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Phytoremediation of ^{137}Cs contaminated sod-podzolic soil in Northern Polissia white sweet clover (*Melilotus albus*)

Volodymyr Snitynskyi, Serhii Razanov, Petro Hnativ, Oleh Bakhmat, Mykola Kutsenko & Oleh Kolisnyk

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Phytoremediation of ^{137}Cs contaminated sod-podzolic soil in Northern Polissia white sweet clover (*Melilotus albus*)

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ABSTRACT
The accident at the Chernobyl nuclear power plant on 26 April 1986 contaminated tracts of Europe with radionuclides. In Ukraine, two million hectares with radiation levels greater than 5.55×10^{11} Bq/km^2 were removed from agriculture and ^{137}Cs, with a half-life of 30 years, is still with us. Phytoremediation by vegetation that accumulates toxic elements has been widely applied. White sweet clover (*Melilotus albus*) accumulates caesium and heavy metals in its biomass but, at the same time, produces nectar and pollen of a safely low level of ^{137}Cs; so this culture is safe for beekeeping in the Chernobyl contamination zone. Growing *M. albus* over two years (2021–2) on a sandy sod podzolic soil within the Zhytomyr region increased the soil's easily-hydrolysable N by 29.9%, decreased mobile phosphorus by 18.2%, and mobile forms of Cd by 38.5%, Hg by 25%, Pb by 24.5%, Cu by 18.5%, Zn by 14.9%, ^{137}Cs by 8%.

KEYWORDS
Phytoremediation; soil; heavy metals; radioactive caesium

Introduction

The threat of pollution in Ukraine and around the world is unabated – and this includes accidental and deliberate damage to nuclear facilities [1]. In 1986, about 3.5 million hectares of agricultural land was radioactively contaminated by the catastrophe at the Chernobyl nuclear power plant in Ukraine. Two million hectares of arable emitting radiation of more than 5.55×10^{11}Bq/km^2 were removed from production [2,3] but there were no funds for decontamination. Radioactive caesium (^{137}Cs), with a half-life of 30 years, is still present. It moves actively in the soil-plant-consumers system [4,5] and causes thyroid cancer, leukaemia and other disorders [6–8]. There is also more general concern about soil contamination by arsenic, selenium, cadmium, mercury, and lead [9,10].

Phytoremediation – growing plants that accumulate toxins in their biomass – is being applied to hasten the return of contaminated land to agriculture [11,12]. Phytoremediation is safe and cost-effective compared to conventional physical and chemical procedures for mitigating soil and water pollution [13,14]. Amongst most-

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promising species, honey clovers (legumes, also known as sweet clovers) accumulate ^{137}Cs but concentrate little of the pollutant in their nectar and pollen, so they can be employed for apiculture [15,16]. White sweet clover (*Melilotus albus* Medik) is undemanding in terms of climate and grows on droughty and, even, moderately saline soils. The green mass serves as stockfeed; the nectar and pollen are attractive to bees [17]; moreover, it is good green manure, drawing nutrients from the deep subsoil and accumulating nitrogen through its symbiotic nitrogen-fixing bacteria [18,19]. Here, we focus on the changes in the agro-ecological parameters of a radioactively contaminated sod-podzolic sandy soil in the course of phytoremediation using white sweet clover.

Materials and methods

Field study was undertaken on a sandy sod-podzolic soil (*Albic Retisol*, arenic, Aric in the World Reference Base [20]) during 2021–2022 within the Narodichi territorial community of the Zhytomyr region, Northern Polissia, Ukraine (51°12′10″ N, 29°04′ 53″ E). The site had been fallow as a result of pollution. For the cultivation of *M. albus*, we established a control, continuing fallow, and an experimental variant under sweet clover, each with four replicates; each plot 25 m^2.

At the outset, soil samples were selected from every plot by the envelope method (four samples from the corners of a square with sides of 100 m and one from the centre). To ascertain the agroecological consequences of phytoremediation, soil sampling was carried out simultaneously in the autumn from the control and experimental variant after collection and removal of the vegetative mass of the experimental plots. Tillage included disking the fallows and the experimental variant, ploughing to 20–22 cm in the autumn and, in spring, pre-sowing cultivation. The Grozynsky variety of *M. albus*, naturalised in Polissia, was sown in early spring at a rate of 14 kg/ha to a depth of 2–3 cm with a row spacing of 45 cm.

Measurements of humus were made by dichromate oxidation [21], reaction (pH_{KCl}) potentiometrically [22], easily-hydrolysed nitrogen following Cornfield [23], available phosphorus following Bray and Kurtz [24], available potassium by ammonium acetate extraction and flame photometry. Mobile Cu, Zn, Pb, Cd and Hg were determined by atomic absorption spectrometry [25] and ^{137}Cs in plants and soil was determined by IAEA methods [26]. To assess the degree of danger of heavy metals, the hazard ratio was determined according to the formula: Hr = C/MPCi, where C is the heavy metal concentration in the soil (mg/kg) and MPCi is the maximum permissible concentration of heavy metals in soil (mg/kg). Statistical analysis of the reliability of the results was performed using Statistics Kingdom online.

Results and discussion

At the outset, the plough layer contained 1.32 ± 0.7% humus, 117.0 ± 1.5 mg/kg easily-hydrolysed nitrogen, 197.2 ± 1.2 mg/kg available phosphorus and 292.4 ± 2.7 mg/kg of available potassium. Two years' cultivation of *M. albus* with annual removal of above-ground biomass had a measurable effect on agrochemical indicators (Table 1): there was a significant increase in the content of easily-hydrolysable N, a decrease in the content of available phosphorus and potassium, but no significant change in pH or humus content.

Table 1. Changes in agrochemical parameters of the soil during the cultivation of *M. albus*.

Agrochemical indicator	Control (fallow)		Culture *M. albus*	
	First year	Second year	First year	Second year
Humus, %	1.32 ± 0.7	1.32 ± 0.2	1.38 ± 0.4	1.40 ± 0.6
Reaction (pH_{KCl})	6.44 ± 0.4	6.42 ± 0.7	6.46 ± 0.2	6.5 ± 0.2
N (easily-hydrolysable), mg/kg	117.0 ± 1.5	117.8 ± 0.4	136.0 ± 4.1	152.0 ± 6.2^{xxx}
P (available), mg/kg	197.2 ± 1.2	198.1 ± 1.6	178.6 ± 1.2	161.3 ± 3.1^{xxx}
K (available), mg/kg	292.4 ± 2.7	293.7 ± 1.4	267.4 ± 3.7	251.6 ± 4.7^{xxx}

$^{xxx}P < 0.001$.

Soil enrichment with available N by fixation of atmospheric N is a clear benefit. Over two years under *M. albus*, easily-hydrolysable N increased by 29.9%, together with some neutralisation of acidity and decrease of available phosphorus and potassium by 13.9% and 18.2%, respectively. Cultivation of *M. albus* also brought about significant changes in the content of mobile heavy metals in the soil (Table 2). Over the two years of cultivation of *M. albus*, the amount of Cu, Zn, Pb, Cd, and Hg decreased by 18.7%, 14.9%, 24.5%, 38.5% and 25%, respectively.

Even one year's cultivation of *M. albus* extracted 22.2% of the mobile cadmium and 21.4% of the lead from the soil (Figure 1). Over two years, the above-ground biomass

Table 2. Changes in the mobile heavy metals in the soil during the cultivation of *M. albus*.

Heavy metal	Content of heavy metals, mg/kg			
	Control		Culture *M. albus*	
	First year	Second year	First year	Second year
Cu	0.32 ± 0.07	0.33 ± 0.09	0.27 ± 0.07	0.26 ± 0.014^{xxx}
Zn	1.07 ± 0.20	1.09 ± 0.02	0.96 ± 0.03	0.91 ± 0.037^{xx}
Pb	3.74 ± 0.60	3.78 ± 0.01	2.94 ± 0.2	2.82 ± 0.031^{xx}
Cd	0.18 ± 0.03	0.18 ± 0.02	0.14 ± 0.02	0.11 ± 0.07^{xx}
Hg	0.0068 ± 0.0005	0.0068 ± 0.0004	0.0060 ± 0.0003	0.0051 ± 0.0002^{xxx}

$^{xx}P < 0.01$; $^{xxx}P < 0.001$.

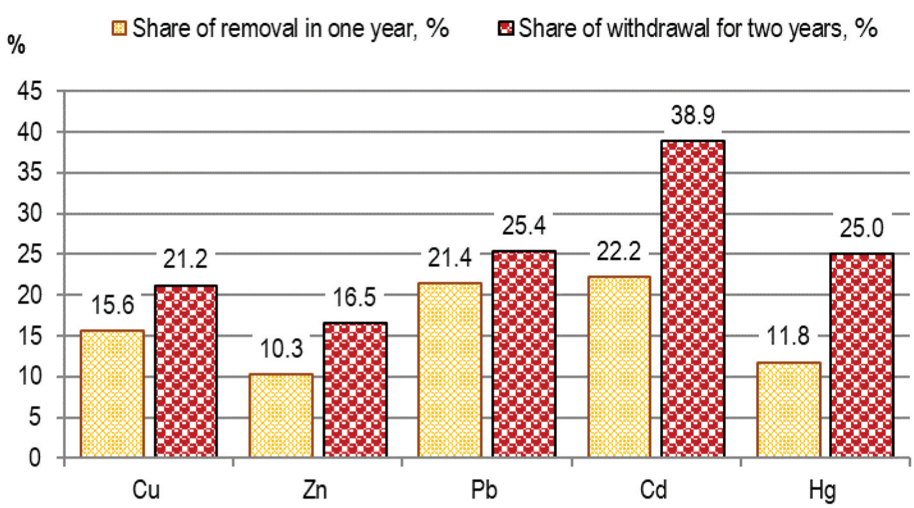

Figure 1. Removal of mobile forms of heavy metals during two years of cultivation of *M. albus*, % of the initial content in the soil.

Table 3. Change in the hazard ratio of heavy metals in the soil during the cultivation of *M. albus.*

		The hazard ratio of heavy metals in the soil			
Heavy metal	MPC	First year	Second year	First year	Second year
Cu	3.0	0.106 ± 0.004	0.11 ± 0.004	0.09 ± 0.0004^{xxx}	0.086 ± 0.002^{xxx}
Zn	23.0	0.046 ± 0.002	0.047 ± 0.0005	0.041 ± 0.0002^{x}	0.039 ± 0.0006^{xxx}
Pb	6.0	0.623 ± 0.002	0.530 ± 0.0001	0.490 ± 0.0008^{xxx}	0.470 ± 0.007^{xxx}
Cd	0.7	0.257 ± 0.004	0.257 ± 0.0009	0.20 ± 0.003	0.157 ± 0.003^{xxx}
Hg	2.1	0.003 ± 0.00003	0.003 ± 0.00004	0.002 ± 0.0007	0.0024 ± 0.0006^{xx}

$^{xx}P < 0.01$; $^{xxx}P < 0.001$.

assimilated 38.9% and 25.4% of these elements, respectively, and there was also significant removal of Hg, Cu and Zn.

The hazard ratio of toxicants (Table 3) indicates the level of soil contamination and its suitability for agricultural use.

In all cases, the ratio was less than the critical value of unity and, after the first year of cultivation of *M. albus*, the hazard ratio of heavy metals in the soil decreased by 15.0% for Cu, 10.8% for Zn, 21.3% for Pb, 22.1% for Cd and 6.6% for Hg. After the second year of phytomelioration, the soil hazard ratio of Cu, Zn, Pb, and Hg decreased by a further 4.4, 4.8, 4.0, 21.5 and 14.2%, respectively. So, over two years of remediation under *M. albus*, the hazard ratio of Cu, Zn, Pb, Cd and Hg in the soil decreased by 18.8, 15.2, 24.5, 38.9 and 20%, respectively.

Figure 2 depicts the changes in the ^{137}Cs radioactivity in the soil. Thus, in the test soil before growing *M. albus*, the radiometric reading was 246.5 Bq/kg. After growing this crop for two years and removing the above-ground biomass, the radioactivity of the soil decreased by 8%.

We find that phytoremediation of sandy soil by growing *M. albus* has a complex effect on its ecological condition; notably, an increase in the content of easily-hydrolysable N and a decrease in the concentration of toxins, including radioactive caesium. This is

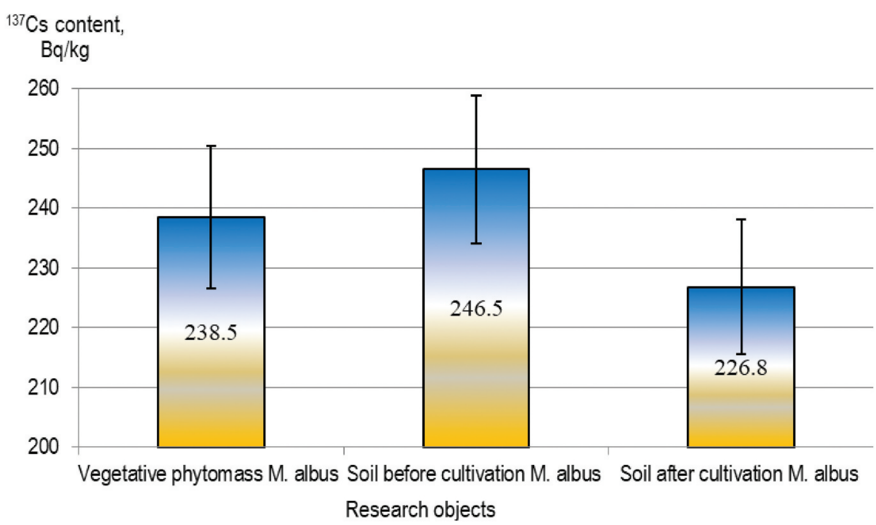

Figure 2. Changes of ^{137}Cs radiation in the soil during the two-year cultivation of *M. albus*, Bq/kg.

highly desirable to increase the ecological safety of soils and return them to agricultural use in the contamination zone of the Chernobyl nuclear power plant.

Moreover, there is consensus that phytoremediation can be effective for the removal of a variety of soil contaminants, including petroleum hydrocarbons, munitions waste (*e.g.*, TNT), metals and metalloids, salt, and radioisotopes [27]: Brown *et al.* [28] recommend metal-tolerant hyperaccumulator plants for phytoremediation of contaminated soils; Hamzah *et al.* [29] report a 71% decrease in the concentration of Cd by vetiver (*Chrysopogon zizanioides* L. Roberty), 59% by Indian goosegrass (*Eleusine indica* L.), 52% by tropical whiteweed (*Ageratum conyzoides* L.) and hairy spurge (*Euphorbia hirta* L.), and 22% by Siam weed (*Chromolaena odorata* L.); tropical black nightshade (*Solanum nigrum* L.) has also been widely studied for the remediation of soils contaminated by heavy metals [30].

Kocira *et al.* [31] focus on Fabaceae for their versatile benefits: reducing soil compaction and erosion, improving soil structure, increasing soil organic matter and the activity of microorganisms and, especially, increasing the nitrogen content by symbiotic fixation of atmospheric nitrogen. Ali *et al.* [32] reported phytoremediation of Cd, Pb, Cu and Zn contamination using annual berseem clover (*Trifolium alexandrinum* L.); the values of the bioconcentration coefficient of the roots for Zn, Pb, Cu and Cd were 4.24, 1.54, 1.07 and 0.60, respectively [32]. Tlustoš *et al.* [33] investigated As, Cd, Pb and Zn uptake and potential phytoremediation efficiency of five plants commonly used as forage and energy crops: *Melilotus alba*, red clover (*Trifolium pratense* L.), Chinese mallow (*Malva verticillata* L.), safflower (*Carthamus tinctorius* L.) and hemp (*Cannabis sativa* L.); the total absorption of elements decreased in the order *C. tinctorius* > *M. verticillata* > *C. sativa* and *M. alba* [33].

Our own field study underscores the phytoremediation potential of the *Fabaceae*, in particular *M. albus*, in polluted landscapes. It enriched the topsoil of sandy *Albic Retisol* by 18.2 mg/kg of easily hydrolysable N in the first year of cultivation, and by another 16 mg/kg in the second – but the main benefit is that, by removing the above-ground biomass, the soil contaminated by the Chernobyl accident was *gradually but noticeably cleaned* of ^{137}Cs, Pb, Cd, Zn, Cu and Hg.

Conclusions

- Phytoremediation of radioactively contaminated agricultural land with the help of a two-year culture of white sweet clover *Melilotus albus* and the removal of above-ground biomass increased the content of easily hydrolysed nitrogen by 29.9% but decreased mobile phosphorus by 18.2% and exchangeable potassium by 13.9%.
- The two-year cultivation of *M. albus* also reduced the content of radiocaesium by 8% and the concentration of heavy metals: Cd by 38.5%, Hg by 25%, Pb by 24.5%, Cu by 18.7%, and Zn by 14.9%, significantly reducing the hazard ratio of these toxins.

Disclosure statement

No potential conflict of interest was reported by the author(s).

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