

RESEARCH – DEVELOPMENT REPORT

Project title: Improvement of methods and techniques for identification and determination at nanoscale of steroid structures contaminants in food

Stage II

Stage duration: December 2008 – November 2009

The objectives of the stage:

- Qualitative determination, based on retention times, the data of NIST libraries of compounds or ionic fragments of contaminants with steroid structure (estrone, beta-estradiol, cholesterol) in SIM or TIC mode;
- Application of derivatization in two stages (oximation and silylation) for separation with significant resolution of contaminants hormone;
- The quantitative determination based on calibration curves with wide dynamic range using a multicomponent stock solution of contaminant compounds with steroid structure, organochloride pesticides and polychlorinated biphenyls in different matrices;
- Application of SPE-GC-MS/MS experimental methodology, and SPE-GC-MS for the extraction, concentration, separation, identification, quantification of contaminants with steroid structure in real samples with different matrices;
- Application of special methods of extraction, purification and concentration on polymer fiber (SPME) of steroids compounds in liquid samples (wastewater, surface, blood serum);
- Identification and determination by multiresidual analysis using liquid-liquid extraction methods, of organochloride pesticides and polychlorinated biphenyls in various food products (milk, grapes) by gas chromatographic techniques coupled with electron capture detection (GC-ECD);
- Detection and quantification by mass spectrometry coupled with advanced analytical techniques (gas chromatography, high performance liquid chromatography): GC-MS, GC-MS/MS (with ion trap), LC-MS/MS, of different steroids at the nanoscale level, in order to determination of contaminants in complex matrices structures

Activities involved:

A.2.1 Official trip to Eötvös Loránd University, Budapest

During 29.08-05.09.2009 Romanian team, consisting of chem. Lacrimioara Senila, chem. Mirela Miclean and chem. Marin Senila, visited the Institute of Chemistry of Eötvös Loránd University, Budapest. On this occasion, together with partner team members (Professor Gyula Záray, Dr. Viktor Gábor Mihucz, Dr. Anikó Vasanits-Zsigrai, Dr. András Helenkár) conducted a qualitative determination of:

- chemical compounds with steroid structure: ► estrone, ► β -estradiol, ► cholesterol in samples with aqueous matrix, by the presentation of fragmentation modes of complete derivatized species of steroid compounds and their derivatives trimethylsilyl ether / ester (oxime);
- qualitative determination of compounds with steroid structures: cholic acids (cholic acid, acid lithocholic, chenodeoxycholic acid, ursodeoxycholic acid, 3-hydroxy 7-ketocolanic acid, dehydrocholic acid), from aqueous samples by derivatized in two stages, then detection by gas chromatography coupled with ion trap mass spectrometry (GC-MS/MS);
- qualitative determination of organochlorinated pesticides (POC) and polychlorinated biphenyls (PCBs) in food samples (milk and grape);

The investigation methods are: solid phase extraction (SPE) and liquid-liquid extraction (LLE). SPE method is a very recent method involving derivatization in two stages: oximation and silylation, gas chromatography coupled with detection by mass spectrometry with ion trap (GS/MS/MS), allowing residual multicomponent analysis by a single injection of the sample.

Qualitative determination of organochlorinated pesticides (POC) and polychlorinated biphenyls (PCBs) by injecting pure standards and real samples under the same chromatographic conditions and comparing the retention times of analytes from the pure standards with those from sample separated by chromatography column.

A.2.2 Receiving visit from the Eötvös Loránd University, Budapest

In 2009, ICIA Cluj-Napoca received visits of the Hungarian partner team, from Eötvös Loránd University - Budapest. On these occasions a quantitative determination of: organochlorinated pesticides (OCPs) and polychlorinated biphenyls (PCBs) in food samples (milk and grape) was conducted. Members of the two partner teams have visited Lech Lacto SRL, in Lechinta, Bistrița-Năsăud County, where they studied the processing

of milk and milk derivatives, and collected milk (raw and pasteurized) samples for OCPs and PCBs determinations.

A.2.3 Activities foreseen in the research program of the young researcher

In the research program of the young researcher chem. Lacrimioara Senila the methodologies for sample preparation prior to chromatographic determination were developed, by performing the purification and concentration using:

- ▶ on solid phase extraction (SPE),
- ▶ solid phase microextraction (SPME),
- ▶ liquid-liquid extraction,

and their application in different analysis for determination of the structure of compounds contaminated with steroid (estrone, estradiol) and organochlorurate pesticides and polychlorinated biphenyls.