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The allelopathic activity of water-soluble biologically active substances from *Hyssopus officinalis* L. of Marquis varieties and their effect on the root growth of *Lepidium sativum* L.

Abstract

Aim. To study the effect of secretions of bioactive water-soluble compounds from leaves, stems, flowers, and the root system of *Hyssopus officinalis* L. on the root growth of the test sample (*Lepidium sativum* L.).

Materials and methods. To determine the effect of biologically active water-soluble compounds released by hyssop plants, a series of biotests was conducted; the allelopathic activity of leaves, stems, and flowers of *H. officinalis* L. and its root system were determined. The preparation of extracts of physiologically active substances was carried out according to the method of A. M. Grodzinsky. The allelopathic effect of *H. officinalis* L. was assessed by the effect of the water extracts of various concentrations (100%, 50% and 10%) on the root length of seedlings of *L. sativum* L. test objects. The inhibition index was determined by Williamson method.

Results and discussion. According to the results of the research, it was found that the allelopathic activity of water-soluble compounds of common hyssop affected the germination and length of seed roots of *L. sativum* L. as it changed with the age of the plant: in the first year of vegetation, the stimulating effect of aqueous solutions of hyssop on seed germination was observed, in the second year, the stimulating effect decreased, and in the third year of vegetation, the inhibitory effect on seed germination of the test object was observed. Thus, the index of the allelopathic activity also changed.

Conclusions. It was found that the highest allelopathic activity of biologically active water-soluble compounds of root secretions, leaves, stems and flowers of *H. officinalis* L. was during the flowering phase in concentrations of 100%, 50% and 10%, affecting the length of the roots of seedlings of *L. sativum* L. test objects. At the same time, the maximum stimulating effect on the length of the roots of the test object was in the variant using an aqueous extract of plant flowers of the second year of vegetation in a 10% concentration. The use of aqueous solutions of plants of the third year of vegetation had an inhibitory effect on the root length of *L. sativum* L. seedlings.

Keywords: *Hyssopus officinalis* L.; *Lepidium sativum* L.; allelopathic activity; water extracts; biologically active water-soluble substances

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Алелопатична активність водорозчинних біологічно активних речовин *Hyssopus officinalis* L. сорту Маркіз та їх вплив на ріст коренів *Lepidium sativum* L.

Мета. Дослідити вплив виділень біологічно активних водорозчинних речовин листків, стебел, квіток і кореневої системи *Hyssopus officinalis* L. на ріст коренів тест-об'єкта (*Lepidium sativum* L.).

Матеріали та методи. Для виявлення впливу біологічно активних водорозчинних речовин, виділюваних рослинами гісопу, було проведено серію біотестів, за якими визначали алелопатичну активність листків, стебел, квіток та кореневої системи *H. officinalis* L. Приготування екстрактів фізіологічно активних речовин проводили за методикою А. М. Гродзінського. Алелопатичний вплив *H. officinalis* L. оцінювали за впливом водних екстрактів різних концентрацій 100%, 50% і 10% на довжину корінців проростків тест-об'єктів *L. sativum* L. Індекс інгібування визначали за методикою Вільямсона.

Результати та їх обговорення. За результатами досліджень з'ясовано, що алелопатична активність водорозчинних речовин гісопу лікарського на проростання та довжину корінців насіння *L. sativum* L. змінюється з віком рослини: на першому році вегетації спостерігалась стимулювальна дія водних розчинів гісопу на схожість насіння, на другому році стимулювальна дія зменшується, а на третій рік вегетації спостерігається гальмувальна дія на проростання насіння тест-об'єкта. Відповідно змінюється і індекс алелопатичної активності.

Висновки. Виявлено, що найбільша алелопатична активність біологічно активних водорозчинних речовин кореневих виділень, листків, стебел і квіток *H. officinalis* L., яка впливає на довжину корінців проростків тест-об'єктів *L. sativum* L., є за фази цвітіння в концентраціях 100 %, 50 % та 10 %. Водночас максимальний стимулювальний вплив на довжину корінців тест-об'єкта виявлено у варіанті з використанням водного екстракту квіток рослин другого року вегетації за 10 % концентрації. Використання водних розчинів рослин третього року вегетації має інгібувальний вплив на довжину корінців проростків *L. sativum* L.

Ключові слова: *Hyssopus officinalis* L.; *Lepidium sativum* L.; алелопатична активність; водні екстракти; біологічно активні водорозчинні речовини

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■ Introduction

One of the most important factors affecting the production of stable and high-quality agricultural yields is soil fertility, which is directly related to the nature of accumulation and transformation of biologically active substances in the soil. Plant waste products, which are a component of this organic soil complex, affect the soil biota and plant communities in different ways, and they are characterized by a certain allelopathic activity. Allelopathic effects of the soil environment include soil fatigue and phytotoxic plant residues [1–3]. Thus, allelopathy is a property of the living phase of soils that affects the level of fertility.

According to Grümmer [4], the active substances released by the plant in the process of allelopathy (allelochemistry) are divided into:

1. senility-toxic substances, which source is microorganisms and some types of fungi. Senility suppresses the vital activity of higher plants;
2. phytoncides are substances released by higher plants that inhibit the vital activity of microorganisms;
3. colins are chemical products of the vital activity of higher plants that inhibit the vital activity of other higher plants.

Colins were chosen as the area of our research since they are a combination of active substances formed naturally in the environment of phytocenosis.

The amount of biologically active substances released by a plant depends on its type, variety, phase of development, the organ under study, the age of the plant, its physiological state and

soil and climatic conditions of cultivation [5]. The allelopathic effect of one plant on another can have a negative or positive effect, depending on the concentration of secretions [6].

Allelopathic substances play a vital role in regulating the structure of plant communities [7]; they can be used as the raw material for the production of biodegradable herbicides and pesticides, creating mixed plantings, which, according to A. M. Grodzinsky [8], can even stimulate the growth of each other in compacted crops. In addition, the determination of allelopathic properties of agricultural crops prevents soil fatigue that occurs during their permanent cultivation as a result of the one-sided development of certain groups of soil microflora to the detriment of other groups [9].

In this regard, the aim of our study was to elucidate the effect of the allelopathic activity of aqueous extracts of leaves, stems and flowers of *Hyssopus officinalis* L. and soil in the rhizosphere zone for the root growth of *Lepidium sativum* L.

To achieve this goal, the following tasks were set:

- to study the allelopathic activity of above-ground organs of the plant *H. officinalis* L. and soils in the zone of its rhizosphere;
- to compare the allelopathic activity of the water extracts obtained in relation to the test object;
- to determine the allelopathic activity of aqueous extracts of aboveground organs of the plant *H. officinalis* L. and the root system in the zone of its rhizosphere in relation to the test object.

Materials and methods

The research was carried out in the model experiments at the premises of laboratories of the Mykolaiv National Agrarian University during 2019–2021. The plant and soil material for the study was collected during the hyssop flowering phase before the experiment began.

The allelopathic activity in biologically active water-soluble plant secretions of *H. officinalis* L. of Marquis varieties were determined by *Grodzinsky* method [8] (the method of test bioassays). Single-day seedlings of *L. sativum* L. were selected as the test culture because of watercress had a high seed germination rate and it was sensitive to external factors.

To determine the allelopathic activity of hyssop plants, extracts of various concentrations were used from flowers, leaves and stems of the plant in the flowering phase and the root layer of the soil, which was the main place of manifestation of allelopathic relationships [1]. The experiment used water extracts from the aboveground organs of hyssop plants and the soil extract with the concentrations of 1:10, 1:50, 1:100.

The experiment was repeated 3 times. In each repetition, 100 watercress seeds were sprouted in Petri dishes at a temperature of 23°C. Control test objects were sprouted when moistened with distilled water. To determine the effect of water extracts of different concentrations on the test object, the root length of *L. sativum* L. was measured. The increase of the root length was calculated as a percentage of the control using mathematical statistics [10].

The response index (RI) or inhibition index, which characterizes the vector and relative value of the effect of allelopathic compounds on the growth rate of watercress seedlings was determined by *Williamson* as follows:

- if $T > C$, then $RI = 1 - (C/T)$;
- if $T < C$, then $RI = (T/C) - 1$

where T – is the morphometric indicator of the embryo in the experiment (treatment response), and C – is the morphometric indicator of the embryo in the control (control response).

The stimulating effect occurs at a value of $RI > 0$, the inhibitory effect occurs at a value of $RI < 0$ [11].

Results and discussion

The active media of allelopathy are allelochemicals formed mainly in the form of secondary plant metabolites or decomposition products of

microorganisms [12]. Around each plant, within its phytogenic field, an allelopathic sphere is formed; it is associated with the accumulation of physiologically active substances of colins released by plants into the environment. These substances can be both direct metabolites of the plant and its secondary metabolites, which the plant produces during its growth and development, as well as under the influence of environmental conditions [13].

The task of our experiment was to determine the activity of colins in the allelopathic sphere of common hyssop, which makes it possible to draw conclusions about the possibility of soil fatigue after permanent cultivation of this crop for years exceeding the recommended time frame. According to research, the recommended growing time in one field of common hyssop (*H. officinalis* L.) is 5–6 years [14].

The results showed that the allelopathic effect of *H. officinalis* on the germination of watercress seeds varied depending on the age of the plant. Both the stimulating effect of the extracts on the germination of the test object seeds and the inhibitory effect on the germination of watercress seeds were revealed.

During 2019–2021, there was an accumulation of colins in the rhizosphere of common hyssop of Marquis variety with both a stimulating and inhibitory effect on the germination of watercress seeds (Table 1). According to Table 1, it can be seen that in 2019 the allelopathic activity of the soil in the rhizosphere zone of common hyssop in the flowering phase was more active. It was expressed in the stimulating development of the test object and amounted up to 59.82%, while in 2020 it was up to 56.55% in relation to the control. This is also confirmed by the value of the allelopathic activity index (Figure). The Figure shows that the index of the allelopathic activity of the soil, which expresses the inhibitory effect in the rhizosphere zone of hyssop, increases in accordance with the age of plants. It was the largest in 2021 for plants of the third year of vegetation. The growth of the roots of the test object was inhibited by 11.02%, which was confirmed by the allelopathic activity index, which was a negative number (-0.11).

In the natural range of the Mykolaiv region, *H. officinalis* L. was not found, therefore, the plant was introduced. Any deviation from the ecological conditions of the plant origin can cause more intensive accumulation of biologically active compounds in the plant organs [15, 16].

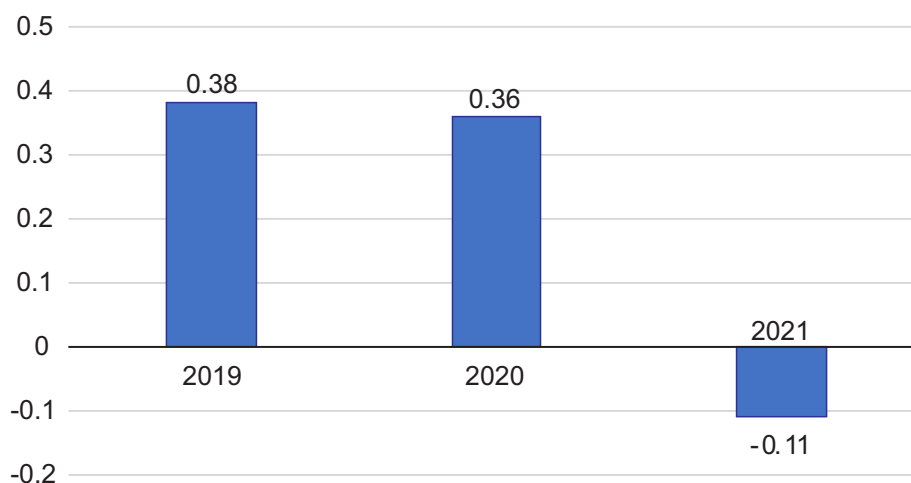


Figure. Changes in the index of the allelopathic activity of the root layer of the soil over the years of research

Table 1. The effect of root secretions of common hyssop plants of different years in the flowering phase on the growth of watercress roots

Vegetation year	Allelopathic activity of the filtrate (dilution 1 : 100)		
	Root length of the test object ($M \pm m$, mm)	Allelopathic activity, %	Allelopathic activity index (RI)
Control	3.36 ± 0.13	–	–
2019	5.37 ± 0.15	+59.82	+0.38
2020	5.26 ± 0.16	+56.55	+0.36
2021	2.99 ± 0.11	-11.02	-0.11

The results of analysis of the root length of the test object and the allelopathic activity of water-soluble biologically active compounds of aboveground organs of *H. officinalis* L. Marquis varieties are listed in Tables 2 and 3.

The use of hyssop flowers in the concentration of 1:50 in the first year of vegetation caused the

growth of the root system of watercress 1.31 times faster compared to the control. Using the (leaf + stem) combination resulted in a slight decrease in the linear growth compared to the flower. All other combinations did not have reliable confirmation.

During the years of research, when studying the allelopathic effect of the age of plants and their aboveground organs on the growth of watercress roots, we found that the highest effect on this indicator was in the variant in which common hyssop flowers were used in the concentration of 1:10 from plants of the second year of vegetation. This concentration contributed to the active growth processes of the watercress root, which was 5.36 mm long and by 2.0 mm longer than the control variant. Decreasing the concentration down to 1:50 and 1:100 reduced this indicator down to 4.56 and 4.73 mm, respectively.

The use of plants in the third year of vegetation led to a sharp decrease in the linear growth

Table 2. The root length of the test object, mm

Plant organ	Year	Solution concentration		
		1 : 100	1 : 50	1 : 10
Leaf	2019	3.64 ± 0.17	3.40 ± 0.17	3.49 ± 0.16
	2020	4.44 ± 0.19	4.54 ± 0.17	4.95 ± 0.14
	2021	2.60 ± 0.07	2.51 ± 0.08	1.02 ± 0.05
Stem	2019	3.84 ± 0.18	3.79 ± 0.15	3.90 ± 0.18
	2020	4.34 ± 0.15	4.83 ± 0.16	4.46 ± 0.15
	2021	2.73 ± 0.09	2.59 ± 0.09	1.46 ± 0.07
Flower	2019	4.04 ± 0.14	4.42 ± 0.16	4.15 ± 0.15
	2020	4.73 ± 0.16	4.56 ± 0.16	5.36 ± 0.15
	2021	2.96 ± 0.11	2.81 ± 0.10	1.85 ± 0.06
Leaf + stem	2019	4.07 ± 0.15	4.37 ± 0.13	3.45 ± 0.13
	2020	4.73 ± 0.18	4.83 ± 0.19	3.80 ± 0.12
	2021	1.39 ± 0.05	1.12 ± 0.05	0.68 ± 0.03
Control	H ₂ O	3.36 ± 0.13	3.36 ± 0.13	3.36 ± 0.13

Table 3. The allelopathic activity, %

Plant organ	Year	Solution concentration		
		1:100	1:50	1:10
Leaf	2019	8.33	1.19	3.87
	2020	32.14	35.12	47.32
	2021	-22.62	-25.30	-69.64
Stem	2019	14.29	12.80	16.07
	2020	29.17	43.85	32.74
	2021	-18.75	-22.92	-56.55
Flower	2019	20.25	31.55	23.51
	2020	40.77	35.71	59.52
	2021	-11.90	-16.37	-44.9
Leaf + stem	2019	21.13	30.06	2.68
	2020	40.77	43.75	13.10
	2021	-58.63	-66.67	-79.76
Control	H₂O	–	–	–

of watercress roots. In addition, aqueous solutions of all variants of concentrations inhibited the growth of the roots of the test object, i.e., an inhibitory effect occurred.

The highest allelopathic activity of 23.51 and 31.55% was demonstrated when using an aqueous solution of the plant flowers of the first year of vegetation in the concentration of 1:10 and 1:50, respectively. Reducing the concentration down to 1:100 stimulated the active linear growth of watercress roots in a lesser extent.

The use of all plant organs in the second year of vegetation had a positive effect, but some of the variants had a lesser effect on the allelopathic activity. Thus, the lowest effect (13.10%) was found in the (leaf + stem) variant in the

concentration of 1:10, and the highest effect (59.52%) of this indicator was when using the flower infusion in the concentration of 1:50.

It is characteristic to note that when using flowers in concentrations of 1:100 and 1:50 the positive effect was observed, but the allelopathic activity of plants of the third year of vegetation was significantly inferior to plants of the first and, especially, the second year of vegetation.

The results of calculating the index of the allelopathic activity of aboveground organs of hysop plants of the Marquis variety are shown in Table 4.

Based on the data given in Table 4, it can be argued that the highest indicator of the allelopathic activity index (RI) was the use of flower

Table 4. The allelopathic activity index (RI)

Plant organ	Year	Solution concentration		
		1:100	1:50	1:10
Leaf	2019	0.08	0.01	0.04
	2020	0.24	0.26	0.32
	2021	-0.23	-0.25	-0.70
Stem	2019	0.12	0.11	0.14
	2020	0.23	0.30	0.25
	2021	-0.19	-0.23	-0.57
Flower	2019	0.17	0.24	0.19
	2020	0.29	0.26	0.37
	2021	-0.12	-0.16	-0.45
Leaf + stem	2019	0.17	0.23	0.03
	2020	0.29	0.30	0.12
	2021	-0.59	-0.67	-0.80
Control	H₂O	–	–	–
Average	2019	0.14	0.15	0.10
	2020	0.26	0.28	0.27
	2021	-0.28	-0.33	-0.63

infusion in the concentration of 1:50 from plants of the second year of vegetation (2020 year) in relation to the control.

As we can see, an average over the years of research, the index of the allelopathic activity of hyssop varied depending on the concentration of an aqueous solution of biologically active substances of aboveground plant organs. Thus, the most stimulating effect of the allelopathic activity index was in the solution with the concentration of 1:50. The maximum inhibitory effect was observed when the solution concentration was 1:10.

■ Conclusions

Thus, all parts of common hyssop plants have an allelopathic effect on the linear growth of the root system of watercress. In the first two years of our research, the stimulating effect of aqueous solutions of root secretions and aboveground organs of hyssop plants was observed regardless

of their concentrations. We found that the variant using hyssop flowers in the concentration of 1:10 from plants of the second year of vegetation had the highest allelopathic activity. Aqueous solutions of root secretions of common hyssop and its aboveground organs of the third year of vegetation showed an inhibitory effect on the linear growth of watercress roots. It was the largest in the (leaf + stem) variant in the aqueous solution with the concentration of 1:10.

Over the years of research, it was also found that on average, the index of the allelopathic activity of water-soluble biologically active substances of aboveground organs of hyssop plants had a stimulating effect in the solution concentration of 1:50, and an inhibitory effect in the solution concentration of 1:10.

Therefore, the additional research is required for determining the soil fatigue in the conditions of permanent cultivation of common hyssop of Marquis variety during the periods exceeding the recommended terms.

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