

### ***Influence of the route of administration of new psychoactive substances on their metabolic profile***

European Monitoring Centre for Drugs and Drug Addiction has already received over 790 notifications of new psychoactive substances (NPS). These substances are freely distributed on European market as "legal" replacement of drugs. Dealers of NPS circumvent legislative restrictions by slight modification of drug molecule. This manipulation leads to a compound with a different structure, which still possesses psychoactive properties, but – what should be stressed – with unknown toxicity and influence on human health and life.

Increasing numbers of NPS overdose deaths became one of the most current social problems and public health threats in the EU. Common diagnostic tests, designed for classical drugs, turned out to be not very useful in the detection of NPS intoxications. Misidentification can lead to the improper diagnosis and incorrect treatment or wrong cause of death determination.

Additional challenge for forensic and clinical toxicologists is the identification of the NPS metabolites excreted with the most common biological material for drug testing – urine. In this kind of specimen metabolites of NPS can be present at much higher concentrations than their parent compound. Metabolic processes may be also altered by different usage profile i.e. various routes of administration of a single NPS (oral, nasal, smoking, inhalation) or poly-drug use. Although a liver is the major organ of drug metabolism, drugs metabolizing enzymes are found also in other tissues like lungs, brain, kidney, skin, intestine. Some enzymes occur selectively or even exclusively in these tissues.

Therefore, the main goal of this project is to study *in vitro* hepatic and extrahepatic metabolism, to propose metabolic pathways of the most recent NPS for which poly-route of administration was reported and compare these pathways depend on the human tissue preparation in which the biotransformation takes place. We suppose that the route of administration of tested NPS may influence their metabolic profile. A thorough understanding of NPS metabolites structures – performed in laboratory conditions – might contribute to the right identification of substance causing intoxication and a proper toxicological risk assessment.

The goals of the project will be achieved by:

- Prediction of potential NPS metabolites using computer simulation software.
- *In vitro* incubation of selected NPS with different kinds of human tissue preparation (which possess metabolizing enzymes).
- Metabolic stability and pharmacokinetic parameters assessments; NPS metabolites formation.
- Identification of NPS metabolites by high-resolution mass spectrometry.
- Confirmation of the chemical structure of the metabolites by modern instrumental techniques.
- Study of the influence of poly-drug usage on their metabolism.
- Investigation of metabolic pathways of selected NPS depending on the biotransformation site.

The conducted studies will allow elucidating and describing previously unknown structures and will fill the existing gap between the detection of new substances on the market and their identification in hospitalized people. We believe that characteristic metabolites which can be treated as markers of NPS intoxication can be predicted on the basis of *in vitro* metabolic studies. The results of this project could be included in toxicological drug screening of biological samples in clinical toxicology laboratories and facilitate the detection and identification of psychoactive substances in intoxication cases, which is crucial for public health prevention. If, as a result of the project, differences in metabolism depending on tissue are pointed out, a new approach in NPS metabolic research could be shown and the key to identifying new factors affecting NPS' fates in the body.