

Level: Specialist			
Course title: Advanced course in chromatographic methods in biochemistry			
Status: elective			
ECTS: 5			
Requirements: none			
Learning objectives To introduce students to modern techniques of biological samples preparation, chromatographic and spectrometric analysis of primary and secondary biomolecules, data analysis and interpretation.			
Learning outcomes Students should be able to independently apply modern gas and liquid chromatographic methods in analysis of biomolecules in a wide range of biological materials of plant, animal and human origin.			
Syllabus <i>Theoretical instruction</i> Sample preparation of biological materials for chromatographic analysis – tissue lysis, quenching, isolation techniques (Soxhlet extraction, ultrasound extraction, SFE, ASE, MWE, headspace, SPME, SBSE), enzymatic hydrolysis, chromatographic purification methods. Gas chromatography (GC) – principles and instrumentation. Spectrometric GC detectors: EI MS, CI MS, IR. Spectral characteristics of selected compound classes. GC in analysis of primary and secondary biomolecules (vitamins, steroids, essential oils, S and N compounds). Derivatization in GC – analysis of primary biomolecules (glycerolipids, carbohydrates). Multidimensional GC (GC×GC). Liquid chromatography (LC) – principles, instrumentation. Retention mechanisms in LC: RP, NP, HILIC, ion-exchange, affinity, gel, ion-pairing, micellar, chiral chromatography. Chemometrics in chromatography optimization. LC analysis of proteins, carbohydrates, vitamins, lipids, secondary biomolecules. Pre- and post-column derivatization in LC – amino acids analysis. Spectrometric detectors in LC: UV/VIS, FLD, API-MS, NMR. Spectral characteristics of common compound classes, primary and secondary biomolecules. Tandem mass spectrometry: QQQ, Q-TOF, ion-trap. Electrospray ionization of lipids, argmentation. Protein-ligand interactions investigation by API-MS. High-resolution MS in biochemistry – protein sequencing. Isotopic markers in metabolic pathways investigation. Chemometric methods in data analysis – PCA. Chemosystematics. Metabolic profiling, metabolomics, proteomics, lipidomics, genomics, volatomics, fluxomics. Modern electrophoresis methods in biochemistry. Capillary electrophoresis (CE). Miniaturization – bioanalyzer. Modern preparative chromatographic methods in biochemistry – preparative HPLC, centrifugal partition chromatography (CPC). <i>Practical instruction</i> Preparation and qualitative, semi-quantitative and quantitative GC-MS and LC-MS-MS analysis of the selected biological materials of plant, animal and human origin.			
Weekly teaching load			Other:
Lectures: 2	Exercises:	Other forms of teaching: 2	
Student research:			