Identification of phenolic constituents in *Lonicera caerulea* L. by HPLC with diode array detection electrospray ionisation tandem mass spectrometry

**Mayya P. Razgonova** 1,2*, Nadezhda G. Tikhonova1, Andrey S. Sabitov1, Natalia M. Mikhailova1, Svetlana R. Luchko1, Alexander M. Zakarenko 1,2, Konstantin S. Pikula 1, and Kirill S. Golokhvatst 1,2,3,4

1 N.I. Vavilov All-Russian Institute of Plant Genetic Resources, B. Morskaya 42-44, 190000, Saint-Petersburg, Russian Federation
2 Far Eastern Federal University, Sukhanova 8, 690950, Vladivostok, Russian Federation
3 Siberian Federal Scientific Centre of Agrobiotechnology, 633501, Krasnoobsk, Russian Federation
4 Pacific Geographical Institute, Far Eastern Branch of the Russian Academy of Sciences, Radio 7, 690041, Vladivostok, Russian Federation

**Abstract**: The purpose of this work was a comparative metabolomic study of extracts of Blue-berried honeysuckle *Lonicera caerulea* L. from (Japan): №860 (Wild *Lonicera* from Amur river) from the collection of N.I. Vavilov All-Russian Institute of Plant Genetic Resources. To identify target analytes in extracts HPLC was used in combination with a Bruker Daltonics ion trap. The results showed the presence of 82 target analytes corresponding to *Lonicera caerulea* L. There were flavonols: Dihydrokaempferol, Rhamnetin I, Rhamnetin II, Taxifolin-3-O-glucoside, Mearnsetin-hexoside, Horridin; flavones: Chrysoeriol, Apigenin-O-pentoside, Chrysoeriol-7-O-glucoside; flavanone Naringenin; flavan-3-ols: Catechin, Epicatechin, Biochanin A-7-O-glucoside; essential amino acids: L-Pyroglutamic acid, Tyrosine; polypeptide 5-Oxo-L-prolyl-L-isoleucine; sterols: Ergosterol, Fucosterol, Beta-Sitosterin; triterpenoids: Betunolic acid, Oleaonic acid; anabolic steroid Vebonol, indole sesquiterpene alkaloid Sespendede; iridoids: Monotropein, p-Coumaroyl monotropein, p-Coumaroyl monotropein hexoside; Myristoleic acid, etc.

1 Introduction

Blue-berried honeysuckle *Lonicera caerulea* L., family *Caprifoliaceae* is known as a natural source of food, beverages and nutraceuticals due to its rich chemical composition, enriched with nutrient and biologically active compounds. The increased focus on these berries is due to their phenolic composition, antioxidant activity, and potential health benefits. The high content of phenols in *Lonicera caerulea* L. is directly related to their biological activity. Popularity of phenolic compounds has grown in recent years as they are excellent antioxidants. Antioxidant intake has been shown to be effective in preventing cancer, cardiovascular disease, osteoporosis, obesity, diabetes, and other health problems [Dias et al., 2017]. The antioxidant properties of plant phenolic compounds are relevant in the field of nutrition (inhibition of lipid oxidation), physiology (protection against oxidative stress) and cosmetology. Phenolic compounds provide antioxidant activity through direct reduction of reactive oxygen species (ROS), inhibition of enzymes involved in oxidative stress, binding of metal ions responsible for ROS production, and stimulation of endogenous antioxidant defense systems [Hossain et al., 2016]. The quality and quantity of phenolic compounds in plants usually depends on the stage of growth, the parts of the plant used and the growing conditions in the environment [Bujor O.-C., 2016].

In this regard, the purpose of this work is the simultaneous assessment of phenolic compounds in the berries of *Lonicera caerulea* L. of various species collected in different climatic-geographical zones of Russia. This study is a complete qualitative study of phenols and other compounds, leading to the identification of a large number of phenolic secondary metabolites isolated from *Lonicera caerulea* L. berries of various species.

* Corresponding author: razgonova.mp@dvfu.ru (M.R.)
Initial LC-MS/MS screening suggested that 82 target analytes detected in EtOH-extracts of Blue-berried honeysuckle. Therefore, tandem mass spectrometry was used in this study for comparative small molecule profiling of four Lonicera varieties cultivated in the collection of N.I. Vavilov All-Russian Institute of Plant Genetic Resources.

2 Experimental

2.1 Materials

The object of the study was the four varieties of Blue-berried honeysuckle Lonicera caerulea L. of breeding varieties obtained as a result of many years of research from the collection of N.I. Vavilov All-Russian Institute of Plant Genetic Resources. There were a varieties: №1043-11 (St. Petersburg); №1043-08 (St. Petersburg); №863 (Japan); №860 (Wild Lonicera from Amur river). The berries were harvested at the end of July 2020. All samples morphologically corresponded to the pharmacopoeial standards of the State Pharmacopoeia of the Russian Federation [SPh XIV, Russia, 2018].

2.2 Chemicals and Reagents

HPLC-grade acetonitrile was purchased from Fisher Scientific (Southborough, UK), MS-grade formic acid was from Sigma-Aldrich (Steinheim, Germany). Ultra-pure water was prepared from a SIEMENS ULTRA clear (SIEMENS water technologies, Germany), and all other chemicals were analytical grade.

2.3 Fractional maceration.

To obtain highly concentrated extracts, fractional maceration was applied. In this case, the total amount of the extractant (methyl alcohol of reagent grade) is divided into 3 parts and is consistently infused on potato with the first part, then with the second and third. The infusion time of each part of the extractant was 7 days.

2.4 Liquid chromatography

HPLC was performed using Shimadzu LC-20 Prominece HPLC (Shimadzu, Japan) was used, equipped with an UV-sensor and a Shodex ODP-40 4E reverse phase column to perform the separation of multicomponent mixtures. The gradient elution program was as follows: 0.01-4 min, 100% CH3CN; 4-35 min, 100-25% CH3CN; 35-50 min, 25-0% CH3CN; control washing 50-60 min 0% CH3CN. The entire HPLC analysis was done with a ESI detector at wavelengths of 230 nm and 330 nm; the temperature corresponded to 17°C. The injection volume was 1 ml.

2.5 Mass spectrometry

MS analysis was performed on an ion trap amaZon SL (BRUKER DALTONIKS, Germany) equipped with an ESI source in negative ion mode. The optimized parameters were obtained as follows: ionization source temperature: 70 °C, gas flow: 41 / min, nebulizer gas (atomizer): 7.3 psi, capillary voltage: 4500 V, and plate bend voltage: 1500V, fragmentary: 280 V, collision energy: 60 eV. An ion trap was used in the scan range m / z 100 -1.700 for MS and MS/MS. The capture rate was one spectrum/s for MS and two spectrum/s for MS/MS. Data collection was controlled by Windows software for BRUKER DALTONIKS. All experiments were repeated three times. A four-stage ion separation mode (MS/MS mode) was implemented.

3 Results and discussion

Four of the most consumed extracts of Lonicera caerulea L. have been selected. All of them have a rich bioactive composition. There were four extracts from a varieties: №1043-11 (St. Petersburg); №1043-08 (St. Petersburg); №863 (Japan); №860 (Wild Lonicera, Amur river) from the collection of N.I. Vavilov All-Russian Institute of Plant Genetic Resources. High accuracy mass spectrometric data were recorded on an ion trap amaZon SL BRUKER DALTONIKS equipped with an ESI source in the mode of negative-positive ions. The four-stage ion separation mode (MS/MS mode) was implemented. The combination of both ionization modes (positive and negative) in MS full scan mode gave extra certainly to the molecular mass determination (Fig. 2,3,4). The positive-negative ion mode provides the highest sensitivity and results in limited fragmentation, making it most suited to infer the molecular mass of the separated polyphenols, especially in cases where concentration is low. By comparing the m/z values, the RT and the fragmentation patterns with the MS² spectral data taken from the literature [Abeywickrama et al., 2016; Abu-Reidah et al., 2015; Rafsanjany et al., 2015; Goufo et al., 2020; Paudel et al., 2013; Jaiswal et al., 2014; De Rosso et al., 2014; Marzouk et al., 2018; Barros et al., 2012; Pradhan & Saha, 2016; da Silva et al., 2019; Ruiz et al., 2013; Ruiz et al., 2010; Razgonova et al., 2020; Kajdzanoska et al., 2010] or to search the data bases (MS2T, MassBank, HMDB). A unifying system table was compiled of the molecular masses of the target analytes isolated from the EtOH-extract of Lonicera caerulea L.
ease of identification (Table 1). The 82 target analytes shown in Table 1 belong to different polyphenolic families: flavones, flavonols, flavan-3-ols, flavanones, anthocyanins, hydroxycinnamic acids, hydroxybenzoic acids, stilbenes, proanthocyanidins and belong to others classes of compounds.

**Fig.2.** Chemical profiles of the *Lonicera caerulea* L. (variety SPb 1043-11) sample represented total ion chromatogram from MeOH-extract.

**Fig.3.** Chemical profiles of the *Lonicera caerulea* L. (variety SPb 1043-8) sample represented total ion chromatogram from MeOH-extract.

In addition to the reported metabolites, a number of metabolites were newly annotated in *Lonicera caerulea* L. There were flavonols: Dihydrokaempferol, Rhamnetin I, Rhamnetin II, Taxifolin-3-O-glucoside, Mearnsetin-hexoside, Horridin; flavones: Chrysoeriol, Apigenin-O-pentoside, Chrysoeriol-7-O-glucoside; flavanone Naringenin; flavan-3-ols: Catechin, Epicatechin, Biochanin A-7-O-glucoside; essential amino acids: L-Pyroglutamic acid, Tyrosine; polypeptide 5-Oxo-L-propyl-L-isoleucine; sterols: Ergosterol, Fucosterol, Beta-Sitosterin; triterpenoids: Betunolic acid, Oleanoic acid; anabolic steroid Vebonol, indole sesquiterpene alkaloid Sespendole; iridoids: Monotropein, p-Coumaroyl monotropein, p-Coumaroyl monotropein hexoside; Myristoleic acid, etc.

**Fig.4.** Chemical profiles of the *Lonicera caerulea* L. (variety Wild lonicera from Amur river) sample represented total ion chromatogram from MeOH-extract.
Table 1. Identified target analytes in MeOH extracts of berries of *Lonicera caerulea* L.

| № | № of collection of N.I. Vavilov All-Russian Institute of Plant Genetic Resources | Class of compounds | Identification | Formula | Calculate d mass | Observed d mass | MS/MS Stage 1 fragmentation | MS/MS Stage 2 fragmentation | MS/MS Stage 3 fragmentation |
|---|---|---|---|---|---|---|---|---|---|
| 1 | SPb 1043-11; SPb 1043-8 | Flavonol | Kaempferol | C₁₇H₁₈O₆ | 286.2363 | 287 | 269; 149 | 239; 181 |  |
| 2 | 863; SPb 1043-8 | Flavonol | Dihydrokaempferol | C₁₇H₂₀O₆ | 288.2522 | 289 | 176; 144; 272 | 144 | 116 |
| 3 | 863; 860; SPb 1043-8 | Flavonol | Quercetin | C₁₆H₁₀O₇ | 302.2357 | 303 | 257; 146 | 229 | 201; 145 |
| 4 | SPb 1043-11 | Flavonol | Rhamnetin I [beta-Rhamnocitrin; Quercetin 7-Methyl ether] | C₁₆H₁₂O₇ | 316.2623 | 317 | 299; 213 | 267 | 239 |
| 5 | 863 | Flavonol | Rhamnetin II | C₁₆H₁₂O₇ | 316.2623 | 317 | 302 | 274; 153; 229; 153; 121 | |
| 6 | 863; SPb 1043-11 | Flavonol | Isorhamnetin [Isorhamnetol; Quercetin 3'-Methyl ether; 3-Methylquercetin] | C₁₆H₁₀O₇ | 316.2623 | 315 | 283 | 255; 211 | 227 |
| 7 | SPb 1043-8 | Flavonol | Kaempferol-3-O-hexoside | C₂₁H₂₀O₁₁ | 448.3769 | 449 | 287 | 213 | 213 |
| 8 | 863 | Flavonol | Quercetin-3-(3-O-arabinosyl)glucoside | C₂₆H₂₈O₁₆ | 596.4909 | 597 | 303; 465 | 257; 165 | 229 |
| 9 | 863; 860 | Flavonol | Quercetin 3'-O-glucoside [Isoquercetin; Hirsutrin; Quercetin 3-O-Glucopyranoside; 3-Glucosylquercetin] | C₂₁H₁₈O₁₂ | 464.3763 | 465 | 303 | 229; 165 | 201; 161 |
| 10 | 863; SPb 1043-11; SPb 1043-8; 860 | Flavonol | Taxifolin-3-O-glucoside | C₂₁H₂₁O₁₂ | 466.3922 | 467 | 449; 287 | 377; 279 | 345; 283 |
| 11 | 860 | Flavonol | Kaempferol acetyl hexoside | C₂₀H₁₉O₁₂ | 490.4136 | 491 | 257 | 183 |  |
| 12 | SPb 1043-11 | Flavonol | Mearnsetin-hexoside | C₂₀H₂₁O₁₃ | 494.4023 | 495 | 477; 387 | 387; 315; 199 |  |
| 13 | 863 | Flavonol | Horridin [Quercetin 3-Rhamnosyl-(1->2)-Rhamnoside] | C₂₁H₁₈O₁₃ | 594.5181 | 595 | 463; 432; 301 | 301 | 286 |
| 14 | SPb 1043-11 | Flavonol | Kaempferol 3-O-(6-O-rhamnosyl-glucoside) | C₂₂H₂₃O₁₄ | 594.5181 | 595 | 287 | 213 | 185 |
|   |   |   |   |   |   |   |
|---|---|---|---|---|---|---|
| 15 | 863; SPb 1043-11 | Flavonol | Rutin (Quercetin 3-O-rutinoside) | C_{27}H_{30}O_{16} | 610.5175 | 611 | 303; 197 | 285; 229; 195 | 229 |
| 16 | 860 | Flavan-3-ol | Catechin [D-Catechol] | C_{15}H_{16}O_{9} | 290.2681 | 291 | 289; 159 | 230; 127 |
| 17 | SPb 1043-11 | Flavan-3-ol | Epicatechin | C_{27}H_{30}O_{16} | 290.2681 | 291 | 273; 137 |
| 18 | 863 | Flavan-3-ol | Biochanin A-7-O-glucoside | C_{22}H_{22}O_{10} | 446.4041 | 447 | 245; 187 | 217 | 148; 182 |
| 19 | SPb 1043-11; SPb 1043-8 | Flavone | Apigenin [5,7-Dioxoxy-2-(40Hydroxyphenyl)-4H-Chromen-4-One] | C_{15}H_{14}O_{5} | 270.2369 | 271 | 225 | 179 |
| 20 | SPb 1043-11 | Flavone | Chrysoeriol | C_{16}H_{12}O_{6} | 290.2681 | 291 | 293; 197 | 195 | 135 |
| 21 | SPb 1043-11 | Flavone | Apigenin-O-pentoside | C_{22}H_{22}O_{11} | 448 | 449 | 403; 287; 216 | 347; 137 | 291 |
| 22 | 863; 860; SPb 1043-11 | Flavone | Luteolin 7-O-glucoside [Cynaroside] | C_{21}H_{20}O_{11} | 448.3769 | 449 | 287 | 269; 241; 132 | 133 |
| 23 | 860 | Flavone | Chrysoeriol-7-O-glucoside | C_{22}H_{22}O_{11} | 462.4035 | 463 | 301; 243 | 183 |
| 24 | SPb 1043-11 | Flavone | Diosmin [Diosmetin-7-O-rutinoside; Barosmin; Diosimin] | C_{28}H_{32}O_{15} | 608.5447 | 609 | 591; 531 | 531 | 487 |
| 25 | 863 | Flavanone | Naringenin [Naringenol; Naringemine] | C_{15}H_{14}O_{7} | 272.5228 | 273 | 147; 246 |
| 26 | SPb 1043-11 | Anthocyanin | Delphinidin | C_{15}H_{11}O_{7} | 303.2436 | 304 | 212; 149 | 212; 145 |
| 27 | SPb 1043-11 | Anthocyanin | Petunidin | C_{16}H_{13}O_{7} | 317.2702 | 318 | 256 | 238; 113 | 238 |
| 28 | 863 | Anthocyanin | Cyanidin-pentoside | C_{20}H_{19}O_{10} | 419.3589 | 419 | 287 | 259; 188; 133 | 160 |
| 29 | 863; SPb 1043-11 | Anthocyanin | Cyanidin-3-O-glucoside [Cyanidin 3-O-beta-D-Glucoside] | C_{25}H_{22}O_{11} | 449.3848 | 449 | 287 | 287; 213; 185; 141 |
| 30 | SPb 1043-11 | Anthocyanin | Peonidin-3-O-galactoside | C_{22}H_{22}O_{11} | 463.4114 | 463 | 301 | 286 | 258; 150 |
| 31 | SPb 1043-8 | Anthocyanin | Peonidin-3-O-glucoside | C_{22}H_{22}O_{11} | 463.4114 | 463 | 301 | 286 | 258; 200 |
| 32 | 863 | Anthocyanin | Peonidin 3-O-acetyl hexoside | C_{24}H_{25}O_{12} | 505.4841 | 506 | 303; 487 | 303; 229; 165 | 201; 159 |
| 33 | SPb 1043-11; SPb 1043-8; | Anthocyanin | Delphinidin 3-O-Beta-D-sambubioside | C_{25}H_{26}O_{13} | 597.4989 | 597 | 303; 465; 229 | 229; 165 | 201; 172 |
| 34 | SPb 1043-11; SPb 1043-8; | Anthocyanin | Peonidin 3-O-rutinoside | C_{25}H_{26}O_{14} | 609.5526 | 609 | 301; 463 | 286 | 258 |
| 35 | 863; 860; SPb 1043-11; SPb 1043-8 | Anthocyanin | Cyanidin 3,5-O-diglucoside | C_{27}H_{30}O_{14} | 611.5335 | 611 | 287; 449 | 287; 213 | 185 |
| #  | Code          | Compound                          | Molecular Formula | Experimental Molecular Mass | Calculated Molecular Mass | Error (%) |
|----|---------------|-----------------------------------|-------------------|-----------------------------|----------------------------|-----------|
| 36 | SPb 1043-11   | Anthocyanin                       | C_{15}H_{15}O_{13} | 274.2827                    | 274.2827                   | 0.0000    |
| 37 | 863; SPb 1043-11 | Hydroxybenzoic acid (Phenolic acid) | C_{15}H_{15}O_{13} | 274.2827                    | 274.2827                   | 0.0000    |
| 38 | 863; 860      | Hydroxybenzoic acid (Phenolic acid) | C_{15}H_{15}O_{13} | 274.2827                    | 274.2827                   | 0.0000    |
| 39 | 863; SPb 1043-11 | Hydroxybenzoic acid (Phenolic acid) | C_{15}H_{15}O_{13} | 274.2827                    | 274.2827                   | 0.0000    |
| 40 | 863; SPb 1043-11 | Hydroxybenzoic acid (Phenolic acid) | C_{15}H_{15}O_{13} | 274.2827                    | 274.2827                   | 0.0000    |
| 41 | 863; SPb 1043-11 | Hydroxybenzoic acid (Phenolic acid) | C_{15}H_{15}O_{13} | 274.2827                    | 274.2827                   | 0.0000    |
| 42 | 863; SPb 1043-11 | Hydroxybenzoic acid (Phenolic acid) | C_{15}H_{15}O_{13} | 274.2827                    | 274.2827                   | 0.0000    |
| 43 | 863; SPb 1043-11 | Hydroxybenzoic acid (Phenolic acid) | C_{15}H_{15}O_{13} | 274.2827                    | 274.2827                   | 0.0000    |
| 44 | 863; SPb 1043-11 | Hydroxybenzoic acid (Phenolic acid) | C_{15}H_{15}O_{13} | 274.2827                    | 274.2827                   | 0.0000    |
| 45 | 863; SPb 1043-11 | Hydroxybenzoic acid (Phenolic acid) | C_{15}H_{15}O_{13} | 274.2827                    | 274.2827                   | 0.0000    |
| 46 | 863; SPb 1043-11 | Hydroxybenzoic acid (Phenolic acid) | C_{15}H_{15}O_{13} | 274.2827                    | 274.2827                   | 0.0000    |
| 47 | 863; SPb 1043-11 | Hydroxybenzoic acid (Phenolic acid) | C_{15}H_{15}O_{13} | 274.2827                    | 274.2827                   | 0.0000    |
| 48 | 863; SPb 1043-11 | Hydroxybenzoic acid (Phenolic acid) | C_{15}H_{15}O_{13} | 274.2827                    | 274.2827                   | 0.0000    |
| 49 | 863; SPb 1043-11 | Hydroxybenzoic acid (Phenolic acid) | C_{15}H_{15}O_{13} | 274.2827                    | 274.2827                   | 0.0000    |
| 50 | 860; SPb 1043-11 | Hydroxybenzoic acid (Phenolic acid) | C_{15}H_{15}O_{13} | 274.2827                    | 274.2827                   | 0.0000    |
| 51 | 863; SPb 1043-11 | Hydroxybenzoic acid (Phenolic acid) | C_{15}H_{15}O_{13} | 274.2827                    | 274.2827                   | 0.0000    |
| 52 | 860; SPb 1043-11 | Hydroxybenzoic acid (Phenolic acid) | C_{15}H_{15}O_{13} | 274.2827                    | 274.2827                   | 0.0000    |
| SPb 1043-11 | Oligomeric proanthocyanidins | (Epi)Catechin-A-(epi)gallocatechin | C_{30}H_{24}O_{11} | 560.5050 | 561 | 399; 278; 201 | 325; 255; 191; 132 |
| SPb 1043-11 | Oligomeric proanthocyanidins | (Epi)catechin-(4,8'/2,6')-(epi)catechin | | 576 | 577 | 559; 447; 377; 396; 265; 179 | 247; 121 175 |
| SPb 1043-11 | Oligomeric proanthocyanidin | 3-O-Galloyl (epi)catechin-(4,8)-(epi)gallocatechin | | 746 | 748 | 575; 466; 377; 306 | 265; 179 247; 121 175 |
| SPb 1043-11; SPb 1043-8 | Aryl-beta-glycoside | Arbutin | C_{12}H_{16}O_{7} | 272.2512 | 273 | 272; 255; 201 |
| SPb 1043-8 | Non-proteinogenic L-alpha-amino acid | L-Pyroglutamic acid | [L-Pidolic acid; 5-Oxo-L-Proline] | C_{5}H_{7}NO_{3} | 129.1140 | 130 | 111 |
| SPb 1043-11 | Amino acid | Leucine | [(S)-2-Amino-Methylpentanoic acid] | C_{6}H_{13}NO_{2} | 131.1729 | 132 | 130 112 |
| 860; SPb 1043-8 | Amino acid | Phenylalanine | [L-Phenylalanine] | C_{9}H_{11}NO_{2} | 165.1891 | 166 | 120 |
| SPb 1043-11; SPb 1043-8; 860 | Cyclohexenecarboxylic acid | Shikimic acid | [L-Schikimic acid] | C_{7}H_{10}O_{5} | 174.1513 | 175 | 128 111 |
| 863 | Amino acid | Tyrosine | [(2S)-2-Amino-3-(4-Hydroxyphenyl)Propanoic acid] | C_{9}H_{11}NO_{3} | 181.1885 | 179 | 133 115 |
| SPb 1043-8 | Propenyl | Methyl eugenol | | C_{11}H_{14}O_{2} | 178.2277 | 179 | 151 123 |
| 863; 860; SPb 1043-8 | Dicarboxylic acid | Azelaic acid | [Nonanedioic acid; Anchoic acid; Finacea] | C_{9}H_{16}O_{4} | 188.2209 | 189 | 171 139 111 |
| 863; 860; SPb 1043-8 | Tricarboxylic acid | Citric acid | [Anhydrous; Citrate] | C_{6}H_{8}O_{7} | 192.1235 | 191 | 111; 173 |
| SPb 1043-8 | Polyhydroxycarboxylic acid | Quinic acid | | C_{6}H_{12}O_{7} | 192.1666 | 191 | 111; 173 111 |
| 863 | Propanoic acid | Dihydroferulic acid | | C_{6}H_{12}O_{4} | 196.1999 | 197 | 127 |
| 863; 860; SPb 1043-8 | Carboxylic acid | Myristoleic acid | [Cis-9-Tetradecenoic acid] | C_{14}H_{26}O_{2} | 226.3550 | 227 | 209; 165 121 |
| SPb 1043-11; SPb 1043-8 | Polypeptide | S-Oxo-L-propyl-L-isoleucine | | C_{9}H_{14}N_{1}O_{4} | 242.2716 | 243 | 196; 137 151 |
| 863 | Medium-chain fatty acid | Hydroxy docosanoic acid | | C_{32}H_{52}O_{4} | 246.3001 | 247 | 229 187 |
| 70 | 863 | Terpenoid trilactone | Bilobalide [(-)-Bilobalide] | $\text{C}_{15}\text{H}_{18}\text{O}_8$ | 326.2986 | 325 | 183; 119; 160 |
| 71 | 860 | Iridoid | Monotropein | $\text{C}_{16}\text{H}_{21}\text{O}_9$ | 390.3393 | 391 | 219; 372; 202; 148; 139 |
| 72 | 863 | Phytosterol | Ergosterol [Provitamin D2; Ergosterol] | $\text{C}_{28}\text{H}_{41}\text{O}_9$ | 396.6484 | 397 | 379; 291; 296; 291; 223; 329 |
| 73 | 860 | Sterol | Fucosterol [Fucoster; Trans-24-Ethylidenecholesterol] | $\text{C}_{29}\text{H}_{43}\text{O}_9$ | 412.6908 | 413 | 395; 324; 219; 329 |
| 74 | 863; SPb 1043-11 | Sterol | Beta-Sitosterin [Beta-Sitosterol] | $\text{C}_{28}\text{H}_{41}\text{O}_9$ | 414.7067 | 415 | 216; 312; 159; 115 |
| 75 | SPb 1043-11 | Anabolic steroid | Vebonol | $\text{C}_{19}\text{H}_{23}\text{O}_5$ | 452.6686 | 453 | 435; 336; 226; 209; 139 |
| 76 | 860 | Triterpenoid | Betunolic acid | $\text{C}_{30}\text{H}_{45}\text{O}_5$ | 454.6844 | 455 | 437; 345; 247; 326; 283; 303; 239; 199 |
| 77 | 863 | Triterpenic acid | Oleanolic acid | $\text{C}_{28}\text{H}_{43}\text{O}_5$ | 456.7003 | 457 | 425; 295; 225; 167 |
| 78 | 863; SPb 1043-11 | Thromboxane receptor antagonist | Vapiprost | $\text{C}_{28}\text{H}_{39}\text{NO}_4$ | 477.6350 | 478 | 337; 263; 121; 119 |
| 79 | 863; SPb 1043-11 | Indole sesquiterpene alkaloid | Sespendole | $\text{C}_{28}\text{H}_{39}\text{NO}_4$ | 519.7147 | 520 | 184; 125 |
| 80 | 863 | Iridoid glucoside | $p$-Coumaroyl monotropein | $\text{C}_{28}\text{H}_{39}\text{O}_{13}$ | 536.4820 | 537 | 375; 256; 185 |
| 81 | 863; SPb 1043-11 | Iridoid | $p$-Coumaroyl monotropein hexoside | $\text{C}_{28}\text{H}_{39}\text{O}_{13}$ | 698.8810 | 699 | 537; 347; 259; 375; 259; 185 |
| 82 | SPb 1043-11 | Steroidal alkaloid | Alpha-chaconine | $\text{C}_{45}\text{H}_{73}\text{NO}_{14}$ | 852.0594 | 852 | 706; 560; 398; 398; 204 |
The CID-spectrum (collision induced dissociation spectrum) in positive ion modes of Dihydrokaempferol from extracts of Lonicera caerulea L. (variety SPb 1043-8) is shown in Fig. 5. The [M + H]^+ ion produced three fragment ions at m/z 270.99, m/z 193.01, m/z 127.03 (Fig. 5). It was identified in the bibliography in extracts from Potato [Oertel et al., 2017]; F. glaucescens [Hamed et al., 2020]; Echinops [Seukep et al., 2020]; Rhodiola rosea [Lee et al., 2016]; Rhodiola crenulata [Daikonya et al., 2011].

![Fig.5. CID-spectrum of dihydrokaempferol from extracts of Lonicera caerulea L. (variety SPb 1043-8), m/z 289.98.](image)

The CID-spectrum in positive ion modes of Dihydrokaempferol from extracts of Lonicera caerulea L. (variety Wild Lonicera from Amur river) is shown in Fig. 6. The [M + H]^+ ion produced one fragment ion at m/z 448.92 (Fig. 6). The fragment ion with m/z 448.92 yields three daughter ions at m/z 376.96, m/z 344.93, and m/z 286.95. The fragment ion with m/z 376.96 yields two daughter ions at m/z 344.92, and m/z 286.99. It was identified in the bibliography in extracts from Rubus ulmifolius [da Silva et al., 2019]; Vitis vinifera [Goufo et al., 2020]

![Fig.6. CID-spectrum of Taxifolin 3-O-glucoside from extracts of Lonicera caerulea L. (variety Wild Lonicera from Amur river), m/z 466.92.](image)

4. Conclusions

Blue-berried honeysuckle Lonicera caerulea L. contains a large number of polyphenolic compounds and other biologically active substances. In this work, we first tried to conduct a comparative metabolomic study of biologically active substances of wild Blue-berried honeysuckle obtained from locations in Khabarovsk territory and from the collection of N.I. Vavilov All-Russian Institute of Plant Genetic Resources (St.-Petersburg). HPLC in combination with a BRUKER DALTONIKS ion trap (tandem mass spectrometry) was used to identify target analytes in extracts.

The results showed the presence of 82 biologically active compounds corresponding to the Blue-berried honeysuckle Lonicera caerulea species. In addition to the reported metabolites, a number of metabolites were newly annotated in blue-berried honeysuckle. There were flavonols: Dihydrokaempferol, Rhamnetin I, Rhamnetin II, Taxifolin-3-O-glucoside, Mearnsin-hexoside, Horridin; flavones: Chrysosierol, Apigenin-O-pentoside, Chrysosierol-7-O-glucoside; flavanone Naringenin; flavan-3-ols: Catechin, Epicatechin, Biochanin A-7-O-glucoside; essential amino acids: L-Prolyglutamic acid, Tyrosine; polypeptide 5-Oxo-L-propyl-L-isoleucine; sterols: Ergosterol, Fucosterol, Beta-Sitosterin; triterpenoids: Betunolic acid, Oleanolic acid; anabolic steroid Vebonol, indole sesquiterpene alkaloid Sespendede; iridoids: Monotropein, p-Coumaroyl monotropein, p-Coumaroyl monotropein hexoside; Myristoleic acid, etc.

The findings may support future research into the production of various pharmaceutical and dietary supplements containing blue-berried honeysuckle Lonicera caerulea L. extracts. A wide variety of biologically active compounds opens up rich opportunities for the creation of new drugs and biologically active additives based on extracts from this family Caprifoliaceae.

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