PHENOLIC COMPOUNDS FROM *Lepidium sativum*

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Lepidium sativum L. (Brassicaceae), garden cress or pepper grass, is the source of valuable biologically active compounds [1]. It is used in folk medicine for cancer, uterine tumors, polyps, and other neoplasms. It is used in medicine as the alcoholic tincture, which possesses sedative and anticonvulsive activity [2].

Herein we communicate results from investigations of phenolic compounds isolated from *L. sativum* seeds grown in culture by us.

Ground air-dried raw material (seeds, 1.0 g) was extracted successively with hexane, CCl_4, and $CHCl_3$ (20 mL each), discarding the produced extracts. Then, the remaining raw material was extracted with boiling ethanol (70%) for 1 h. The extract was cooled, filtered through paper into a volumetric flask (100-mL), and adjusted to the mark with ethanol (70%). The resulting extract (2 mL) was placed into a volumetric flask (25-mL) and adjusted with the same solvent to the mark (stock solution). A series of reference solutions in ethanol (70%) was prepared in parallel. These included rutin, quercetin, luteolin, luteolin-7-glycoside, hyperoside, hesperidin, apigenin, vicenin, vitexin, 4-hydroxycoumarin, scopoletin, robinin, dicoumarin, coumarin, umbelliferone, dihydroquercetin, catechin, orientin, and gallic, chlorogenic, neochlorogenic, cinnamic, caffeic, ferulic, salicylic, and ellagic acids. The stock and reference solutions (20 μL) were injected into a chromatograph.

The qualitative composition of the phenolic compounds from *L. sativum* seeds was studied on a Gilston Model 305 (France) high-performance liquid chromatograph (HPLC) with a Rheodyne 7125 (USA) manual injector with subsequent computer processing of the results using the program Multichrom for Windows.

The mobile phase was CH_3OH:H_2O:H_3PO_4 (400:600:5). The analysis was performed at room temperature. The eluent flow rate was 0.5 mL/min; analysis time, 120 min. Detection used a Gilston Model 151 UV/Vis UV detector operating at 254 nm.

A total of 12 compounds was isolated by the HPLC analysis of *L. sativum* seeds. Of these, gallic acid (9.44%, content in the isolated mixture by internal normalization method), chlorogenic acid (14.77%), ferulic acid (5.63%), neochlorogenic acid (2.22%), luteolin-7-glycoside (14.67%), dihydroquercetin (4.37%), and quercetin (3.15%) were identified.

Quantitative analysis of phenolic compounds from *L. sativum* seeds was based on the UV absorption spectra of this group of compounds. The absorption spectra were defined using the extract (70% ethanol) of seeds and a Helios (USA) self-recording spectrophotometer in the wavelength range 210-400 nm. According to preliminary results on the identification by HPLC of phenolic compounds from *L. sativum* seeds, chlorogenic acid was the predominant component. Therefore, the absorption spectrum of chlorogenic acid in ethanol (70%) was recorded in parallel as a working standard in the same wavelength range. Absorption spectra of the studied solution and the chlorogenic acid solution had a common absorption maximum at 329 ± 2 nm. This enabled us to calculate the total phenolic compounds in *L. sativum* seeds using chlorogenic acid as a working standard. The studied solution was prepared by the method given above (weight 1.0 g). The working standard (chlorogenic acid, 0.02 g) was placed in a volumetric flask (50-mL), treated with ethanol (70%, 30 mL), stirred until dissolved, and adjusted to the mark (solution A). Solution A (1 mL) was placed in a volumetric flask (50-mL) and adjusted with the same solvent to the mark (working standard). The optical density of the resulting solutions was measured at 329 nm relative to ethanol (70%). The contents of total phenolic compounds calculated as chlorogenic acid in *L. sativum* seeds was 0.85%.

REFERENCES

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