

## ESSENTIAL OIL FROM ORNAMENTAL PLANT *JUNIPERUS SABINA* L. (*CUPRESSACEAE* GRAY) WITH ANTIFEEDANT, REPELLENT AND DETERRENT ACTIVITY AGAINST *GALLERIA MELLONELLA* L.

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### Abstract

*Juniperus sabina* L. (Cupressaceae Gray) planted in the Central Europe as an ornamental shrub for gardening. In China twigs and leaves of *Juniperus sabina* have been traditionally used as the medicinal herb for the treatment of many ailments. At the same time, the biologically active properties of various species of *Juniperus*, including *Juniperus sabina*, are studied for the possible use of their extracts in the control of pests and plant diseases. We carry out investigation of the essential oil from dry needles of the plant *Juniperus sabina*. Essential oil was evaporated by heating a mixture of water and plant materials at atmospheric pressure, followed by the liquefaction of the vapors in a condenser and collection in receiving vessel Ginsberg according to a hydrodistillation method (HD). *Galleria mellonella* (Linnaeus, 1758) (Lepidoptera: Pyralidae) was as a test-object for studying the antifeedant effect of essential oil from *J.sabina* in the laboratory conditions. For these purpose in the Petri dishes (diameter 14 cm, 4 replicates) were placed treated (200 µl essential oil/ 1.0 g feed) and untreated feed (1.5 g both). After that in the centre dishes were placed the larvae of 2-3 instars of *Galleria mellonella*. In the control in the Petri dishes was only untreated feed. We observed by the movement of larvae. It was noted that all the larvae in the experiment moved toward to the untreated feed, some larvae lost orientation. It was established statistically significant ( $p \leq 0,05$ ) that in the experiment with the choice larvae took more than 30 to 120 seconds in search of food in comparative to the control. It was determined that the essential oil from the *Juniperus sabina* has not only a repellent effect on the larvae of *Galleria mellonella*, but also an antifeedant effect. The amount of consumed treated food was significantly lower (by 31.42%) than the untreated. It was also found that the essential oil had a deterrent effect. Two weeks after the start of the experiment, the amount of the consumed food in the experiment was significantly lower than in the control on 41.2%.

**Key words:** *Juniperus sabina*, ornamental plant, essential oil, repellent, antifeedant, deterrental, *Galleria mellonella*

*Juniperus sabina* L. (Cupressaceae Gray) is one of the most widespread conifer species in the world, native to the mountains of central and southern Europe, western and central Asia, as well as Northern Africa (Algeria). It grows in a wide variety of ecological conditions: on sand, chalk deposits, rocks, foothills and high in the mountains, rising to 3,300 m, occurring on southern stony and fine-earthed northern slopes.

Needles of *Juniperus sabina* (savin juniper or savin) contains bitter glycoside pinopicrin, gallic acid, tannins, flavonoids, resin, wax, vitamin C, organic acids and more. In addition, savin juniper contains essential oil (up to 2.5-4.8%), which has a specific odor and consists of alcohol sabinol and various terpenic compounds (Ayoshina E.N., Velichko N.A., 2004; Myrzagaliyeva A.B., Medeubaeva B.Z., 2014; Novikov O.O. et al,

2014). Such a variety of chemical compounds, as well as the availability of extraction of biologically active substances from various plant organs, has led to the fact that the savin is widely used in medicine, perfumery, for the production of various alcohol drinks, as well as dyes, lacquers, joinery, bath besom, etc.

One of the beneficial properties of extracts from *Juniperus sabina* is the availability of insecticidal activity against various species of harmful insects. Earlier, we showed the possibility of reducing the population density of dangerous pests Solanaceae - Colorado potato beetle *Leptinotarsa decemlineata* Say (Coleoptera: Chrysomelidae) and cotton budworm *Helicoverpa armigera* Hbn. (Lepidoptera: Noctuidae) with alcohol extracts from the Cossack juniper (Elisovetskaya D., 2015; Elisovetcaia D. et al., 2013; Elisovetskaya D. et al.,

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2014; Elisovetcaia D. et al., 2015; Elisovetcaia D.S., Nastas T.N., Odobescu V.A., 2013; 2014).

The purpose of this work consisted in to study the antifeedant, repellent and deterrent activity of essential oil from the *Juniperus sabina* against the *Galleria mellonella* (Linnaeus, 1758) (Lepidoptera: Pyralidae).

## MATERIAL AND METHOD

Scientific *researches* were carried out in the laboratory conditions during 2017 year in the Laboratory of Integrated Protection, Institute of Genetics, Physiology and Plant Protection, Chisinau, Republic of Moldova and in the Institute of Biodiversity Conservation and Biosafety, Department of Genetics and Plant Breeding (FAaFR), Slovak University of Agriculture in Nitra.

The needles (twigs) of *Juniperus sabina* were collected in spring, dried and crushed in a laboratory mill, Type MRP-1 (asynchronous motor, Ministry of Agriculture of the USSR, Research, Development and Production Facility "Agropribor", 1987).

There are several ways and methods to deriving of essential oil. One of the easy methods of deriving them is by distillation them. (Hesham H.A. et al., 2016). Output rate of the essential oil was defined by the Ginsberg Method (Ginzberg A.S., 1932).

Extraction oil from plant raw materials was performed according to a hydrodistillation method (HD) in which the essential oils are evaporated by heating a mixture of water and plant materials at atmospheric pressure, followed by the liquefaction of the vapors in a condenser (reflux) and collection in receiving vessel Ginsberg.

Determination of antifeedant, repellent and deterrent activities of essential oil were carry out on the 2-3 instars larvae *Galleria mellonella* L. (Linnaeus, 1758) (Lepidoptera: Pyralidae) according to standard methods (Indrumări metodice..., 2002; Elisovetcaia D. et al. 2013). However, we have made some additions to the testing scheme. Thus, for a laboratory experiments Petri dishes of a larger diameter, 14 cm, were used. In the Petri dishes were placed treated (200  $\mu$ l essential oil / 1.0 g feed) and untreated feed (1.5 g both). After that in the centre dishes the larvae of *Galleria mellonella* (in each of 5 larvae, 4 replicates) were placed. In the control in the Petri dishes was only untreated feed. We observed by the movement of larvae during the first hour after the accomodation and then daily monitored the behavior of the larvae. Also, the feed was periodically weighed (on 4, 12 and 18 days), up to the beginning of pupation of the larvae.

The statistical data processing was carried out by generally accepted methods (Dospheov B.A., 1979).

## RESULTS AND DISCUSSIONS

Our observation by the movement of larvae *Galleria mellonella* during the first hour after the accommodation in the Petri dishes showed that larvae in the experiment lost orientation (*figure 1*). They circled in the center of the Petri dishes, sometimes approached to the treated feed. And, in this case, larvae immediately, very quickly, started moving in the opposite direction, from the treated feed. This fact indicates at the presence of a repellent effect of essential oil from *Juniperus sabina*. Comparative analysis showed, that in the control larvae moved to the feed in the 5-10 seconds. In the experiment they moved to the untreated feed during 35-130 seconds. Consequently, it was established statistically significant ( $p \leq 0,05$ ) that in the experiment with the choice larvae took more than 30 to 120 seconds in search of food in comparative to the control (*figure 1, 2*).



Note: treated feed with essential oil marked "V"

Figure 1. Distribution larvae of *Galleria mellonella* L. in the Petri dishes between the untreated and treated feed with essential oil, 2017



Figure 2. Larvae of *Galleria mellonella* L. in the Petri dishes with untreated feed in the control, 2017

During experiment we weighted the feed from all Petri dishes. The results showed in the table 1. It was determined that the essential oil from the *Juniperus sabina* has not only a repellent effect on the larvae of *Galleria mellonella*, but also an antifeedant effect. The amount of consumed treated food was significantly lower (by 31.42%) than the untreated (table 1, figure 3-5).

Table 1  
Dynamics of food consumption of the larvae *Galleria mellonella* L. in the laboratory conditions, 2017

| Variants                           |                                   | Weight of <u>unconsumed</u> feed, g |                |                 |                 |
|------------------------------------|-----------------------------------|-------------------------------------|----------------|-----------------|-----------------|
|                                    |                                   | before treatment                    | through 4 days | through 12 days | through 18 days |
| EXPERIMENT                         | untreated feed                    | 1,4837 ±0,05                        | 1,3802 ±0,04   | 1,2519 ±0,05    | 0,9885 ±0,01    |
|                                    | treatment feed with essential oil | 1,5045 ±0,04                        | 1,5040 ±0,04   | 1,5043 ±0,04    | 1,4752 ±0,03    |
| CONTROL                            |                                   | 1,5058 ±0,02                        | 1,3994 ±0,06   | 1,1059 ±0,02    | 0,3541 ±0,08    |
| p≤0,05, HSD <sub>0,05</sub> = 0.43 |                                   |                                     |                |                 |                 |

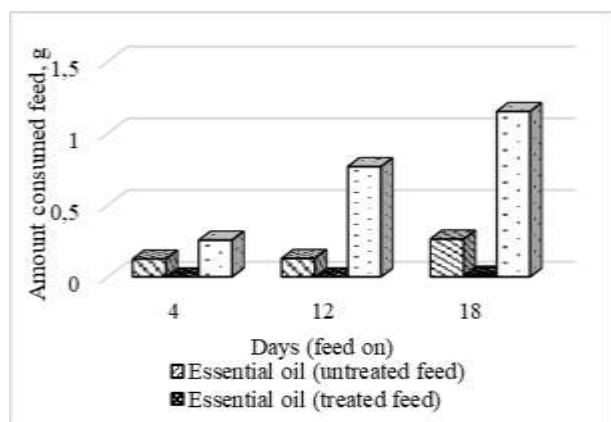


Figure 3. Amount of consumed feed by larvae (n=5, 18 days in the total) of *Galleria mellonella* L. in the laboratory conditions during the experiment, 2017

It was also found that the essential oil had a deterrent effect. Two weeks after the start of the experiment, the amount of the consumed food in the experiment was significantly lower than in the control on 41.2% (figure 3). The larvae in the experiment lagged behind in development from control and later pupated (figure 4-5).

Mathematical treatment revealed some significant differences between variants: with treatment feed of essential oil and control (p≤0,05, HSD<sub>0,05</sub>= 0.43) (table 1).



Note: treated feed with essential oil marked "V"

Figure 4. Food consumption larvae (n=5) of *Galleria mellonella* L. in the experiment after 18 days – untreated and treated feed with essential oil, 2017



Figure 5. Food consumption larvae (n=5) of *Galleria mellonella* L. in the control after 18 days, 2017

Consequently, it was noted that all the larvae in the experiment moved toward to the untreated feed, some larvae lost orientation. It was established, that the essential oil from ornamental plant *Juniperus sabina* possessed antifeedant, repellent and deterrental properties against larvae *Galleria mellonella*.

## CONCLUSIONS

Determined antifeedant, repellent and deterrental properties of the essential oil from ornamental plant *Juniperus sabina*. Amount of consumed treated food was significantly lower (by 31.42%) than the untreated, time for choice of feed in the experiment significantly increased, and was observed behind in development of larvae in the experiment from control.

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