Phytchemical Research of Roots of the Trifolium Pratense L

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The purpose of work consist in studying of biologically active substances of roots clover meadow (Trifolium pratense L.) and them antioxidant's activity. We found out in 70 % ethanol's extract of roots Trifolium pratense L: flavonoids and its glycosides, amino acids, coumarins, tannic flavonoids – mainly flavonols. The content of extracted substances, flavonoids, tannic substances and polysaccharide was determined by the standard techniques described in Russian Pharmacopoeia XI. Using HPLC was established, that in roots Trifolium pratense L. contain apigenin, quercetin, routines, chlorogen's and coffe's acids. The noted substances are found out for the first time.

By using the method of fractional extraction and column chromatography it was allocated 2 individual substances: (1) - an amorphous white powder, dissolving in hexane, benzene and ether; $R_f = 0.81$ in system of butanol acid : acetic acid : water = 4:1:5. It has violet fluorescence in UV light. (2) - colorless crystals, dissolving on ethanol, water.. $R_f = 0.65/0.48$ in system of butanol acid : acetic acid : water = 4:1:5/ 2% CH₃COOH. It has blue fluorescence in UV light.

In PMR spectrum of substance (1) there are signals of protons, characteristic for methyl, methylene groups, alcohol's –OH groups, connected double bonds. There are no signals of protons, characteristic for an aromatic ring and methoxy- groups.

In NMR ¹³C spectrum of (1) there are marked signals, characteristic for atoms of carbon of methyl and methylene groups. Signals with chemical shifts, characteristic for aromatic atoms of carbon and carbonyl- groups were not presence.

On the base of ¹H and ¹³C NMR spectra there were supposed that substance (2) is glucoside, connected with not conjugated cycle. Anomeric proton of glucose had signal at 5,11 ppm, another protons are at 3,26-4,11 ppm. There are signals, characteristic for keton CO - group which is not connected with aromatic ring. There are no signals of protons and atoms of carbon, characteristic for an aromatic ring, too. The establishment of structure of substances (1) and (2) are in proceeding. Antioxidant activity of the common extract of roots and flowers, as a separate fraction was studied. All samples have shown this specific activity one measured in relation to process of reduction of oxygen.

It was shown that mechanochemical treatment in planetary mill AGO-2 of common dry extract from roots and flowers of clover meadow raised the antioxidant activity.