and held constant for 15 min. Main substances were determined by comparison with database mass spectra of compounds or analyzing ions characteristics of mass spectra.

Results and discussion. Terpene profiles of Santhica and Fedora were analyzed by GC-MS after hydrodistillation. Terpene profiles and abundant of terpenes were slightly different depending on a breed. Most abundant terpenes according to chromatograms were those that took up the widest areas of the peaks in chromatograms. In Santhica most abundant terpenes were sesquiterpenes Shyobunol (34,09%), Aromandendrene (26,57%), trans-α-Bergamotene (11,36%), cis-α-Bergamotene (1,98%) and monoterpene Isogeranial (11,14%). In Fedora most abundant terpenes were sesquiterpenes β-Elemene (24,38%), trans-α-Bergamotene (16,21%), β-Selinene (8,17%), trans-beta-Farnesene (6,78%) and monoterpene trans-Verbenol (16,30%).

Conclusion. We compared two different breeds of Cannabis from Lithuania and determined their terpene compositions. In Santhica and Fedora most dominating terpenes are different, however, trans-α-Bergamotene is common for both. In the Santhica breed dominating terpene profile was Shyobunol, Aromandendrene, trans-α-Bergamotene, cis-α-Bergamotene and Isogeranial. For Fedora the major terpenes were β-Elemene, trans-α-Bergamotene, β-Selinene, trans-beta-Farnesene, trans-Verbenol.

OPTIMISATION OF PRETREATMENT AND DERIVATIZATION METHOD FOR ANALYSIS OF ORGANIC ACIDS BY GAS CHROMATOGRAPHY-MASS SPECTROMETRY

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Introduction. Chromatography is the preferred method of analysis because it adequately addresses the simultaneous identification and quantification of targeted compounds. However, not all chromatographic protocols are suitable for the given task. Although the separation using this method generally targets volatile, non-polar species, the use of derivatization for polar low molecular weight species enables detection with a good resolution and sensitivity. Derivatization as and pretreatment can improve chromatographic results.

The **aim** of our work was to develop reliable and accurate method for quantitative and qualitative analysis of organic acid, such as levulic, heptanoic, malic, lactic, glycolic, oxalic, nonanoic, maleic, succinic , stearic, citric.

Materials and methods. The reaserch was done using methodology on SHIMADZU GC-MS-QP2010 Ultra chromatography system with RXI-5ms (Restek Corporation) capillary column (30 m long, with 0.25 mm outer diameter and 0.25 μm liquid-stationary phase thickness) with a liquid stationary phase (5% diphenyl and 95% polysiloxane), as carrier gas of chromatography we used helium. Organics acids were identified by comparison with database mass spectra of compounds or analyzing ions characteristic of mass spectra. The oven temperature was programmed from 75 °C for 5 min, then 10 °C/min to 290 °C for 5 min, after 20 °C/min to 320 and held constant for 5 min. The injector temperature was 260 °C, injection volume 1 μL, injection mode split, split ratio 1:10, the ion source voltage 70eV. Mass spectra scan range of m/z 35-500 amu with mass scan time 0.2 seconds, interface temperature 280 °C. All solvents were HPLC grade.

Results and discussion. In presence work we examined an effects of different derivative agents, solvents for sample dissolution and reaction solvents. The weights of samples were placed into 5 ml volumetric flasks, 3 ml (of the following solvents: water, acetonitrile, methanol, 80% methanol and 80% acetonitrile) were added and ultrasonicated for 5 minutes, then the volume was filled up to mark. 1 mL of test solutions were evaporated to dryness with a gentle stream of nitrogen, after that 100 μL of acetonitrile (methanol) and 100 μL of N,O-Bis (trimethylsilyl) trifluoroacetamide (BSTFA) or N-tert-Butyldimethylsilyl-N-methyltrifluoroacetamide (MTBSTFA) were added. The derivatization time 4 hours and temperature 70 °C were selected.

In suggested conditions the investigated organic acids have the following retentions times (as shown at the Figure 1): Levulic acid (12,39 min), Heptanoic acid (12,85 min), Malic acid (13,05 min), Lactic acid (14,28 min), Glycolic acid (14,51), Oxalic acid (15,06 min), Nonanoic acid (15,53), Maleic (2-Butenedioic) acid (17,49 min), Succinic (Butanedioic) acid (17,63), Stearic acid (24,72), Citric acid (25,81).

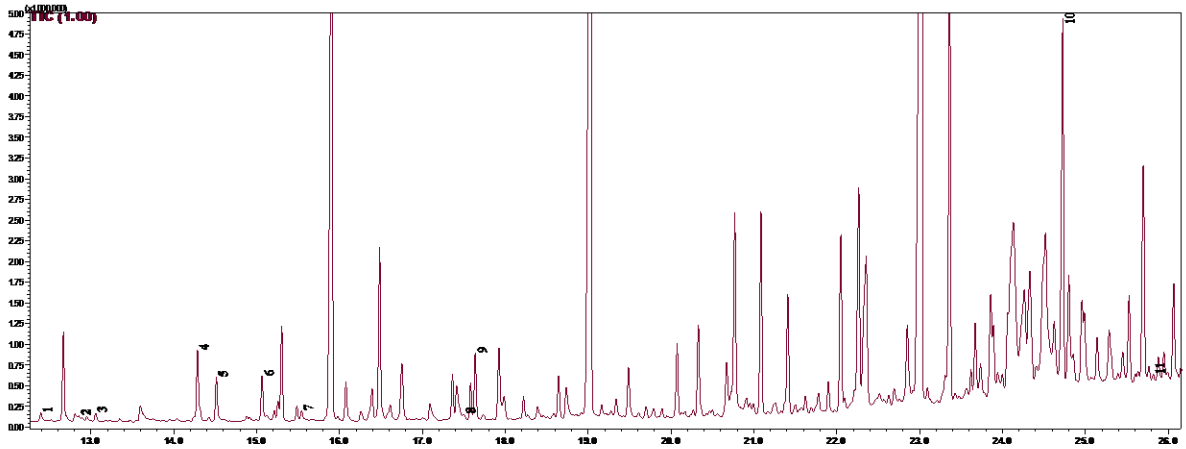

Fig. 1 Typical chromatogram of mitochondria sample

Conclusion. According to received data, the most optimal derivatization conditions were as the following: as derivatization agent was MTBSTFA, as the solutions for dissolution and reaction – water and acetonitrile, respectively. We detected the most effective chromatographic conditions for analysis of organic acids, the results suggest that the method could be usefully integrated in analysis of organic acid.

DEVELOPMENT AND VALIDATION OF HPLC METHOD FOR DETERMINATION OF BIOLOGICAL ACTIVE SUBSTANCES OF *CROCUS SATIVUS* L.

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Introduction. *Crocus sativus* L. is a parennial plant of Iridaceae family. Its usege for medical purpose is good known from ancient time. It contains many biological active substances (such as phenolic acids, flavonoids, isoflavonoids,). *Crocus sativus* L. is used for treatment inflammatory, depressive, cardiac, hepatic, diabetic and other diseases. Besides, earlier studies reported that *Crocus sativus* L. was safe at various doses and did not show any severe side effects. Different growing conditions like humidity, amount of sunlight or soil composition in various geographic areas can effect impact for plant qualitative and quantitative consist. Therefore it is important to have more data from different locations more often in order to have comprehensive knowledge about main components of plant. Stigma is main part of this herb, it is most widely exploitable, although, leaves, stems, tepals or corms also could be quite good source of some bioactive compounds. Thus would reduce plant material wasting and have added value.

Aim of our work was determination and comparing of bioactive components that contains in raw material of *Crocus sativus* L., collected from different locations.

Matherial and methods. We used high performance liquid chromatography (HPLC) for the qualitative and quantitative analysis. Method was carried out by using a C18 reversed-phase column (250*4.6 mm with particle size 5µm) with temperature 25°C. The binary solvent system of the mobile phases were used, 0,1 % acetic acid in water as mobile phase A and acetonitrile as a mobile phase B at the flow rate 1.0 ml/min with following gradient program: 0-8 min, 5-15% B; 8-30 min, 15-20% B; 30-48 min, 40-50% B; 58-65 min, 50% B; 65-66 min, 50-95% B. Extraction by methanol was selected as optimal solvent for raw material. We analyzed different parts of *C.sativus* L. such as pistils, tepals, stems