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Composition for Targeted Plant Root Treatment in Drylands: Justification of Components and Concentrations for Field Tests

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Abstract

The study deals with the problem of combating the growth of undesirable vegetation on the borders of agroecosystems and in unrecoverable areas (intrusions) within these systems. The immediate objective of the study was to reduce the growth of dominant and subdominant species in arid plant communities on the example of *Artemisia lerchiana*.

To solve this problem, we used an approach based on the impact on the irremediable or difficultly eliminated herbaceous areas using plant root treatment in the early vegetative phase. To do this, we developed an aqueous suspension of the original variable composition, which provided delivery to the plant roots, based on 1 kg of surface soil layer: 20-40 mg of copper ions and/or 1-2 mg of zinc ions (1); 8-20 mg of citric and/or succinic acid, permissible in the form of sodium or potassium salts (2); and 10-20 mg of chitosan (3).

As a result, a complex biogenic effect of the received suspension on the soil microbiota and plants was achieved. The improved inhibiting the plant growth and reducing the projective coverage (only for zinc ions or combination copper with chitosan in high concentrations), are the experimental basis for subsequent field tests. For this study we developed a working algorithm for the selection of components and its concentration, based on mathematical modeling of different responses of plant communities to variations in the composition of the suspension for plant root treatment.

Keywords: arid plant communities, undesirable vegetation, plant traits, copper, zinc, chitosan, plant biotechnology, plant root treatment, *Artemisia lerchiana*.

1. Introduction

The approaches to observation and control of ruderal vegetation using mechanical methods, if necessary, in combination with various herbicides and their compositions are well-known from the literature (Monaco et al., 2008; Kraehmer et al. 2014). These works present in detail some technical devices for mechanical soil treatment (Tu et al., 2001; Monaco et al., 2008) and technologies of their application depending on the type of soil, main crop, season, nature and degree of contamination of the agroecosystem and surrounding area. We may find a lot of developed algorithms for calculating the concentrations of active substances in herbicide compositions depending on the dominant types of undesirable vegetation (Tu et al., 2001; Ani et al., 2018), protocols of their application, including the use of the sprayer shape, the angles of solution direction to the plants, and the speed of movement along the processing area (Zimdahl, 2017).

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Thus, although research and innovation activity in this direction does not weaken due to the emergence of new agricultural technologies and the presence of plant invasions, the fundamental approaches to this issue can be considered well-established.

At the same time, there are often situations that represent unrecoverable areas of undesirable vegetation penetrating the territory of managed agroecosystems, the so-called technogenic intrusions. As a usual characteristic, it is shown for intrusions to have reduced species composition, direct or indirect negative impact on the adjoining agroecosystem contributing the violation of consort links in it.

It was shown that agroecosystem demonstrates the decrease in the projective coverage, biological productivity of arable crops in the contact zone with the intrusion. These plots form a special microrelief, changing moisture conditions and transfer of pollutants with subsequent accumulation in the system 'soil – plant' (Ivantsova et al., 2017; Ivantsova, Novochadov, 2019).

Such areas in the area of power lines, gas and oil pipelines, etc. can invade significantly deep into the territory of agroecosystem, and the use of classical methods of combating undesirable vegetation here is limited both by agricultural protocols, and ecology.

There is, therefore, an objective need to find and develop new methods of combating undesirable vegetation on the territory of technogenic intrusions, on one hand providing for the preservation of a certain part of this ecosystem (ecological aspect), on the other hand being strongly aimed to reduce the negative impact of vegetation on the adjoining agroecosystem (production aspect). For obvious reasons, these technical solutions must be adapted to modern agricultural technologies, labor-intensive, and economically feasible.

In this area, we know a way to combat weeds by forming a seed mixture of different plant types, so that the new herbaceous cover completely suppressed the growth of weeds in the surrounding area. The combined crop mix helped to increase the full employment of ecological niches and, consequently, reduced the diversity and density of weed plants (Hossain et al., 2012; El-Sayed et al., 2014). The disadvantage of this method is that it is not quite suitable for solving the problem of man-made intrusions. In addition, the soil needs to be pre-machined, and use of this method is almost impossible in intrusion territory.

Velisevich et al. (2010) described a method of protection against undesirable vegetation, including treatment with herbicides by contact spraying, followed by removal of the plant mass by mechanical means. The main drawback of this approach is the same, because we have no possibility to carry out mechanical tillage in areas bordering the agroecosystem. In addition, the herbicide application may be limited by the main production process.

Currently, there is information about the use of fertilizers based on polymer matrices containing polycarboxylic acids or polysaccharides (Danilova, 2016; Sarkar et al., 2018). Their use is an effective way to retain the liquid and redistribute of trace elements or microorganisms both on the plants and in the soil. Such polymers can contribute to changing the species composition of intrusions and become a component of technologies to combat undesirable vegetation.

Based on the above, the immediate task of the study was to overcome the negative impact of irremediable or difficultly eliminated grass vegetations on the adjoining agroecosystems. In order to solve this problem, we used an approach based on the impact on weeds in these areas by means of plant root treatment in the early vegetative phase.

2. Material and methods

To select the components of the suspension for root treatment, we used a forecast based on a mathematical model that was compiled earlier and tested on the results of our own observations (Ivantsova et al., 2018).

As follows from these studies, control effects should not actively affect the agroecosystem (1); they should be achieved by introducing an additional component, not by reducing the existing one (2); they should not have herbicidal effect (3). The formula of the introduced compound should not contain more than three components, since in this case the costs increase significantly, and it is impossible to reliably predict the result (4). We refused to introduce microorganisms due to the difficulties of dosage and increasing labor intensity at all stages of the technology.

As a result, the main components in the suspension formula for plant root treatment, which allows for minimal impact on the plant communities of intrusion to sufficiently reduce their negative impact on the adjoining agroecosystem, were a trace element (copper or zinc), a metabolite stimulating the soil microbiota (citric or succinic acid), and a polymer for moisture protection and activation of micromycetes (chitosan).

Table 1 illustrates the preliminary justification for the required amounts of individual substances to be applied to the soil.

Table 1. Calculation of the required number of suspension components for plant root treatment (all values are expressed in mg/kg of dry soil)

Suspension component	Range of values in soil intrusion	Decision rule based on a mathematical model (Ivantsova et al., 2018)	Interval of checked values for root treatment
Chitosan	–	Ensure the presence of at least 100 mg/kg in the surface layer of the soil (up to 5 mm). Dose of chitosan should increase as the metal dose increases.	5.0 – 40.0
Copper	12.0 – 45.0	The increase of concentration by 30-100 % is effective. The effect is less, the greater metal concentration in soil	10.0 – 80.0
Zinc	2.0 – 7.0		0.5 – 4.0
Succinic acid	Not defined	Application of up to 2.5 mg/kg for every 10 % of the expected projective coverage is effective	8.0 – 20.0
Citric acid			

As a result, twenty working formulas of suspension were prepared for the test. In the first group, the chitosan was introduced into the soil at doses from 0.05 to 4.0 mg/kg together with citric acid. The suspensions in the second group were a combination of citric acid and copper in concentrations that ensure the introduction of a trace element into the soil from 10.0 to 80.0 mg/kg. The third group tested a similar composition, varying in zinc in amounts from 0.5 to 4.0 mg/kg of soil. The fourth group was represented by suspensions including copper, citric acid and chitosan, the fifth group varied in zinc content, respectively.

Table 2 details the compositions of individual suspensions, which will be referred to in the text in the future. For example, subgroup IV-A denotes a suspension that provides 10.0 mg/kg of copper, 10.0 mg/kg of citric acid, and 0.05 mg/kg of chitosan to the soil.

Table 2. Calculated increments of substance concentrations (mg/kg of dry soil) after plant root treatment using test suspensions

Group*	Subgroup			
	A	B	C	D
I	Chitosan 0.05	Chitosan 0.1	Chitosan 0.2	Chitosan 0.4
II	Cu 10.0	Cu 20.0	Cu 40.0	Cu 80.0
III	Zn 0.5	Zn 1.0	Zn 2.0	Zn 4.0
IV	Cu 10.0 + Chitosan 0.05	Cu 20.0 + Chitosan 0.1	Cu 40.0 + Chitosan 0.2	Cu 80.0 + Chitosan 0.4
V	Zn 0.5 + Chitosan 0.05	Zn 1.0 + Chitosan 0.1	Zn 2.0 + Chitosan 0.2	Zn 4.0 + Chitosan 0.4

* – in all cases, citric acid is present at a concentration that provides 10 mg/kg of this substance to the soil.

As the object for testing, we chose the plant *Artemisia lerchiana* (Web.), as well-studied, having the necessary cultural properties and found us earlier as a dominant or subdominant in more than 70 % of the intrusions in arid zone.

To obtain plants in the early vegetative phase, we placed ehe native soil from intrusions in the pots, two plants were placed by root planting in each one, cut off to 3-4 root buds. After 2 weeks, the root treatment was performed, randomly forming 2 cases (four plants) in each subgroup. To form the control group, we exclude root treatment in two cases. Considering the number of plants and the number of buds that produced shoots, the average number of measured objects in each sample was from 10 to 15, which was enough for their representativeness.

Inspection and photo documentation of plants we performed immediately before the plant root treatment and 30 days after it (Figure 1).

The main indicators characterizing the effect of substances received in the soil were the average increase in the length of new shoots (mm) and the average projective coverage (%) in each subgroup.

A

B

C

Fig. 1. Digitized images of growing *Artemisia lerchiana* for morphometry. A. Side view for measuring the shoot height. B. Top view for measuring projective coverage. C. Binarization of the previous image, cleaning from artifacts and noises, performed by ImageJ, before automatic determination of projective coverage value

The measurements were performed using the capabilities of the free access program ImageJ (NIH, USA), and MS Excel (Microsoft, USA) was used for subsequent processing and visualization of the results. Since the normal distribution hypothesis was rejected, non-parametric criteria were used: the distribution was expressed as the median and the interquartile range (Me [Q1 ÷ Q3]), and the Kruskal-Wallis test was used to compare samples ($p < 0.01$).

3. Results and discussion

Table 3 shows the results of measurements of the average shoot height and the projective coverage after application of suspensions depending on their different composition

In group I, where we included only chitosan and citric acid in the suspension formula, as the concentration of chitosan increased, the negative effect on the growth of shoots increased. The effect was detected starting from a dose of 0.1 mg/kg of soil. At the same time, we did not find any significant effect in relation to the projective coverage.

In group II, we tested the effect of copper in combination with citric acid and obtained similar results. A dose-dependent negative effect on the growth of shoots was detected, starting from the addition of 20.0 mg/kg of copper to the soil. No effect was found with respect to the projective coverage.

Completely different changes we registered due to testing zinc together with citric acid (group III). This suspension significantly slowed down the growth of shoots and caused the formation of foliage with a low projective coverage. The effects of zinc did not depend on the administered dose of the trace element.

In group IV, where we tested the co-action of copper and chitosan, a distinct effect on both analyzed indicators was detected in subgroups IV-C and IV-D, that is, when adding 40.0 mg/kg of trace element and 0.2 mg/kg of chitosan to the soil or more.

Table 3. Changes in growth and development after plant root treatment of *Artemisia lerchiana* with suspensions of different composition

Group/Subgroup	Indicators, Me [Q1 ÷ Q3]	
	Average growth of shoot length, mm	Average projective cover, %
Control	1183 [1020 ÷ 1285]	84.2 [75.9 ÷ 89.1]
Chitosan and citric acid		
I-A	1109 [961 ÷ 1243]	82.0 [70.5 ÷ 87.4]
I-B	796 [675 ÷ 910] *	77.3 [66.1 ÷ 83.8]
I-C	644 [558 ÷ 736] *	74.5 [64.8 ÷ 77.3]
I-D	302 [259 ÷ 334] *	77.0 [65.5 ÷ 82.6]
Copper and citric acid		
II-A	1057 [894 ÷ 1218]	83.5 [72.2 ÷ 88.8]
II-B	841 [717 ÷ 965] *	80.7 [69.7 ÷ 85.0]
II-C	696 [601 ÷ 805] *	81.4 [70.0 ÷ 87.4]
II-D	638 [550 ÷ 726] *	76.0 [65.5 ÷ 78.9]
Zinc and citric acid		
III-A	445 [383 ÷ 512] *	67.2 [60.1 ÷ 73.2] *
III-B	497 [429 ÷ 570] *	68.5 [61.3 ÷ 75.0] *
III-C	436 [375 ÷ 501] *	63.0 [57.8 ÷ 70.9] *
III-D	400 [344 ÷ 464] *	62.7 [55.6 ÷ 68.7] *
Copper, chitosan and citric acid		
IV-A	1215 [1033 ÷ 1395]	80.0 [69.1 ÷ 86.6]
IV-B	977 [840 ÷ 1128]	73.5 [62.3 ÷ 79.0]
IV-C	323 [281 ÷ 370] *	70.2 [61.0 ÷ 76.8] *
IV-D	304 [265 ÷ 354] *	67.8 [59.5 ÷ 74.9] *
Zinc, chitosan and citric acid		
V-A	895 [763 ÷ 1028]	65.3 [59.0 ÷ 71.2] *
V-B	502 [430 ÷ 577] *	61.8 [56.1 ÷ 68.5] *
V-C	800 [684 ÷ 927] *	56.0 [49.7 ÷ 63.0] *
V-D	711 [615 ÷ 822] *	55.4 [49.4 ÷ 61.9] *

* – significant differences with values in the control group

Group V was represented by samples that received together zinc, chitosan and citric acid. In this case, we identified the maximum negative changes from the growth of shoots and projective coverage. The changes in the growth of the shoot length dominated at low doses of the introduced substances, the effects on the projective coverage dominated at higher ones.

The effect of trace elements on plant communities can only partially be reduced to direct metabolic consequences of their penetration into plant cells, where metals are included in the complex proteins with regulatory, transport, and enzymatic properties (Singh, Parihar, 2015; He et al., 2016; Novochadov et al., 2018). Equally important events occur directly in the soil, where metals, including copper and zinc, can significantly change the growth rate and metabolic activity of microorganisms. The microbiota provides a complex of protective and trophic processes, based on stimulation of oxidation, deposition, complexation, transport and conjugation. As a result, this leads to changes in the growth and development of plants, as well as their sensitivity to environmental influences (Ma et al., 2016; Meena et al., 2017; Kertész et al., 2017; Bargaz et al., 2018).

The root treatment options used by us provided, after natural alignment of the resulting concentration and pH gradients, the content of mobile trace elements exceeding from 30 % to 100 % above the initial concentrations. It is the combined effects of these metals in the suspension on the soil microbiota and plants that explain the results obtained for inhibiting the growth and development of the model plant (*Artemisia lerchiana*).

Regarding chitosan, the use of polymers for soil treatment has been shown to be effective due to the ability to thus provide water retention and trace elements near plant roots (Danilova, 2016; Sarkar et al., 2018). Chitosan after entering the soil becomes insoluble and forms a three-dimensional matrix structure. Perhaps such deposits serve as a matrix for the growth of soil microfungi, which use these polymers as the main source of carbon. The organic acids received as part of the suspension, which exist in these conditions in the form of soluble and bioavailable anions, support all the above processes. This, in our opinion, achieves a complex biogenic effect of the received suspension on the soil microbiota and plants, resulting in an increase in the contribution of microfungi to the transformation of humus, mobilization of soil trace elements, intensification of vegetative processes in plants with subsequent depletion of consort bonds in the system 'soil – plant microbiota'.

The direct composition for plant root treatment can be selected empirically, which requires information about the content of mobile forms of copper and zinc in the soil, the species composition of plant community, its projective coating, and the influence on the adjoining agroecosystem. If we have a low influence on agroecosystem, the use of any actions is not advisable. For direct field testing, we have developed a working algorithm that uses only the soil concentrations of copper and zinc, as well as the influence of the intrusion on the adjoining agroecosystem for the selection of suspension components and their dosage (Figure 2).

It is known that plant communities living in more severe and stressful habitat conditions are less susceptible to invasion by alien species, compared with similar ones in more temperate habitats. This is due to limited reproduction, low rates of population movement, while the plant communities have enough resistance to invasive impacts (Zefferman, 2015; Hudson et al., 2017).

Both restriction of distribution and resistance to invasion can simultaneously contribute to the low aggressiveness of plant communities themselves in harsh habitats in relation to surrounding territories (Liu et al., 2014; Uroy et al., 2019).

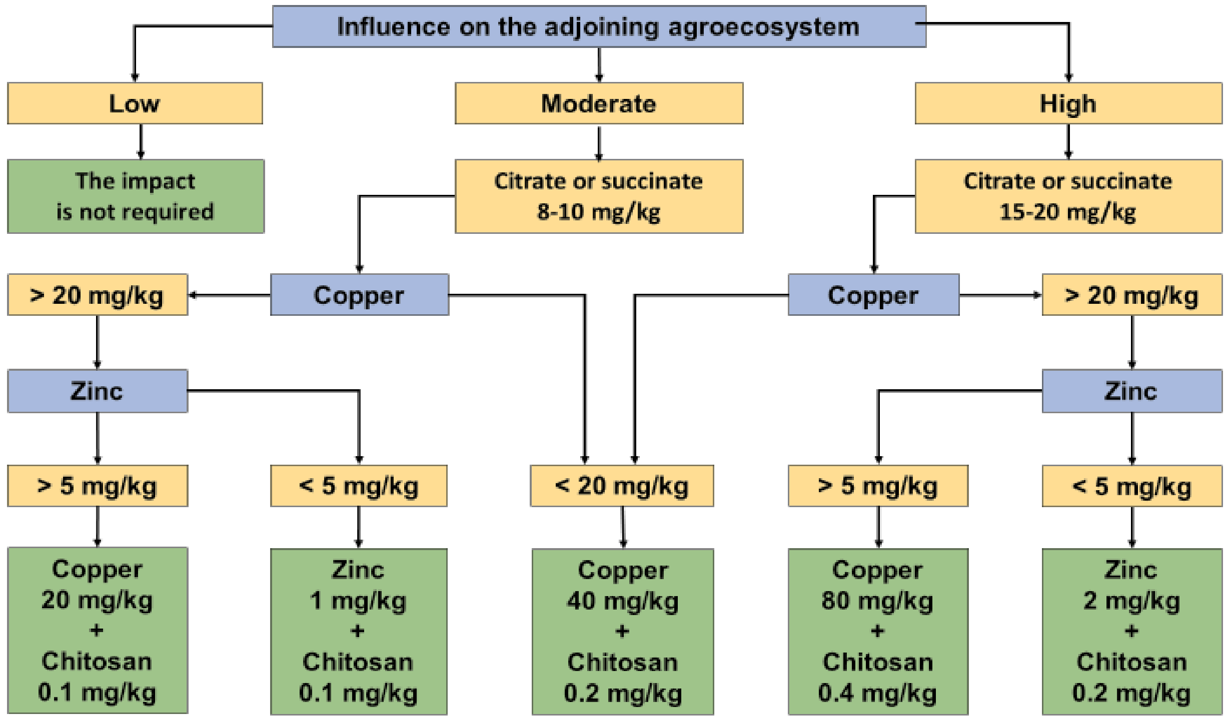

Fig. 2. A working algorithm for field testing the effectiveness of the developed suspensions to reduce the negative impact of undesirable plant communities on the adjoining agroecosystem

We associate the potential effectiveness of the developed suspensions in their application in real land use conditions with the emerging stress conditions in relation to plants that are dominant and subdominant in arid plant communities.

4. Conclusion

This study has shown the effectiveness of new suspensions for plant root treatment as a tool to reduce the growth of dominant and subdominant species in arid plant communities on the example of *Artemisia lerchiana*. To do this, we developed an aqueous suspension of the original variable composition, which provided delivery to the roots of plants, based on 1 kg of surface soil layer: 20-40 mg of copper ions and/or 1-2 mg of zinc ions (1); 8-20 mg of citric and/or succinic acid, permissible in the form of sodium or potassium salts (2); and 10-20 mg of chitosan (3).

As a result, a complex biogenic effect of the received suspension on the soil microbiota and plants was achieved. The improved inhibiting the plant growth and reducing the projective coverage (only for zinc ions or combination copper with chitosan in high concentrations), are the experimental basis for subsequent field tests. For this study we developed a working algorithm for the selection of components and their concentration, based on mathematical modeling of different responses of plant communities to variations in the composition of the suspension for plant root treatment.

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