ASPECTS REGARDING THE QUALITATIVE AND QUANTITATIVE PHYTOCHEMICAL ANALYSIS OF THE *INULA HELENIUM* L. SPECIES

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Abstract: In the present paper, we aim to achieve a study referring to the qualitative and quantitative chemical composition of *Inula helenium* L., a species known in our country under the name of horse-heal. The rhizome and roots of the plant have therapeutic use in human and veterinary medicine. In the rhizome and the roots there is inulin in a ratio of 44% (Grigorescu et al., 2001; Istudor, 2001), volatile oil (1-3%), superior terpens as β-elemen, fridelin, stigmasterol, alantol, and also phenolic acids, mucilages, proasulens, E-vitamins and saponins (Bruneton, 1995; Grigorescu et al., 2001). The presence of active principles confers it choleretic, colagog, diuretic, antiinflammatory and antihistaminic properties. It has a diuretic action favouring water elimination, that of nitrogens and chlorides in rheumatism and gout, in renal affections, but especially in affections of infectious nature (Temelie, 2006). It also has antiinflammatory properties in hepatic congestions, in cholecystitides, in renal and biliary lithiasis, oliguria, arthritis (Pârvu, 2003, 2006). The inulin extracted from the roots represents a polysaccharide without nutritive value, but when swallowed renders a sensation of repletion, yet it does not determine the increase of glycemia (Ciulei et al., 1993). In this respect we analyzed the aerial vegetative organs: the inflorescences (*Inulae flos*), and the underground vegetative organs: the roots (*Inulae radix*). We performed both qualitative chemical analyses made up of the study of the volatile oil fractions, of the flavonoids, of the polyphenolic acids and of the triterpenes with the help of TLC, and quantitative chemical analyses constituted of spectrophotometric determinations.

Keywords: *Inula helenium* L., polyphenols, volatile oil, interspecific variability.

Introduction

*Inula helenium* L. is a plant used in folk medicine of numerous countries from Europe and Asia. The extracts obtained from horse-heal (*Inula helenium* L.) are used, mainly, due to its content of inulin, volatile oil, alantolactons and helenin (Bruneton, 1995).

In our study, we analyzed the chemical composition of five samples of *Inula helenium* prelevate in July 2011 (the inflorescences) and in November 2011 and March 2012 (the roots), from the north-eastern area of Moldavia, the *Miclauseni* population.

The objectives targeted by the performed phytochemical study comprised the determination of the level of the biosynthesis of flavonoidal, polyphenolic, triterpenic and volatile compounds.

Materials and methods

The vegetal material was made up of samples belonging to the *Inula helenium* species harvested in the anthesis phase, in the neighbourhood of Miclauseni, Iasi County.
The vegetal material was dried at room temperature and extracted in absolute methanol (DER=2.5:100g/mL), in warm conditions, to exhaustion. To get an idea of the polyphenolic, triterpenic and volatile compounds spectra that exist in the vegetal material, we first performed a Thin Layer Chromatographic Study (S) using for this the exhaustions which had the ratio drop:extract (DER ) 2.5:100 g/mL (Reich et al., 2006).

The spectrophotometric determinations aimed the highlighting of the level of secondary metabolites biosynthesis, the methanolic extracts having DER 2.5:100 g/mL. In these extracts, we determined the flavonoids by the treatment with a solution of alluminium chloride, when the bioactive components form internal complexes with Al3+, intensely coloured in yellow, for which the extinction was read on the spectrophotometer at $\lambda=413$nm, using luteolin as standard. The dosing of the polyphenolic acids was done by treating the extracts of phosphowolframic acid in an alkaline medium, when we obtained blue colourings, colourimetered at $\lambda=660$nm, compared to the chlorogenic acid standard (Reich and Schibi, 2006).

**Results and discussions**

The vegetal material prelevated at Miclauseni-Iasi was used to determine the content in fruitosans expressed in fruitose. This determination was made due to the fact that one of the major horse-heal root components is inulin, which is a polysaccharide (a water soluble polyfruitosan), is lacking nutritive value, but is a substitute of sugar, especially for persons with raised glycemia. The swallowing of inulin renders the sensation of repletion and does not cause the increase of glycemia.

In the same time, we determined the content in polyphenols and flavons in the aerial and flower part of *Inula helenium* (Table 1).

Table 1. The polyphenol and flavone content of *Inula helenium* samples

<table>
<thead>
<tr>
<th>Sample</th>
<th>Flavonoids (g % rutosid)</th>
<th>Polyphenolcarboxylic Acids (g% caffeic acid)</th>
</tr>
</thead>
<tbody>
<tr>
<td><em>Inula helenium</em> - inflorescence</td>
<td>0.4710</td>
<td>0.5169</td>
</tr>
<tr>
<td><em>Inula helenium</em> – herba</td>
<td>0.1038</td>
<td>0.3925</td>
</tr>
</tbody>
</table>

The greatest quantities of rutosid and caffeic acid are in the inflorescence extracts, and the smallest in *herba*. The results obtained are due to those of scientific literature (Grigorescu et al., 2001). In Figure 1 we highlight the content with a low variability in all the sample investigated but with greater values than those registered in scientific literature, Istudor (2001) mentions a variable content of 19 - 44%.
Figure 1. The variation of the frutiosan content of *Inula helenium*

![Graph showing the variation of frutiosan content](image1)

Figure 2. The variation of aminoacid content of *Inula helenium*

![Graph showing the variation of aminoacid content](image2)

The average value of the samples is 49.10 g %/d.s. with a minimum of 43.80 g%/d.s. for individual 3 (plant harvested in November) and a maximum of 58.42 g%/d.s. for individual 4 (plant harvested in March). In the same time, we analyzed the aminoacid content (Fig. 2) to evaluate the biosynthetic capacity for other secondary metabolites, too, with a rich aminoacid content that confers the plant good curative properties.

The TLC qualitative analysis highlighted the presence of volatile, terpenic, flavonoidic and polyphenolic compounds.
In the analyzed volatile oil, due to the standards used by us, we identified: limonene, linalol and β-pinene (Fig. 3).

The triterpenic compounds are represented by stigmasterol and oleanolic acid (Fig. 4).

We noticed their existence and the biosynthetic spectrum for the flavonoids in all the analyzed samples.

The flavonoids are represented by evercetol and rutosid (Fig. 6), also being present some of their aglicons (Fig. 5).

The identified polyphenolcarboxylic acids are due to the standards: caffèic acid, chlorogenic acid, p-cumaric acid and ferulic acid (Fig. 7).
Figure 5. TLC chromatogram for flavonoidic aglicons in *Inula helenium* samples

Figure 6. TLC chromatogram for flavonoids in *Inula helenium* samples

Figure 7. TLC chromatogram for polyphenolcarboxylic acids in *Inula helenium* samples

Legend: Samples: 1,2= *Inula helenium*. Flavonoid standards: Cv.=cvercetol, R=rutosid, L=luteolin, K=kemferol
Polyphenolcarboxylic acids Standards: Ac.caf.=caffeic acid, Ac.cl.= chlorogenic acid, Ac.fer.= ferulic acid, Ac.p-cum.= p-cumaric acid
Conclusions

As a result of the phytochemical study achieved on samples of *Inula helenium*, the de Miclauseni-Iasi population, we noticed that the vegetal extracts were analyzed from the phytochemical, qualitative and quantitative point of view; these contain numerous biologically active compounds.

The quantitative phytochemical analysis proved the existence of fruitosans (inulin) in great quantities, the average per experiment being of 49.10 % g /d. s. (dry substance).

The total aminoacids expressed in glutamic acid have a reduced quantitative variability except a single sample prelevated in 2011 year.

The greatest quantities of flavons and polyphenolcarboxylic acids of the rutosid type, caffeic acid respectively, are to be found in the inflorescences.

One may conclude that there is a very reduced intraspecific variability expressed both at the qualitative level the quantitative one.

REFERENCES


