FLAVONOID CONTENT AND ANTIOXIDANT ACTIVITY 
OF CARAWAY ROOTS (CARUM CARVI L.)

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Summary
Flavonoid content in the roots of caraway plants cv. Kończewicki and the antioxidant activity of extracts from those roots were determined in the first year of cultivation. The experiment revealed a significant relationship between the concentration of flavonoid compounds and the age of caraway roots. During the vegetative season the concentration of flavonoids in caraway roots gradually increased and ranged from an average level of 0.153 mg·g⁻¹ of air-dry matter (a.d.w.) in June to 0.512 mg·g⁻¹ a.d.w. in October. The largest amounts of these compounds were contained in the roots of the plants grown in 2006 (an average of 0.312 mg·g⁻¹ a.d.w.).

The ability to scavenge the DPPH radical by the extracts from caraway roots increased in direct proportion to the length of the vegetative period, flavonoid content and extract concentration.

key words: flavonoid content, antioxidant activity, DPPH, caraway, length of vegetative period

INTRODUCTION
Flavonoids are an important group of biologically active compounds that can be found in fruit and vegetables (Bravo 1998, Wolski & Dyduch 2000, Horbowicz 2000, 2003, Sembrotowicz & Czech 2005, Mysiak & Tendaj 2006). They exhibit multidirectional pharmacological action and the effects they produce in humans and animals are to a considerable degree dependent on their solubility in body fluids (Shahidi & Nazck 1995, Bravo 1998, Malolepsza & Urbanek 2000). Flavonoid compounds seal and strengthen the walls of capillary vessels, improve blood circulation in the heart muscle, have a spasmodic, diuretic, anti-aggregation effect on blood platelets, and also have anti-inflammatory, anti-ulcerative, anti-allergic and anti-hepatotoxic action. They have also been found to possess antifungal and antiviral properties, and have an
inhibiting effect on the action of some enzymes. Moreover, the importance of
flavonoids as so-called detoxifiers should be emphasized, as they easily form
complex connections (chelates) with heavy metals (Robak & Gryglewski 1996,

The effectiveness and availability of antioxidants of plant origin arouse
widespread interest. Flavonoids, apart from other natural antioxidants such as
phenolic acids, tannin and tanning agents, are an important group of secondary
metabolites capable of both preventing and eliminating the effects of reactive
oxygen species (ROS). They exhibit a high antioxidant potential and are classed
as natural non-nutrients (Smith & Yang 1994, Bravo 1998, Williamson 1999,
Troszyńska et al. 2002, Gow-Chin et al. 2002). Polyphenolic compounds, espe-
cially flavonoids, flavonones and isoflavones, enhance the action of other anti-
oxidants, e.g. vitamins soluble in fats and low-molecular substances soluble in
water. This indicates that the presence of these compounds in the diet is very
important (Politycka & Wójci-Wojtkowiak 2001, Troszyńska et al. 2002, Pod-
sędek & Anders 2002).

The fruit of the caraway plant (Carum fructus) are commonly used as sea-
soning in cooking, prepared foods, food industry, perfume and cosmetics indus-
try, etc. They are also a herbal raw material that contains mainly ethereal
(essential) oil, fatty oil, protein compounds, waxes, tanning agents, resins, fla-
vonoids, organic acids, coumarin compounds, and mineral and other salts (Ru-
way plant can also be grown as a vegetable. Use is then made of the whole
plants in the first year of cultivation – the leaves (soups, sauces, salads) and the
roots, in which there are also many biologically active compounds (Weiss 2002,
Dyduch et al. 2006).

The aim of the experiment described in this paper was to determine the
amounts of flavonoid compounds in the roots of caraway plants and to assess
the antioxidant activity of extracts from those roots in relation to their age at
harvest in the first year of cultivation.

MATERIALS AND METHODS

The experimental material consisted of the roots of caraway plants cv.
‘Kończewicki’, which had been obtained in the first year of cultivation in our
own agrotechnical experiments with caraway conducted on the Felin Experi-
mental Farm of the University of Life Sciences in Lublin. Fruits of the studied
variety of caraway were sown between the 24th and 26th of April 2005 and 2006,
on a flat plot, in rows 40 cm apart and 40 m long, with a seed barrow at 8 kg·ha−1.
After germination, the plants were thinned, leaving in each row 20 plants per
running meter, which is around 50 plants per square meter. Maintenance and
cultivation treatments were applied in accordance with the recommendations for
growing carrot. In the two years of the experiment, plants were harvested on
five different dates in the last ten days of each month from June through Octo-
ber. After harvesting, the roots of the caraway plants were separated from the
above-ground parts, cleaned and dried at 30°C.
In air-dry and pulverized material, the concentration of flavonoid compounds (mg·g⁻¹) was determined using the spectrophotometric method according to Christ & Muller (FP VI, 2002). In addition, antioxidant activity (%) measured by the ability to scavenge the DPPH radical was determined with the spectrophotometric method according to Chen & Ho (1997). For that purpose, aqueous extracts from caraway roots were prepared. The extracts were evaporated until dry and then lyophilized. The analyses were carried out on extracts at three concentrations: 20 μg·mL⁻¹, 40 μg·mL⁻¹, 80 μg·mL⁻¹. Absorbance measurements were carried out at a wavelength of λ=517 nm in a Canberra Packard UVIKON 932 spectrophotometer.

The results of the laboratory tests were evaluated statistically using variance analysis and T-Tukey’s confidence intervals at the 0.05 level of significance.

RESULTS AND DISCUSSION

The obtained results showed that the concentration of flavonoid compounds in the roots of caraway plants varied during their vegetative growth in the field in each year of the experiment (Table 1). The flavonoid content in the roots was at a level of 0.140 to 0.536 mg·g⁻¹ of air-dry weight (a.d.w.), with decidedly larger amounts of those compounds found in the roots of plants harvested in October (an average of 0.512 mg·g⁻¹ a.d.w.) than in June (an average of 0.153 mg·g⁻¹ a.d.w.). Irrespective of the harvest date, the largest amounts of flavonoids were found in caraway roots in the year 2006 – an average of 0.312 mg·g⁻¹ a.d.w. While analyzing the roots of two horseradish varieties, Horbowicz & Rogowska (2006) had revealed a different relationship. During the growing season, the level of kempherol in the analyzed roots was seen to decrease irrespective of the horseradish variety.

<table>
<thead>
<tr>
<th>Month of harvest</th>
<th>Years of experiment</th>
<th>Average for years 2005-2006</th>
</tr>
</thead>
<tbody>
<tr>
<td>June</td>
<td>0.140 a *</td>
<td>0.166 a</td>
</tr>
<tr>
<td>July</td>
<td>0.152 ab</td>
<td>0.186 ab</td>
</tr>
<tr>
<td>August</td>
<td>0.208 b</td>
<td>0.238 b</td>
</tr>
<tr>
<td>September</td>
<td>0.439 c</td>
<td>0.493 c</td>
</tr>
<tr>
<td>October</td>
<td>0.488 c</td>
<td>0.536 c</td>
</tr>
<tr>
<td>Average</td>
<td>0.273</td>
<td>0.312</td>
</tr>
</tbody>
</table>

Values followed by the same letter are not significantly different at P=5%

Studies conducted by Shahidi & Naczk (1995) revealed that rich sources of flavonoids include vegetables, fruit, seeds of various plants, some cereals, as well as red wines, tea (especially green tea), coffee, fruit juices and many seasonings. Particularly high concentrations of polyphenolic compounds can be
found in vegetable plants of the family Brassicaceae (Cruciferae) such as white cabbage, red cabbage, brussels sprouts, cauliflower, kale and broccoli (0.7-3.2 mg·g⁻¹ f.w.), and also other vegetables such as tomato (0.9-2.4 mg·g⁻¹ f.w.) and capsicum (0.6 mg·g⁻¹ f.w.). Large amounts of these compounds are also contained in the seeds of oil plants such as soya-bean, rape and flax (1.5-18.4 mg·g⁻¹ d.w.), and leguminous plants such as pea, bean and lentil (1.1-6.3 mg·g⁻¹ d.w.).

On the other hand, Mysiak & Tendaj (2006) showed that plants of the various species of alliums grown for bunch harvest can be a valuable source of flavonoid compounds in early spring. The authors found out that among the three studied species – onion, shallot, and bunching onion – irrespective of the conditions for growing in the field or forcing in a greenhouse, the largest amounts of flavonoids were present in shallot plants (an average of 0.754 mg·g⁻¹ f.w. in field-cultivated plants and 0.611 mg·g⁻¹ f.w. in greenhouse-forced plants). In our experiment, the total concentration of flavonoid compounds in the roots of caraway plants was at a level of between 0.140 and 0.512 mg·g⁻¹ a.d.w., regardless of the harvest date.

The aqueous extracts from the roots of caraway plants showed the ability to reduce the DPPH radical to diphenylpicrylhydrazine, whose amount increased with the increase in the concentration of the tested extracts (Table 2). After 30 minutes, at the extract concentration of 80 μg·mL⁻¹, the highest ability to scavenge the DPPH radical was shown by the extracts from the roots harvested in October - 47.32%. The effect of scavenging the DPPH radical by the extracts from the roots that had been harvested in the months June through September was considerably smaller and was at an average level of between 17.38% and 23.21% at the extract concentration of 80 μg·mL⁻¹.

Table 2. Antioxidant activity (%) of aqueous extracts from the roots of caraway plants expressed in terms of their ability to scavenge the DPPH radical

<table>
<thead>
<tr>
<th>Month of harvest</th>
<th>Extract concentration (μg·mL⁻¹)</th>
<th>2005</th>
<th>2006</th>
<th>Average for years</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>20 40 80</td>
<td>20 40 80</td>
<td></td>
<td></td>
</tr>
<tr>
<td>June</td>
<td>3.98 a 9.17 a 14.93a</td>
<td>4.24 a 10.11 a 19.83 a</td>
<td>4.11 a 9.64 a 17.38 a</td>
<td></td>
</tr>
<tr>
<td>July</td>
<td>4.11 a 10.02 ab 15.02 a</td>
<td>4.41 a 10.64 a 20.16 a</td>
<td>4.26 a 10.33 a 17.59 a</td>
<td></td>
</tr>
<tr>
<td>August</td>
<td>9.44 b 15.37 bc 15.94 ab</td>
<td>10.12b 15.99 bc 24.56 ab</td>
<td>9.78 b 15.68 b 20.25 ab</td>
<td></td>
</tr>
<tr>
<td>September</td>
<td>15.79 c 18.36 c 20.71 b</td>
<td>16.49 c 18.86 c 25.71 b</td>
<td>16.14 c 18.61 bc 23.21 b</td>
<td></td>
</tr>
<tr>
<td>October</td>
<td>18.03 c 18.97 c 43.31 c</td>
<td>18.67 c 19.57 c 51.33 c</td>
<td>18.35 c 19.27 c 47.32 c</td>
<td></td>
</tr>
</tbody>
</table>

Values followed by the same letter are not significantly different at P=5%

The ability of the extracts to neutralize the DPPH free radical at the concentrations of 20 and 40 μg·mL⁻¹ was below 20% in both years of the experiment and irrespective of the age of the roots. A considerably lower ability to reduce the DPPH radical was shown by the extracts from the roots of the caraway plants produced in 2005.
The ability to reduce the DPPH radical by the extracts from the roots of caraway plants was several times greater in relation to the results presented by Gawlik-Dziki & Świeca (2007), which had been obtained for extracts from dried greens of dill (8-12%), parsley (3-6%) and the fruits of black pepper (6-16%) and Nigella sativa (5-7%).

On the basis of the obtained results it can be stated that the analyzed roots of caraway plants contain chemical compounds which are secondary metabolites exhibiting antioxidant properties that make the caraway plant a vegetable of a high biological value.

CONCLUSIONS

1. Flavonoid content in caraway roots was determined. There was a significant relationship between the concentration of flavonoid compounds and the age of caraway plants.
2. Considerably greater amounts of flavonoids were found in the roots of caraway plants harvested in October (0.512 mg·g\(^{-1}\) a.d.w., on average) in comparison with the roots of the plants harvested in June (0.153 mg·g\(^{-1}\) a.d.w., on average).
3. The ability to reduce the DPPH radical to diphenylpicrylhydrazine increased with the increase in the concentration of the tested extracts.
4. The greatest ability to scavenge the DPPH radical was shown by the aqueous extracts from the roots harvested in October (47.32%, on average) at the extract concentration of 80 μg·mL\(^{-1}\).
5. The lowest ability to reduce the DPPH radical was shown by the extracts from the roots harvested in June (4.11%, on average) at the extract concentration of 20 μg·mL\(^{-1}\).

REFERENCES

Streszczenie

Oznaczono zawartość flavonoidów oraz określono aktywność antyoksydacyjną ekstraktów z korzeni kminku zwyczajnego odmiany 'Kończewicki' w pierwszym roku uprawy. Na podstawie przeprowadzonych badań wykazano istotną zależność pomiędzy zawartością związków flavonoidowych a wiekiem korzeni kminku. Podczas sezonu wegetacyjnego poziom flavonoidów w badanych korzeniach stopniowo wzrastał i kształtował się na poziomie średnio od 0,153 mg·g⁻¹ p.s.m. w czerwcu do 0,512 mg·g⁻¹ p.s.m. w październiku. Najwięcej tych związków zawierały korzenie roślin uprawianych w roku 2006 (średnio 0,312 mg·g⁻¹ p.s.m.).

Zdolność znoszenia rodnika DPPH przez ekstrakty z korzeni kminku zwyczajnego wzrastała wprost proporcjonalnie do długości okresu wegetacji, zawartości flavono- idów i stężenia ekstraktów.