JMBFS

Journal of Microbiology, Biotechnology and Food Sciences

International peer-reviewed scientific online journal

Published by Faculty of Biotechnology and Food Sciences

ALLELOPATHIC POTENTIAL OF SUGAR SORGHUM (*SORGHUM BICOLOR* (L.) MOENCH) SEEDS

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doi: 10.15414/jmbfs.2019.9.1.93-98

ARTICLE INFO

Received 13. 9. 2018
Revised 19. 3. 2019
Accepted 19. 3. 2019
Published 1. 8. 2019

Regular article

OPEN ACCESS

ABSTRACT

Allelopathic effect of water extracts of crushed sweet sorghum seeds of hybrids 'Medovyi' and 'Dovista' and variety 'Sylosne 42' and their effect on sugar beet (*Beta vulgaris* L.) seeds germination of hybrid 'Ukrainian MS 97' is presented. Water extracts of various concentrations (from 5 to 50 %) were prepared. The results were compared with the control treatment (distilled water). To estimate the growth rate of plants, the number of buds and leaves, plant height and the general condition of plants were examined. In regard to the chemical interaction of crops in mixed cropping, it was found that water extracts from sorghum seeds have a significant amount of allelopathically active substances revealing both phytotoxic and stimulating effects on the germination and germination vigour of sugar beet seeds. The water extracts of 'Medovyi' seeds showed the lowest allelopathic effect compared to other cultivars under study. Water extract of 'Sylosne 42' appeared relatively tolerant to germination of sugar beet seeds.
The extract of 'Medovyi' seeds was filtered and added into the agar medium on which the clones of *Beta vulgaris* L. were planted. Allelopathically active substances did not affect sugar beet plants on the 7th day after planting. A decrease in the number of buds, leaves and plant height was recorded on the 14th day. On the 21st day, sugar beet plants looked suppressed and eventually died off. Understanding of physical and biochemical mechanisms of plants interaction allows selecting physiologically compatible plants for high-productive phytocoenoses.

Keywords: germination vigour, inhibiting effect, *in vitro*, seed extracts

INTRODUCTION

Scientific and technological progress builds on the created by man material resources and technologies which often do not consider natural phenomena. This resulted in a number of such negative consequences as an ecological crisis, the lack of raw resources, etc. The most serious changes in the environment are connected with agro-industrial production, and they are clearly seen in the disorder of natural, biological and geological cycles of substances and energy, the reduction of biological diversity, the change of structure and major properties of natural landscapes, pollution and violation of the reproduction processes of renewable resources. Therefore, at the current state of the economy, allelopathy, which is a rather new tendency in biology, is becoming the most important trend to save biological diversity and to expand plant resources (**Khalaj *et al.*, 2013**; **Uddin, 2014**). The physiologically active substances demonstrate allelopathic properties; their chemical nature is very diverse and unstable even within one plant. Allelopathic relationships are very complex because the direct and indirect effects are closely intertwined. Chemical regulation in a plant grouping exists due to the fact that every plant creates a certain allelopathic sphere around it, that is, accumulates collines. The higher the concentration of these collines, the worse the components of the coenosis will grow. **Gpiazdowska and Bogatek (2005)** presented five levels of plant organization where the effect of allelopathically active substances is manifested: molecular, structural, biochemical, physiological and ecological. In this article, the levels are highlighted in connection with the allelopathically induced symptoms and mechanisms.
Allelopathic relationships are one of the most complex because direct and indirect effects are closely connected (**Jafariehyazdi and Javidfar, 2011**). Therefore, the central issue of allelopathy is the study of the presence, concentration and chemical composition of collines at all stages of plant development, their physiological activity and the role in agrophytocoenosis. Therefore, the composition of stable high-productive agrophytocoenoses in regard to allelopathic effect is an urgent task which needs a solution (**Haig, 2008; Weir *et al.*, 2008; Moosavi *et al.*, 2011**). It was shown that most agricultural crops have certain allelopathic effects that can be revealed already at the cell level, specifically, inhibition of enzyme activity, changes in membrane permeability, in exchange of proteins and organic fatty acids; allelopathy affects hormonal status which can activate or suppress antioxidant cell systems. At the level of the whole organism, there are the following effects: stretching of cells, changes in the intensity of respiration, in membrane permeability, in the size of leaf stomata, inhibition of photosynthesis and growth **(Rice, 1974; Nekonam *et al.*, 2014; Arowosegbe and Anthony, 2012; Kheibaryan *et al.*, 2012; Weston *et al.*, 2013; Shtyka *et al.*, 2015**).
Sorghum (*Sorghum bicolor* (L.) Moench) is one of the crops possessing a strong allelopathic potential, i.e. capable of forming and allocating physiologically active substances in the medium, and allelopathic tolerance i.e. tolerates the active secretions of other crops. The allelopathic potential of sorghum is demonstrated in numerous publications **(Marinov-Serafimov, 2010; Alsaadawi and Dayan, 2009; Shah *et al.*, 2016; Głąb *et al.*, 2017). Storozhyk (2016)** found that vegetative and generative parts of sugar sorghum contain biologically active substances such as glycosides, tannins and acids that affect the development of other crops in mixed cropping, specifically height and the number of leaves, the content of sugar, starch, fat, protein, etc.
Growing sugar sorghum is advisable due to its high productivity and versatile applications. In Ukraine, at the beginning of the 1990s, the area under sorghum was 20,000–25,000 hectares. Since the beginning of the 2000s, the area under the crop increased to 76,000 hectares. Such a considerable increase in the area under this crop is due to the fact that sorghum is an alternative feedstock for biofuel production and can be grown to harvest grain and green mass (**Saballos, 2008**). However, it is not advisable to grow sorghum as a single crop for fodder. Previous research showed that sorghum productivity increases in mixed cropping with maize, soybean and sugar beet **(Elemo, 2010; Sani *et al.*, 2011; Kheibaryan *et al.*, 2012).**
Mixed planting of two fodder crops is studied and widely used worldwide Mixed cropping of biologically compatible components is a tool to both increase protein content and the yield of fodder **(Javanshir *et al.*, 2001; Ngongoni et al., 2008).** However, mixed germination of seeds of two crops has ambiguous effects on the development of each crop. For example, when germinating, seeds of sugar sorghum release biologically active substances that

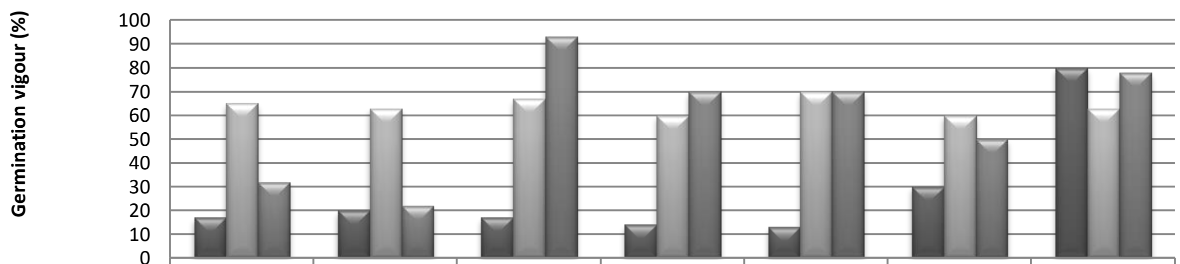
can affect the growth and development of the other crop in mixed cropping **(Storozhyk *et al*., 2015, 2016).**
In view of this, the study of allelopathic substances released by sugar sorghum seeds and their effect on other crops in mixed cropping deserves a special attention. In consideration of the above, the purpose of this research was to study the allelopathic effect of sugar sorghum on the germination of sugar beet seeds.

MATERIALS AND METHODS

Allelopathic activity of sugar sorghum was studied using a technique by **Grodzinsky and Grodzinsky (1973), Scott (2008), Thompson and Hannah (2008)** and methodological recommendations developed by the Institute of Bioenergy Crops and Sugar Beet of the National Academy of Agricultural Sciences of Ukraine (**Voitovska *et al*., 2016; Ingle *et al*., 2018**). The extraction method was used to study the chemical interaction of plants.
In the experiment, fresh seeds of sugar sorghum hybrids 'Medovyi', 'Dovista' and variety 'Sylosne 42' were used. A 50 g sample of crushed seeds was placed in a glass container, and 250 ml of distilled water was added (1:5). The container was shaken and covered with a lid. The extraction process lasted 24 h at a temperature of +20°C. After that, the extraction solution was poured into a reservoir. Sugar beet seeds of hybrid 'Ukrainskyi MS 97' were put on a filter paper in Petri dish and 7 ml of the extracted solution was added. Petri dishes were put into a thermostat at a temperature of +25°C. Germination vigour was determined after 4 days, and germination after 7 days. The experiment was designed in eight replications. The results were compared to the control treatment. Water extractions of various concentrations (5%−50%) were prepared. Distilled water was used in the control treatment. In the evaluation of sorghum allelopathic effect on sugar beet, we used the most common sorghum hybrid 'Medovyi' with a sugar content of 18%. The extracts of crushed sugar sorghum seeds were filtrated and added to agar culture medium according to the **Murashige and Skoog (1962)** recommendations with some modifications **(George *et al*., 2008).** Clones of sugar beet were planted *in vitro* on the culture medium. The design of the experiment was as follows: 1) culture medium without sorghum seed extracts (control); 2) culture medium with sorghum seed extracts. The following parameters were used to evaluate the growth rate: the number of buds, the number of leaves and plant height. General condition of the plant was assessed by a five-point scale, where 5 – plants are green and have newly-formed buds; 4 – yellowing of lower leaves, weak bud-formation; 3 – partial change in plant colour and no bud formation; 2 – change in plant colour and partial necrosis; 1– necrosis of 95% of the plant. Analysis of variance, software *Microsoft Excel 2010* and *AgroStat* (Trial version) was used in statistic processing of the obtained data.

RESULTS

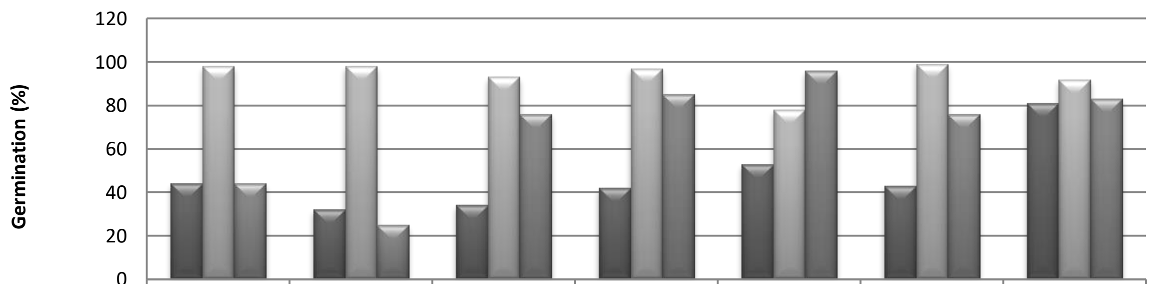
The results of the experiment on the chemical interaction of crops showed that sugar sorghum seeds during germination have both a negative and positive effect on germination and germination vigour of sugar beet. It is important to note that regardless of some cases of stimulating effect, sorghum extracts mostly caused suppression (inhibition) of sugar beet seeds germination. It was found that water extracts from the seeds of 'Medovyi' demonstrated a lesser allelopathic effect compared with other cultivars under study (Fig. 1).



	5%	10%	20%	30%	40%	50%	control
hybrid Medovyi	17	20	17	14	13	30	80
cultivar Sylosne 42	65	63	67	60	70	60	63
hybrid Dovista	32	22	93	70	70	50	78

Figure 1 Allelopathic effect of sugar sorghum water-soluble extracts of various concentrations on germination vigour of sugar beet seeds

In the treatments with 40% water extract from the seeds of 'Medovyi', germination vigour of sugar beet seeds was 13%, which is lower than the control indicators by 83.7% (Fig. 2). Water extracts of 'Medovyi' with a concentration of 5% and 10% demonstrated a considerable phytotoxic effect on sugar beet seeds (Fig. 3).



	5%	10%	20%	30%	40%	50%	control
hybrid Medovyi	44	32	34	42	53	43	81
cultivar Sylosne 42	98	98	93	97	78	99	92
hybrid Dovista	44	25	76	85	96	76	83

Figure 2 Allelopathic effect of sugar sorghum water-soluble extracts of various concentrations on germination vigour of sugar beet seeds

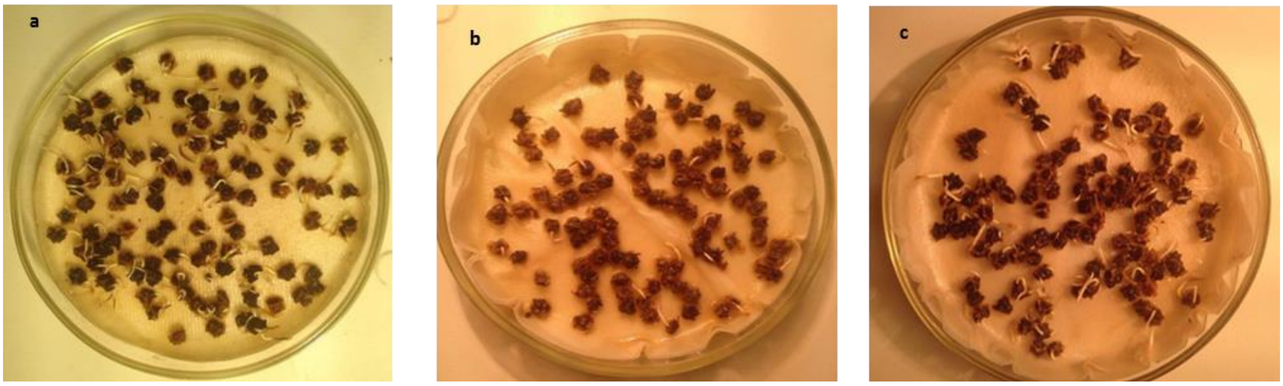


Figure 3 Effect of water extract of sugar sorghum hybrid 'Medovyi' on germination vigour of sugar beet seeds: (A) distilled water – control; (B) 10%; (C) 40%.

Figure 4 Effect of water extract of sugar sorghum hybrid 'Dovista' on germination vigour of sugar beet seeds: (A) distilled water – control; (B) 5%; (C) 10%.

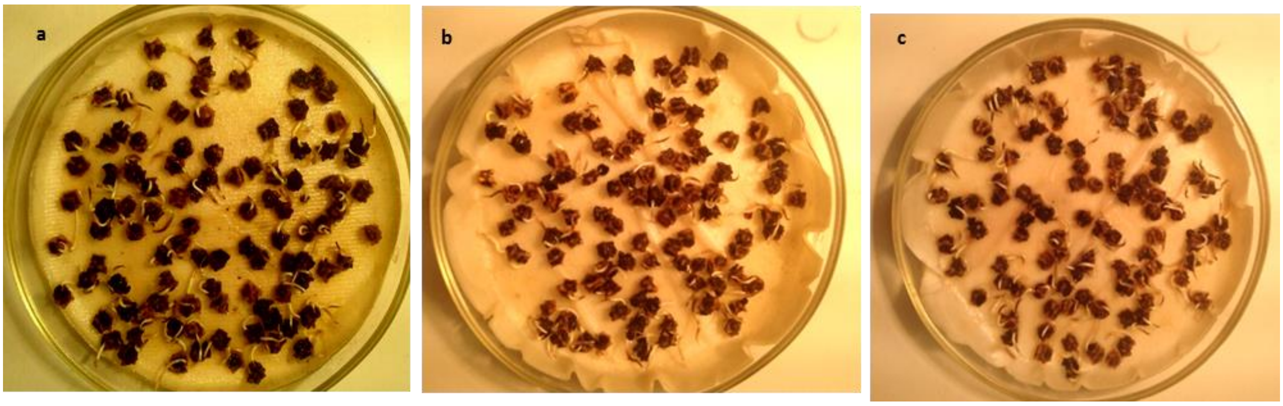


Figure 5 Effect of water extract of sugar sorghum variety 'Sylosne 42' on germination vigour of sugar beet seeds: (A) distilled water – control; (B) 10%; (C) 40%.

Water extracts (concentrations 5% and 10%) of 'Dovista' caused a serious decrease in germination vigour of sugar beet seeds as compared to the control by 58.9% and 71.8%, respectively (Fig. 4).

Allelopathically active substances from water extracts of 'Sylosne 42' appeared to be relatively tolerant to sugar beet seeds, with 63–65% germination vigour, i.e. equal to control treatment (Fig. 5).

Accordingly, water extracts from sugar sorghum of various cultivars have both a phytotoxic and stimulation effect on germination and germination vigour of sugar beet seeds. A low stimulation effect (2–5% compared to control) was recorded in the treatments with 10% solution of 'Sylosne 42' and manifested in more intensive seed germination.

To study the allelopathic effect of extracts from sugar sorghum seeds *in vitro*, biometric indicators of plants were measured on the 7th, 14th and 21st day after planting. The results of the experiment showed that on the 7th day, the number of newly formed buds in the treatments was smaller by 0.26 ± 0.04, as compared with the control. The number of leaves and a plant height also decreased (3.78 ± 0.04 and 1.6 ± 0.04 cm, respectively). The general condition of the plants was scored 4. This proves that on the 7th day, allelopathically active substances have no effect on the plant (Table1).

Table1 Biometric indicators of sugar beet clones *in vitro* (n = 8) as affected by water extracts from sugar sorghum seeds (hybrid 'Medovyi')

Treatment	Number of buds	Number of leaves	Plant height (cm)	The general condition of plants (score)
7th day				
Without extracts – control	1.94 ± 0.03	4.95 ± 0.02	1.9 ± 0.03	5
Culture medium with seed extract	1.68 ± 0.05	3.78 ± 0.04	1.6 ± 0.04	4
14th day				
Without extracts – control	2.73 ± 0.04	6.12 ± 0.03	3.5 ± 0.04	5
Culture medium with seed extract	1.34 ± 0.04	3.35 ± 0.04	1.6 ± 0.03	3
21st day				
Without extracts – control	3.86 ± 0.02	8.04 ± 0.02	5.0 ± 0.04	5
Culture medium with seed extract	1.21 ± 0.03	3.67 ± 0.05	1.6 ± 0.02	1

On the 14th day after planting, a decrease in the number of newly formed buds (1.34 ± 0.04) was recorded in the treatments where culture medium contained sorghum seed extract. Similar changes were observed in regard to the number of leaves and plant height. To illustrate, in the control treatment these indicators were 6.12 ± 0.03 and 3.5 ± 0.04 cm, respectively but on the culture media containing sorghum seed extract 3.35 ± 0.04 and 1.6 ± 0.03 cm, respectively. In the treatment with seed extracts, the score of the general condition of plants decreased from 5 to 3. On the 21st day in the control treatment (culture medium without sorghum extracts), the number of newly formed buds was 3.86 ± 0.02 and the number of leaves 8.04 ± 0.02, respectively, plant height was 5.0 ± 0.04 cm and general condition 5 (Fig. 6). On culture media with sorghum seed extracts, these biometric indicators were 11.21 ± 0.03, 3.67 ± 0.05, 1.6 ± 0.02 cm and 1 point, respectively.

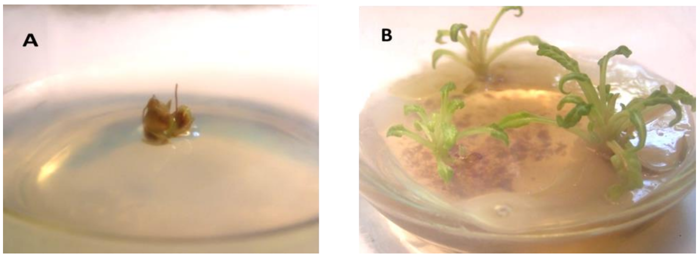


Figure 6 Sugar beet planted on culture medium without sugar sorghum seed extract on the 7th day (A) and on the 21st day (B)

On the 21st day of the experiment, the effect of allelopathically active substances resulted in gradual suppression of sugar beet plants, and eventually in their dying off (Fig. 7).

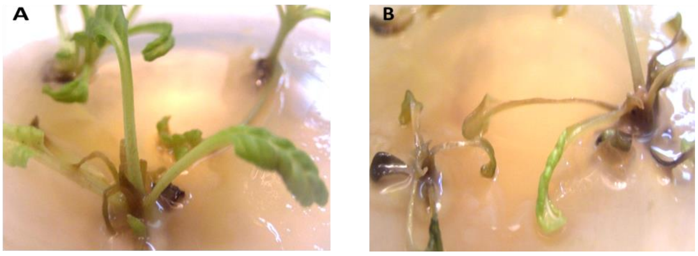


Figure 7 Sugar beets planted on culture medium with seed extract of sugar sorghum *in vitro*: 14th day, gradual suppression (A) and 21st day, death (B)

The above-described effect has to be taken into account when planning mixed cropping or using sugar sorghum as green manure. These issues have not been studied enough yet and require further investigation.

DISCUSSION

The research of Ukrainian (**Grodzinsky and Grodzinsky, 1973**; **Đikic, 2005 a; b; Koval and Yeshchenko, 2011; Storozhyk, 2015**) and foreign scientists **(Chou, 2006; Seigler, 2006; Fujii, Hiradate, 2007; Bhadoria, 2011; Baličević, 2015; Alsaadawi et al., 2015; Novak et al, 2018**) proved a possible allelopathic or chemical interaction of plants through the release of biologically active substances. For example, in the laboratory experiment, water extracts from harvest residues of barley, winter wheat, pea, maize, sunflower and spring rapeseed demonstrated a positive effect on germination vigour of spring rapeseed, speeding up its initial development. On the contrary, water extracts from sugar beet tops were toxic to plants and inhibited growth. The laboratory germination of flax seeds increased as the result of the effect of water extracts of soybean, maize, and pea harvest residues, while water extracts of sugar beet considerably suppressed germination and inhibited the initial growth of flax sprouts.
Allelopathic properties of the plant material vary by the kind of plant tissue, the age of plant and cultivar. Allelopathic activity changes in the process of ontogenesis and under the effect of external factors (**Marchi *et al*., 2008; Weston *et al*., 2013**). For example, green leaves contain many physiologically active substances; their number increases as the age of a plant increases, and decreases after plant dying off. **Kumar and Gautam (2008), Mohanan and Rajendiran (2013**) found that collines are mostly concentrated in leaves and generative organs of plants. Their content in roots is 1.6–9 times lower, and the highest values are observed at the stage of flowering. Literature data prove that seeds also demonstrate the allelopathic effect (**Moosavi *et al*., 2011; Makoi and Ndakidemi, 2012; Alsaadawi *et al*., 2015**). American researchers presented an experimentally proven hypothesis that seeds release some chemical substances in soil solution in a very little amount; being accepted by seeds of other species these substances determine a level of interaction in phytocoenosis. Because of a very small amount of extracted substances, they are regarded as 'information molecules' (**Khaliq *et al*., 2013; Weston *et al*., 2013; Yarnia *et al*., 2009; Al-Tawaha and Odat, 2010**.). There are several ways of using allelopathic activity of sorghum for weed control in field conditions. They are not limited to the growing sorghum as a single crop, or in mixed cropping. Allelopathic effect of sorghum mulch or sorghum as cover crop may be used too (**Marinov-Serafimov, 2010; Al-Bedairy *et al*., 2013; Rab *et al*., 2016; Jabran, 2017**).
Koval and Yeshchenko (2011) point out to the contradiction of allelopathic effects in laboratory and field experiment. Despite the fact that the effects obtained in laboratory conditions do not always conform to the results obtained in field, the contradiction can be explained as follows: active substances released in soil undergo fast mineralization and participate in humus formation; phenolic compounds, which are intermediaries in humus substances and available in the soil in a free state, can act allelopathically (**Tharayil *et al*., 2008a,b; Alsaadawi *et al*., 2015**). Thus, their toxic and stimulating effect are equal at the moment of sowing. **Ashrafi *et al*., (2008)** and **Al-Tawaha and Odat (2010)** found that decomposition of maize and sorghum residues increases phenolic compounds in the soil affecting barley germination. **Anjum and Bajwa (2010)** showed that allelopathically active maize substances affect wheat yield.
In a research carried out earlier it was found that when sugar sorghum was cultivated in mixed cropping with mother roots of sugar beet, a mutual effect on growth and development of both crops was observed. At the beginning of vegetation, when there is enough moisture and nutrients in the soil, each crop is supplied with an equal amount of environmental factors (solar radiation, temperature), and no sign of mutual suppression of the crops is recorded. As the main crop grows, the growth of mother roots of sugar beet slows down. In fact, this situation lasts until the harvesting of the main crop. However, some inhibition of the growth and development of sorghum was observed as well.
The results of the laboratory experiments showed that water extracts from sugar sorghum seeds have both phytotoxic and a stimulating effect on germination and germination vigour of sugar beet seeds. The degree of the phytotoxic effect depends on the concentration of extracts.
In the treatment with 40% water extract from seeds of 'Medovyi', germination vigour of sugar beet seeds was 13%, i.e. 83.7% lower compared to control; meanwhile, 70% water extract of 'Sylosne 42' increased the value of by 7% the control. The extract of 'Dovista' of the same concentration resulted in 70% germination vigour. That is, 40% extract of 'Sylosne 42' had a weak stimulating effect on sugar beet germination compared with 'Medovyi', which had a significant phytotoxic effect on seed germination.
The inhibiting effect of the extracts on germination in most cases increases along with the increase in the extract concentration (**Yarnia, 2009**). **Nganthoi (2014), Msafiri *et al*., (2013)** and others found out that a high concentration of water extract of *Parthenium hysterophorus* leaves also had a strong inhibiting effect on the germination of some cultivated and wild plant varieties. It was shown that the allelopathic effect of all sunflower extracts increases as their concentration increases (**Ashrafi *et al*., 2008**). **Devi and Dutta (2012)** recorded a strong positive correlation between the increase in the concentrations of harvest residues extract and the decrease of a seedling height of genus *Brassica*.
The technologies of plant cultivation *in vitro* are used to study potential allelopathic activity. From the methodological point of view, it remains unclear whether phytotoxicity found *in vitro* turns into allelopathic effect under more complicated ecological conditions (**Singh *et al*., 2009; Qasem, 2010; Rad *et al*., 2014; Jandová *et al*., 2015**).
Analysis of the research results showed that allelopathically active substances from sorghum seed extracts revealed no effect on the clones and plants of sugar beet on the 7th day after planting; a decrease in the number of formed buds, leaves and plant height was observed on the 14th day and gradual suppression and eventually dying off on the 21st day.

CONCLUSIONS

Water extracts from sugar sorghum seeds of different cultivars and of various concentrations (from 5% to 50%) have both a toxic and stimulating effect on germination and germination vigour of sugar beet seeds. In the treatment with 40% water extract from the seeds of sugar sorghum hybrid 'Medovyi', germination vigour of sugar beet seeds was 13%, which was by 83.7% lower compared the control treatment. A phototoxic effect in regard to sugar beet seeds is typical for 10% water extract of hybrid 'Dovista' decreasing germination vigour by 71.8% and seed germination by 69.9% as compared to the control.
Water extracts of sorghum variety 'Sylosne 42' appeared relatively tolerant to sugar beet seed germination. In this treatment, the value of germination (63–65%) conformed to the results of the control treatment.
Allelopathically active substances from sorghum seed extracts revealed no effect on the clones and plants of sugar beet on the 7th day after planting; a decrease in the number of formed buds, leaves and plant height was observed on the 14th day and gradual suppression and eventually dying off on the 21st day.

Investigation of the possibilities of using water extracts from sugar sorghum as natural inhibitors of weed growth will be the topic of our further research.
When planning agrophytocoenoses, the effects of plant extracts should be taken into account. The random selection of physiologically incompatible components for mixed cropping may result in a sharp decline in yield and worsen its quality.

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