test probability of $p < 0.01$ was considered highly significant.

3. Results

Selected results of chemical tests (heavy metals) for biochar obtained from *S. hermaphrodita* and their comparison with the limit values permissible in Poland [4], which determine the maximum permissible amount of contaminants in fertilizers and soil conditioners, are presented in Table 1.

Table 2 also presents a comparison of the results for biochar obtained from *S. hermaphrodita* with other binding standards for this type of products in some European

Table 1
Results of chemical determinations (heavy metals) in the biochar obtained from *Sida hermaphrodita*

Heavy metal	Content of contaminants determined in biochar (mg/kg d.m.)	Maximum permissible content of contaminants (mg/kg d.m.) [4]
Chromium (Cr)	2.12 ± 0.20	100
Cadmium (Cd)	1.3 ± 0.33	5
Nickel (Ni)	1.7 ± 0.64	60
Lead (Pb)	5.2 ± 0.87	140
Mercury (Hg)	0.0039 ± 0.0010	2

±: standard deviation.

Table 2
Comparison of the results of chemical analyses of the tested biochar from *Sida hermaphrodita* with the guidelines for biochar in selected European countries and the USA

Type of contaminant present in biochar (mg/kg d.m.)	Content of contaminants determined in biochar	Permissible values according to the European Biochar Certificate – EBC	Permissible values according to International Biochar Initiative – IBI [12]
Arsenic (As)	<2.0	13	13–100
Cadmium (Cd)	1.3 ± 0.93	15	1.4–39
Chromium (Cr)	2.12 ± 0.73	90	93–1.200
Copper (Cu)	19.0 ± 7.33	1.000	143–6.000
Mercury (Hg)	0.0039 ± 0.0010	1	1–17
Nickel (Ni)	1.7 ± 0.5	50	47–420
Lead (Pb)	5.2 ± 0.9	150	121–300
Zinc (Zn)	51.0 ± 6.3	400	416–7.400
Selenium (Se)	n.m.	–	2–200
Molybdenum (Mo)	4.3 ± 3.8	–	5–75
Fluorine (F)	2.69 ± 0.7	–	–
PAHs (16)	1.395	12	6–300
PCB	n.m.	0.2	0.2–1
Dioxins and furans (ng/kg d.m.)	n.m.	20	20

n.m.: not marked; d.m.: dry mass, ±: standard deviation.

countries (European Biochar Foundation – EBF) and (International Biochar Initiative – IBI) in the United States [12]. The content of PAHs not included in Polish standards was also taken into account.

Other determinations in the examined biochar were as follows:

- pH (in H_2O): 4.80,
- pH (in KCl): 4.53,
- total carbon (TC): 65.5%,
- nitrogen content (N): 0.7%,
- phosphorus content (P): 0.2%,
- potassium content (K): 0.4%,
- TOC (in water extract): 3,620 mg/L,
- calcium content (Ca): 1.4%.

Comparative analysis of the contents of heavy metals in the tested fertilizers with standards for fertilizers and conditioners found no contamination of the tested biochar with these elements. Other determinations and their comparison with the guidelines for biochar used in the European Union and the USA indicated no excessive contaminants such as PAHs. Based on the tests, no other permissible levels of contaminants were exceeded in the biochar.

Results of toxicological tests showed that the biochar produced at 350°C is characterized by phytotoxic properties. Test plants in its water extract (both cress and mustard) did not germinate and did not grow (Fig. 1). Therefore, this biochar was excluded from further research. A lot of tarry compounds migrating to water extract were observed.

The study showed that the biochar produced from the biomass of *S. hermaphrodita* during the pyrolysis process at 450°C did not exhibit phytotoxic properties with respect to *L. sativum* after 2 and 4 d after the experiment was started (Fig. 2).

The results presented in Tables 3 and 4 concern the effect of only chemical compounds contained in biochar formed at 450°C and soluble in water on germination of seeds of *L. sativum* and *S. alba*.

The results of physical and chemical tests of sandy soil used in the pot experiment are presented in Table 5.

Fig. 1. Lack of germination of plants 4 d after the experiment was set up: germination test on a water extract from biochar formed at 350°C.

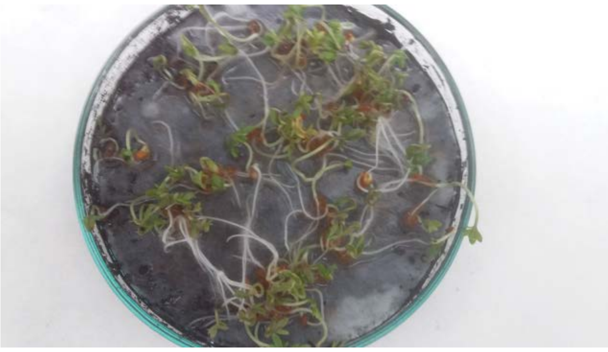

Fig. 2. Germination of garden cress (*Lepidium sativum*) seeds 4 d after the experiment was set up: germination test on a water extract from biochar formed at 450°C.

The results of the second toxicological test on soil mixtures fertilised with biochar are presented in Fig. 3 and Table 6. The number of germinating seeds, biomass, and the growth index of garden cress (*L. sativum*) after 4 d from starting the experiment were determined.

The results concerning the amount of the biomass of the plants studied in the pot experiment are presented for individual fertilization combinations in Table 7.

Photographic documentation of an example test object is shown in Fig. 4.

4. Discussion

The analysis of the content of heavy metals in the examined biochar and comparison with the limit values permissible in Poland indicates the lack of contamination with these elements and their usefulness for soil fertilization (Table 1). The results indicate that the material tested can be characterized as biochar because the content of total carbon exceeds 50% (65.5%). The content of calcium (1.4%) is important in this material. However, the very low pH 4.8 (in H_2O) is worrying. This may indicate an incomplete decomposition of organic matter in the pyrolysis process and the presence of undesirable components. During the examinations, a brown colouring of water extracts obtained from biochar was also observed, which may indicate the presence of tarry compounds. This is also evidenced by the high content of total organic carbon (TOC) in the biochar tested, amounting to 3,620 mg/L. It is likely that during the torrefaction process, the biomass was not fully decomposed. These conditions may lead to an increased content of PAHs.

The tests of the presence of PAHs in biochar did not show an increased concentration of these compounds (Table 2).

Table 3
Effect of biochar on germination of *Lepidium sativum* and *Sinapis alba* seeds based on the phytotest performed in water solution

Growth medium type	Mean amount of germinated seeds (%)			
	Garden cress (*Lepidium sativum*)		White mustard (*Sinapis alba*)	
	After 2 d of the experiment	After 4 d of the experiment	After 2 d of the experiment	After 4 d of the experiment
Control (tissue paper)	93	94	95	94
Biochar (water extract)	89*	91*	91*	90*

Significance of differences: *$p < 0.05$, **$p < 0.01$, s.i.: statistically insignificant difference.

Table 4
Effect of biochar on root formation and root length in *Lepidium sativum* and *Sinapis alba* seedlings based on the phytostest performed in water solution

Growth medium type	Mean root length in germinated seeds (mm)			
	Garden cress (*Lepidium sativum*)		White mustard (*Sinapis alba*)	
	After 2 d of the experiment	After 4 d of the experiment	After 2 d of the experiment	After 4 d of the experiment
Control (tissue paper)	6.2	18.2	8.3	21.2
Biochar (water extract)	5.6 s.i.	16.6*	6.0*	19.4*

Significance of differences: *$p < 0.05$, **$p < 0.01$, s.i.: statistically insignificant difference.

Table 5
Physicochemical characteristics of the sandy soil used in the pot experiment variant

Determination	Physicochemical parameters of the sandy soil
pH (in H_2O)	5.94 ± 0.68
pH (in KCl)	5.48 ± 0.37
Hydrolytic acidity (H_h) (me/100g)	3.0 ± 1.3
P_2O_5 (mg/100g soil)	17.6 ± 5.9
Kjeldahl N (mg/kg)	2,363 ± 454
TC (mg/g)	39.0 ± 7.9
Organic matter content (loss on ignition) (%)	5.8 ± 0.77
Lead (Pb) (mg/kg d.m.)	30.2 ± 6.93
Zinc (Zn) (mg/kg d.m.)	76.5 ± 7.92
Copper (Cu) (mg/kg d.m.)	6.3 ± 1.3
Nickel (Ni) (mg/kg d.m.)	3.6 ± 2.9
Cadmium (Cd) (mg/kg d.m.)	0.3 ± 0.1
Chromium (Cr) (mg/kg d.m.)	7.3 ± 2.6
Mercury (Hg) (mg/kg d.m.)	n.d.

n.d.: not detected, ±: standard deviation

The total content of 16 PAHs in the tested biochar was 1.395 mg/kg d m. Table 2 also presents a comparison of the results of the examinations of biochar obtained from *S. hermaphrodita* with other binding standards for this type of products in some European countries (European Biochar Foundation – EBF) and (International Biochar Initiative – IBI) in the United States [12]. Also in this case, the results of biochar tests did not exceed the permissible limits.

However, the results of toxicological tests showed that the biochar produced at 350°C is characterized by phytotoxic properties (Fig. 1). Test plants (both cress and mustard) did not germinate and did not grow. A large number of tarry compounds was observed, which may indicate that the pyrolysis temperature was too low. This phenomenon has been also emphasized by other researchers [13]. Toxic compounds contained in this material migrated to water extracts and in 100% inhibited the process of seed germination. The material was excluded from further testing because of its phytotoxicity.

The study showed that the biochar produced from the biomass of *S. hermaphrodita* during the pyrolysis process at 450°C did not exhibit phytotoxic properties with respect to test plants, including *L. sativum* and *S. alba* for 2 and 4 d after the experiment was started (Tables 3 and 4, Fig. 3).

The analysis of physical and chemical properties used to fertilize sandy soil (Table 5) showed that it can be classified as clean soils, not contaminated with heavy metals. However, it was soil with a weak acidic reaction according to the reaction classification [9]. It did not exhibit phytotoxic properties.

The results of toxicological tests consisting in introducing selected doses of biochar (0.5%, 1%, and 2%) into sandy soil and evaluation of their potential toxic effect on test plants (Table 6) showed that after the application of this product in all fertilizing doses, there was a slight decrease in all determined parameters (germination and seedling growth) in relation to control objects. However, in the case of 0.5% and 1% doses, the decrease in the number of germinating seeds of *L. sativum* was not statistically significant (very low doses). Only soil fertilization with 2% dose caused a statistically significant decrease in seed germination capacity by 12% on average compared to objects from the control soil. Similar results were obtained in the case of shoot weight and determination of the growth index. However, the plants did not show any physiological reactions indicating the toxic effect of biochar tested (Fig. 3).

Analysis of the results of the examinations of the amount of biomass grown during the pot experiment revealed pronounced differences in the reaction of selected test plants to fertilization of soils with biochar (Table 7). Similar conclusions were drawn based on experiments by other researchers [1]. The best yielding was found for all fertilized mixtures of *S. hermaphrodita*. The increase in the amount of biomass obtained compared to control objects after the application of biochar was 7.5%, 51%, and 24% for doses of 0.5%, 1%, and 2%, respectively. In the case of doses of 1% and 2%, the increase in this amount was noticeable and statistically significant. Other authors cultivating *S. hermaphrodita* [8] confirmed its good response to various types of fertilization. Sunflower fertilized with biochar also yielded more biomass. However, the increase was less

Fig. 3. Germination of garden cress (*Lepidium sativum*) seeds 4 d after the experiment was set up: germination test on mixtures of sandy soil with biochar formed at 450°C (from the left: control and fertilization doses of 0.5%, 1%, and 2%).

Fig. 4. Examples of research objects with the cultivation of *Sida hermaphrodita* in sandy soil mixtures (from the left: control soil with 0.5%, 1%, and 2% of biochar doses, respectively).

pronounced and amounted to 3%, 7%, and 7% after application of 0.5%, 1%, and 2% biochar, respectively. The fertilization of *Z. mays* with biochar resulted in a statistically insignificant decrease in yielding compared to control soils. The best biochar dose for this plant was 0.5%. White mustard exhibited the poorest response to biochar fertilization (yielding decreased statistically significantly after the application of the doses of 1% and 2%). Gładki [1] observed higher values of biomass and the better response of plants to soil amendment with biochar. The growth and yielding of plants depend on many factors. Soil reaction is also very important. The biochar studied was characterized by a low pH of 4.8. Different results were obtained by Soudek et al. [14]. The authors observed alkaline biochar reaction in all experiments. The use of biochar in their research has always reduced soil acidification. However, despite the low pH in the present study, biochar did not cause a decrease in pH after introduction into sandy soil. The reaction in the control soil after the experiment was 6.22, whereas after fertilization with biochar with contents of 0.5%, 1%, and 2%, respectively, this was 6.34, 6.45, and 6.31. This phenomenon can be explained by the high calcium content in biochar (1.4%).

The Soudek et al. [14] observed differences in the germination of selected plants (sorghum) depending on the material used for the production of biochar. Mumme et al. [15] also observed the effect of biochar addition on the reduction of soil toxicity. The same researchers also observed an increase in plant yield (watercress) after fertilization with biochar with compost and a decrease in the yield after the application of biochar with sewage sludge. The authors attributed the factors responsible for phytotoxicity to pH and organic pollutants.

It was found that fertilization of soils with 0.5% and 1% of biochar from *S. hermaphrodita* resulted in the best plant

Table 6
Results of the evaluation of the mean number of germinating seeds, biomass, and the growth index of garden cress (*Lepidium sativum*) on sandy soil fertilized with selected biochar doses after 4 d from starting the experiment

Test soil type	Type of determination		
	Mean amount of germinating seeds (%)	Mean weight of fresh shoots (g)	Growth index
Control (sandy soil)	95	4.11	95.0
Soil + biochar 0.5%	92 s.i.	3.81 s.i.	88.1 s.i.
Soil + biochar 1%	92 s.i.	3.78 s.i.	87.4 s.i.
Soil + biochar 2%	90*	3.63*	83.9*

Significance of differences: *$p < 0.05$, **$p < 0.01$, s.i.: statistically insignificant difference.

Table 7
Amount of green biomass of selected plants on soil mixtures with the addition of biochar

Type of plant grown	Mean weight of biomass grown on individual soil mixtures (g/pot)			
	Control (sandy soil)	Soil + biochar 0.5%	Soil + biochar 1%	Soil + biochar 2%
Virginia fanpetals (*Sida hermaphrodita*)	16.03	17.23 s.i.	24.19**	20.58**
Maize (*Zea mays*)	12.79	12.60 s.i.	11.40 s.i.	11.23 s.i.
White mustard (*Sinapis alba*)	16.90	14.80 s.i.	9.11**	8.89**
Common sunflower (*Helianthus annuus*)	20.38	20.92 s.i.	21.70 s.i.	21.74 s.i.

Significance of differences: *$p < 0.05$, **$p < 0.01$, s.i.: statistically insignificant difference.

growth. In the case of some plants (especially mustard), application of 2% biochar dose leads to a decrease in yield. Different results were obtained by Bouqbis et al. [16]. These researchers added 0%, 0.5%, 1%, 2%, 4%, and 8% biochar from argan shells and studied the effect of the addition on germination of lettuce seeds. Scientists did not find any negative effect of argan biochar on lettuce germination or on germination rate and fresh weight of seedlings. Furthermore, the use of biochar increased the germination rate and the amount of fresh biomass. No toxic or adverse effects of biochar on plant growth were observed.

5. Conclusions

- Biochar produced from *S. hermaphrodita* during the pyrolytic process at 350°C turned out to be phytotoxic for the test plants.
- Toxicological plant tests carried out on water extracts obtained from biochar (formed at 450°C) showed a small effect of this material on inhibition of germination of the test plants (*S. alba* and *L. sativum*). The amount of germinated seeds *S. alba* and *L. sativum* on water extracts of biochar decreased from 94% (on the control material) to 90% and 91%, respectively.
- The biochar studied formed during the biomass pyrolysis process of the biomass of *S. hermaphrodita* introduced into soils slightly inhibited germination of a selected test plant species (*L. sativum*). Optimal doses of biochar (not causing noticeable germination disturbances) are 0.5% and 1%. The growth index of plants on the water extracts from soils after application of these doses was 88.1 and 87.4 (95 in the control soil), respectively.

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