THE EFFECT OF EXTRACTS FROM \textit{APIUM GRAVEOLENS} \textit{L.} AND \textit{LEVISTICUM OFFICINALE} KOCH LEAVES ON THE GERMINATION OF CERTAIN DICOTYLEDONS SPECIES

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Abstract: The paper presents the results of a study regarding the influence of aqueous extracts obtained from \textit{Levisticum officinale} and \textit{Apium graveolens} leaves on seeds germination and seedlings growth in \textit{Raphanus sativus} and \textit{Lens culinaris} species. The following parameters have been determined: the pH value of aqueous extracts; the germination percentage between 24 - 96 hours, and the average length of the root. The results of the investigations that were carried out reveal the following aspects: the aqueous extracts from leaves have a low acidic - neutral pH; the germination percentage and the average length of the seedlings root (at seven days after the experiment’s setting up) present specific value variations according to the extract’s concentration, the type of extract and the test species that was used. It was noticed a delay of the growth in length of the root at both species.

Keywords: \textit{Apium graveolens} \textit{L.}, \textit{Levisticum officinale} Koch, aqueous extracts, germination.

Introduction

The studies in the specialty literature have highlighted that the aqueous extracts obtained from seeds, leaves, stems from different species of umbelliferous plants have an effect of inhibition which is not specific to germination (Mihăilescu, 1958; Füzi et al., 1965; 1966; Lamoureux and Koning, 1998) or self-inhibitory effect (Friedman et al., 1982).

The above-mentioned effects are due to the presence in the vegetal organs of certain inhibitory +/- specific substances, with a varied chemical nature (aromatic organic acids, terpenoids, phenolic compounds, coumarins, furocoumarins etc.) (Bewley and Black, 1978; Friedman et al., 1982; Bilderback, 1985; Lamoureux and Koning, 1998; Kordali et al., 2007; Razavi, 2011).

The paper continues our investigations regarding the influence of some extracts from different organs from the umbelliferous species which is exerted on the seeds’ germination and growth processes in the first ontogenetic stages (Stratu et al., 2002; 2012; Zamfirache et al., 2002).

Starting from these reasons, in this paper are presented aspects regarding the influence of aqueous extracts obtained from leaves of celery (\textit{Apium graveolens} \textit{L.}) and lovage (\textit{Levisticum officinale} Koch) regarding the germination of seeds and the growth of seedlings at the species of \textit{Raphanus sativus} \textit{L.} and \textit{Lens culinaris} Medik.

Materials and methods

As biological material, we used fresh leaves, harvested from celery (\textit{Apium graveolens} \textit{L.}) and lovage (\textit{Levisticum officinale} Koch) plants. Were studied two test species: seeds of radish (\textit{Raphanus sativus} \textit{L.}) and lentil (\textit{Lens culinaris} Medik.)

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The seeds were placed to germinate in Petri dishes, on a filter paper humidified with distilled water (control variant) and aqueous extracts (obtained by hot extraction) with different concentrations (5%, 10% and 20%). Were used two type of extract: not filtered through powder of animal coal and filtered through powder of animal coal (Table 1).

To obtain the aqueous extracts, the amount of biological material (5 g; 10 g; 20 g) was kept in a volume of distilled water (95 ml; 90 ml; 80 ml) at 90°C. Extraction time was 30 minutes. After filtering half the volume of extract obtained was passed through animal coal powder. The volume of liquid (aqueous extracts / aqueous extracts) used to wet the filter paper was 8 ml – at assembling the experiment. During the experiment, germination substrate was wetted with aqueous (control variant) and aqueous extracts (treatment variants).

For each test species were used 100 seeds for each experimental variant. For each test species were made seven experimental variants: a control and six treatment variants (Table 1).

Table 1. The experimental variants

<table>
<thead>
<tr>
<th>Experimental variants</th>
<th>The type of extract</th>
</tr>
</thead>
<tbody>
<tr>
<td>M</td>
<td>distilled water</td>
</tr>
<tr>
<td>V1</td>
<td>the aqueous extracts from leaves; concentration 5 % not filtered through powder of animal coal</td>
</tr>
<tr>
<td>V2</td>
<td>the aqueous extracts from leaves; concentration 5 % filtered through powder of animal coal</td>
</tr>
<tr>
<td>V3</td>
<td>the aqueous extracts from leaves; concentration 10 % not filtered through powder of animal coal</td>
</tr>
<tr>
<td>V4</td>
<td>the aqueous extracts from leaves; concentration 10 % filtered through powder of animal coal</td>
</tr>
<tr>
<td>V5</td>
<td>the aqueous extracts from leaves; concentration 20 % not filtered through powder of animal coal</td>
</tr>
<tr>
<td>V6</td>
<td>the aqueous extracts from leaves; concentration 20 % filtered through powder of animal coal</td>
</tr>
</tbody>
</table>

We analyzed the following indicators: the pH of extracts (a CONSORT C532 multiple parameters were used); the percentage of germinated seeds (at 24 - 96 hours after assembling the experiment); the length of root at seven day since the experiment beginning. The data obtained from the length of the root were interpreted statistically. It was used the unifactorial Anova test and in order to test the difference between averages the Tukey test was used (Zamfirescu and Zamfirescu, 2008; Microsoft Excel program).

Results and discussions

The germination of seeds. The germination percentage increases progressively in the analyzed range.

For the lentil, 24 hours after assembling the experiment, the percentage of germination presented the following values: 19% at the witness variant; between 3% - 7% for the variants of treatment with extracts of celery (Fig. 1). In the variants treated with extracts of lovage, there were not noticed any germinated seeds (Fig. 2). After 96 hours, the percentage of germination of the variants of treatment presented equal values (100%) or near to the control (97 - 99%).
For the radish (Figs. 3, 4), 24 hours after assembling the experiment, the percentage of germination registered the following values: 86% at the control variant; between 32% - 78% for the variants of treatment with extracts of celery; between 78% - 92% for the variants of treatment with extracts of lovage - V1, V2, V3, V4. For the variants V5 and V6, there were not noticed any germinated seeds. After 96 ore, the percentage of germination of the control variant registered an average intermediary value (97%) of the ones obtained for the variants of treatment (between 83 - 97% for the variants of treatment with extracts of celery; 96 - 99% for the variants of treatment with extracts of lovage). The extract of celery with concentration 20%, not filtered through powder of animal coal, presented an effect of delay with 14% of the germination of the seeds of radish.

![Figure 1](image1.png)

Figure 1. The percentage of germinated seeds of lentil – aqueous extracts of celery

![Figure 2](image2.png)

Figure 2. The percentage of germinated seeds of lentil – aqueous extracts of lovage

The pH of the extracts used for the treatments presented values slightly acid – neutral, ranged between 6.7- 7.25 for the extracts of celery, and between 6.74 and 7.97 for
the extracts of lovage. Comparing with the extracts not filtered through coal powder, the extracts filtered through coal powder presented values less high, respectively neutral.

**Length of the root.** For the two species test analysed, the average length of the root for the control variant presented values superior to those recorded for the variants of treatment with extracts of lovage and celery.

For the variants of treatment, the average length of the root registered a decrease in value with the growth of the concentration of the extract, except for the test species of radish treated with extract of celery (Tables 2, 3). This fact makes us state that the aqueous extracts obtained from leaves of lovage and celery have an effect of delay of growth in length of the root for the test species.

![Figure 3](image1.png)

**Figure 3.** The percentage of germinated seeds of radish – aqueous extracts of celery

In the case of the extracts not filtered through powder of coal, the average length of the root for the test species presented higher (lentil - extracts of lovage; radish - extracts of
celery) or lower (lentil - extracts of celery; radish – extracts of lovage) values than in the case of the extracts filtered through powder of coal.

The statistics of unifactorial Anova test (F calculated : 4.77 – length of root at radish – variants treated with extract of celery; 5.03 – length of root at lentil - variants treated with extract of lovage; 28.55 – length of root at radish - variants treated with extract of lovage) was higher than the critical value (2.20 for length of root at radish - variants treated with extract of celery; 2.52 for length of root at lentil - variants treated with extract of lovage; length of root at radish - variants treated with extract of lovage); p<0,001 (length of root at radish – variants treated with extract of celery; length of root at radish - variants treated with extract of lovage); p<0,01 (length of root at lentil - variants treated with extract of lovage). This fact indicates that the used extracts have a significant influence on the growth in length of the root.

Table 2. The length of the root at test species- aqueous extracts of celery

<table>
<thead>
<tr>
<th>Experimental variants</th>
<th>Lentil Average</th>
<th>Standard deviation</th>
<th>Radish Average</th>
<th>Standard deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>M</td>
<td>95.23</td>
<td>30.10</td>
<td>82.38</td>
<td>18.5</td>
</tr>
<tr>
<td>V1</td>
<td>86.92</td>
<td>29.98</td>
<td>58.69</td>
<td>16.1</td>
</tr>
<tr>
<td>V2</td>
<td>90.46</td>
<td>17.73</td>
<td>58.46</td>
<td>13.61</td>
</tr>
<tr>
<td>V3</td>
<td>84.23</td>
<td>34.99</td>
<td>67.07</td>
<td>16.03</td>
</tr>
<tr>
<td>V4</td>
<td>89.84</td>
<td>20.46</td>
<td>57.92</td>
<td>10.71</td>
</tr>
<tr>
<td>V5</td>
<td>69.38</td>
<td>11.25</td>
<td>65.92</td>
<td>11.97</td>
</tr>
<tr>
<td>V6</td>
<td>76.53</td>
<td>16.29</td>
<td>61.23</td>
<td>11.66</td>
</tr>
</tbody>
</table>

Table 3. The length of the root at test species- aqueous extracts of lovage

<table>
<thead>
<tr>
<th>Experimental variants</th>
<th>Lentil Average</th>
<th>Standard deviation</th>
<th>Radish Average</th>
<th>Standard deviation</th>
</tr>
</thead>
<tbody>
<tr>
<td>M</td>
<td>95.23</td>
<td>30.10</td>
<td>82.38</td>
<td>18.5</td>
</tr>
<tr>
<td>V1</td>
<td>78.07</td>
<td>15.52</td>
<td>41.61</td>
<td>10.5</td>
</tr>
<tr>
<td>V2</td>
<td>74.76</td>
<td>16.48</td>
<td>52.38</td>
<td>17.35</td>
</tr>
<tr>
<td>V3</td>
<td>77</td>
<td>15.74</td>
<td>32.15</td>
<td>10.37</td>
</tr>
<tr>
<td>V4</td>
<td>62.16</td>
<td>12.15</td>
<td>37.15</td>
<td>7.18</td>
</tr>
</tbody>
</table>

The results of the Test Tukey in the case of the test species of radish indicate the following: the extracts of celery of concentration 5% (non-filtered and filtered through powder of animal coal), 10% and 20% filtered through powder of animal coal influence significantly, unfavourably the growth in length of the root comparing with the control; all the extracts of lovage influence significantly, unfavourably the growth in length of the root comparing with the control.

The results of the Test Tukey in the case of the test species of lentil indicate the following: extracts of lovage of concentration 10% (filtered through powder of animal coal) influence significantly, unfavourably the growth in length of the root comparing with the control.

Comparing with the lentil, the radish was more sensitive to the treatment with extracts obtained from the two species of umbelliferous. In other experiments carried out,
we have noticed the fact that the aqueous extracts of concentration 5% and 10% obtained from leaves of *Anethum graveolens* delay the germination of the seeds and the growth of the radish root 72 hours after assembling the experiment (Stratu et al., 2012).

The aqueous extracts obtained from dried leaves (of concentration 1%, 5%, 10%) of *Heracleum sphondylium* (Fuzi et al. 1965), *H. lanatum* and *H. lehmanianum* (Füzi et al., 1966) inhibit seed germination of *Sinapis alba, Brassica nigra, Linum usitatissimum, Sorghum vulgare*. The aqueous extracts obtained from dried leaves of *Heracleum sphondylium* (Fuzi et al., 1965), negatively influenced the increase in length of root of *Sinapis alba, Brassica nigra* and *Linum usitatissimum*. The *Heracleum* species contain furanocoumarins (sphondin, pimpinellin, bergapten, etc) (Füzi et al., 1965; 1966; Steck, 1970).

According to the literature information, the delay of the process of growth in length of the root during the first ontogenetic stages would be due to the substances with inhibiting potential present in various organs of the plant, in our case leaves, substances which, once entered with the water in the seeds of the test species, would determine a series of modifications.

The results obtained might be due to the chemical composition of the leaves, especially to the chemical composition of the volatile oil. The literature of specialty quotes the presence in leaves: in celery and lovage, of some macro-elements, in significant quantities, microelements (Enăchescu, 1984; Popa et al., 2010), carbohydrates, proteins, fats, carotenoids, vitamin C (Enăchescu, 1984; Caunii et al., 2010), essential oil (Enăchescu, 1984; Raal et al., 2008; Burzo and Toma, 2012); in celery – flavones glycosides, bitter principles, vitamins B1, B2 (Enăchescu, 1984); in lovage – umbelliferone and coumarin (Enăchescu, 1984), polyphenolic compounds (Najda et al., 2003).

In the composition of the leaves of celery there are monoterpenes (myrcene, α limonene, α și β pinene, etc.) in significant quantities, sesquiterpenes (β-cariophyllene, α și β-selinene etc.) (Enăchescu, 1984; Sipailiene et al., 2005; Burzo and Toma, 2012), derivatives phthalides (Enăchescu, 1984).

In the composition of the essential oil of lovage leaves, were identified monoterpenes (β-phellandrene, α - terpinyl acetate in high concentration, α și β pinene, trans β- ocimene, terpinene, α-terpineol) (Raal et al., 2008; Burzo and Toma, 2012). The research carried out by Kordali et al. (2007), showed the fact that the monoterpenes inhibit the germination of the seeds and the growth of the seedlings in species of weeds.

**Conclusions**

The extracts, in the used concentrations influence specifically the germination and growth processes in the first ontogenetic stages.

It was noticed a delay of the growth in length of the root at both species.

Between the two analysed species, the radish was more sensitive to treatment with extracts, in the used concentrations.
REFERENCES


