

## CHARACTERISTICS OF LIPIDS FROM INDIVIDUAL ORGANS OF *Veronica beccabunga*

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*Lipids from various organs of the aquatic plant Veronica beccabunga were studied. It has been demonstrated that neutral and polar lipids are qualitatively typical of higher photosynthetic plants. Lipids in various organs of aquatic veronica were unevenly distributed. The most significant differences were observed in neutral lipids for the accumulation of glycerin esters, free fatty acids, alcohols, and hydrocarbons. The ratio of phospholipids varied considerably in the polar lipids.*

**Key words:** neutral and polar lipids, phospho- and glycolipids.

Brooklime (*Veronica beccabunga* L.) is a widely distributed Eurasian boreal forest species that is found in very moist and swampy habitats on the banks of rivers, marshes, rivers, and ponds [1, 2]. The species grows from the western border of Russia to the southern part of the Urals and from the Karelian peninsula in the north to the southern shores of lakes Ladoga and Onega. It is common in all regions of the Caucasus and missing from the steppes along the Volga [3]. It belongs to Magnoliopsida class, Scrophulariales order, Scrophulariaceae (figwort) family, and *Veronica* genus [4, 5].

The plant is used as a cholagogue, anesthetic, and antiscorbutic [6-8]. The leaves and stems can be used in food. They contain a significant amount of vitamin C [1]. The carbohydrates and related compounds, steroids, and flavonoids have been studied [7]. However, data on the lipid composition have not been published.

*Veronica beccabunga* can be assigned to the helophytes, i.e., aerial-aquatic plants, according to the ecobiomorphological classification of macrophytes found in swamps and aquifers and also the habitat conditions of the population that we studied. In addition to hydrophytes, or actual aquatic plants, this species belongs to the higher aquatic plants [8-10] that are considered secondary aquatics, i.e., adaptable to life in water [9]. Aquatic plants are similar to terrestrial plants in their phylogenetic features whereas they have different structural and functional traits [11, 12]. We compared the lipids in various organs of brooklime as a function of habitat. Plants were collected in June 1999 during rapid growth and flowering on the banks of a stream in Studenets in the south of Ul'yansovsk region. The water temperature in the stream was 7.2°C for one year. *Veronica beccabunga* inhabits the banks of this aquifer during the year. We collected leaves, stems, roots, and flowers of several plants of approximately equal height in a quantity sufficient to isolate and analyze the lipids. The leaves (9.3 mg/g dry mass) and flowers (12.5 mg/g dry mass) are richest in lipids. The neutral lipids (NL) of the leaves, roots, and flowers contain the highest amount of glycerin esters (47.4, 43.6, and 53.0%, respectively) whereas the stems have only 22.2% of them in the total NL (Table 1). Glycerin esters are the main component of the cell energy stores. The ratio of mono-, di-, and trisubstituted glycerin esters was shown that the triacylglycerides dominate in the flowers, reaching 37.5% of the total NL or 70% of the total amount of acylglycerides. Diacylglycerides are more significant in the lipids of leaves and roots (21.2 and 24.7%) whereas monoacylglycerides appear in the leaves (16.9%). Free sterols in the various organs are distributed evenly from 3.8% in flowers to 7.3% in leaves. The amount of sterol esters is greater than the sterols in all organs. Differences are observed in the distribution of free fatty acids and alcohols. Acids in the flowers make up 4.0% of the total NL; in stems, 11.4%. The alcohol content varies from 11.9 to 27.1%.

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TABLE 1. Total (mg/g Dry Mass) and Neutral Lipids (% of Total Neutral Lipids) of *Veronica beccabunga*

Lipids	Leaves	Stems	Roots	Flowers
Total lipids	9.3±0.4	1.4±0.0	3.4±0.4	12.5±2.5
Monoacylglycerides	16.9±0.6	1.8±0.1	8.4±0.5	1.9±0.2
Diacylglycerides	21.2±0.4	6.1±0.4	24.7±0.3	13.6±0.1
Triacylglycerides	9.3±0.5	14.3±0.6	10.5±1.0	37.5±2.5
Free fatty acids	8.0±0.6	11.4±0.5	10.2±0.5	4.0±0.4
Free sterols	7.3±0.3	5.1±0.3	5.3±0.3	3.8±0.2
Sterol esters	10.2±0.4	13.8±0.2	5.9±0.1	9.5±0.9
Waxes	8.2±0.7	14.1±0.8	4.3±0.1	7.8±0.3
Alcohols	11.9±0.2	24.0±1.0	27.1±0.3	16.0±1.0
Hydrocarbons	15.0±0.7	9.3±0.9	3.6±0.2	5.9±0.3

The first system was used to separate NL of leaves, stems, and flowers; the second, NL of roots.

TABLE 2. Polar Lipids of *Veronica beccabunga*, % of Total

Lipids	Leaves	Stems	Roots	Flowers
Glycolipids:				
monogalactosyldiacylglycerides	45.8±3.0	39.2±2.7	42.7±2.0	48.8±0.4
digalactosyldiacylglycerides	43.6±5.0	45.2±3.4	35.3±0.8	27.6±1.3
sulfoquinovosyldiacylglycerins	10.5±0.8	15.6±0.8	22.0±2.2	23.6±1.0
Phospholipids:				
phosphatidylcholines	53.6±1.6	22.2±0.7	31.4±2.2	29.4±2.0
phosphatidylethanolamines	2.5±0.0	8.3±0.4	20.5±1.9	6.3±0.4
phosphatidylglycerines	23.8±0.5	18.1±0.3	8.7±1.2	24.5±0.9
diphosphatidylglycerides	6.0±0.1	3.4±0.1	0.9±0.0	7.5±0.7
phosphatidylinosite	9.8±0.6	8.1±0.7	8.5±0.3	14.7±0.1
phosphatidic acid	4.3±0.5	39.9±0.6	30.0±2.4	15.3±0.8
phosphatidylserines	-	-	-	2.3±0.2

Twelve components were identified in the polar lipids of brooklime. Three of these give a positive reaction with anthrone and can be assigned as glycolipids (GL). The mole ratio of GL as measured from the amount of galactose in the lipid fractions isolated from the various organs indicates that monogalactosyldiglycerides (MGDG) dominate in the leaves, roots, and flowers. Their content varies from 42.7 to 48.8% of the total GL. Digalactosyldiglycerides (DGDG) are in second place (27.6-43.6%), then sulfoquinovosyldiacylglycerides (SQDG, 10.5-23.6%). The stems have slightly more DGDG than MGDG.

The qualitative content of phospholipids (PL) is in general typical of both terrestrial and aquatic photosynthetic plants [13]. However, the localization of phosphatidylserines (PS) in the flower lipids of veronica is an important distinguishing feature. Table 2 shows that phosphatidylcholines (PC) are the principal PL in the leaves and flowers of brooklime. Whereas the amount of PC in the leaves is 53.6% of the total PL, the PC in the flowers and stems make up only 29.4% and 22.2%, respectively, of the PL. Phosphatidylglyceride (PG), the principal phospholipid of the photosynthetic apparatus, are second in content in the PL of these organs. Its level in flowers is practically equal to that of PC. Phosphatidylethanolamines (PE) in the leaves and flowers amount to only 2.5-6.3%. The levels of synthesized phosphatidylinositols (PI) and diphosphatidylglycerols (DPG) are very similar in the PL fractions of all organs. The amount of phosphatidic acid (PA) differs more significantly. There is almost three times more of it in flowers than in leaves. PA contributes most to the PL in stems and roots (39.9 and 36.0%, respectively) (Table 2). If it is considered that PA is the principal substrate in PL synthesis, then the large quantity of PA observed by us in the roots and stems may be related to the slower exchange of lipids in these organs.

Therefore, the ratio of NL and PL in lipid extracts isolated from various organs differs considerably. The morphological and physiological differences between the individual plant parts, which led during evolution to the specialization of organs, are evident at the cellular level, in particular, they influence the lipid composition. The principal differences of the root, stem, and leaf structures consist primarily of the relative distribution of tissue systems: integumentary, conductive, and basic. Apparently, the differences in lipid composition are related to the morphological difference of the tissues and functions associated with each organ.

## EXPERIMENTAL

Lipids were extracted by the method of Bligh and Dyer [14] and were separated into individual classes by TLC on microplates (6×6 cm or 10×10 cm) with a fixed layer of silica gel (Haapsalu, Estonia). NL were separated using one-dimensional chromatography with successive application of the solvent systems toluene—hexane—formic acid (70:30:0.5, first system) and hexane—diethylether—formic acid (60:40:1, second system). Glycolipids were separated using acetone—benzene—water (91:30:8); PL, chloroform—methanol—benzene—ammonia (130:60:20:12) in the first direction and chloroform—methanol—benzene—acetone—acetic acid (140:60:20:10:8) in the second. NL were identified using TLC and comparison of the mobilities of the studied compounds and standards [stearic acid, glycerol tripalmitate,  $\beta$ -sitosterol, and heptadecanol (Sigma)]. PL were identified by known methods using specific reagents; P-containing lipids, molybdenum blue; lipids containing the trimethylammonium group, Dragendorff's solution; amino-containing lipids, ninhydrin [15]. GL were recognized from a positive reaction with anthrone reagent [16]. The amount of NL was determined by the method of Kabara and Chen [17]; GL, from the galactose content with anthrone reagent [16]; PL, by the Vaskovsky method [18].

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