Nootropic drugs, also known as cognitive enhancers or smart drugs, improve mental functions, such as memory, attention, concentration and motivation. The current lifestyle contributes to the rise of off-label and non-prescription use of nootropics by the general public. As a consequence, many nootropic substances and numerous new supplements are still being developed and introduced on the market. These substances have become very popular among students, as reports suggest that approx. 7% of US students take these stimulants regularly and even 25% of students have tried them in recent years. Thus the global sale of non-prescription enhancers in 2015 already exceeded one billion dollars and is still increasing. Recently, nootropics have been detected in **sewage, surface- ground-** and **even drinking water** and became a **new class of emerging micropollutants** with potential bioactive behavior towards wildlife. **Methylphenidate** is the most commonly prescribed nootropic drug, mainly for people suffering from ADHD. It is metabolized in human body and **excreted as ritalinic acid**. **Piracetam**, as the first nootropic drug, was created to treat cognitive disorders resulting from aging, dementia, epilepsy, depression, autism and dyslexia. Its popularity as a *smart drug* is rising, especially among students. Their environmental fate and transformation pathways (including biodegradation, photo-degradation and abiotic transformation) have received very little attention in the scientific literature to date. It is worth to emphasize that biodegradation of nootropic drugs and determination of their metabolic pathways is extremely important in context of ***sewage epidemiology***, which can be used to monitor the consumption of illicit substances based on samples from WWTPs. Recently, ritalinic acid has been proposed as a biomarker in *sewage epidemiology* studies to monitor the consumption of methylphenidate among students, however no information regarding its biotransformation was presented. In addition, the **metabolites and products of partial metabolism may be more toxic than the parent compounds**, especially in case of pharmaceuticals. Therefore, understanding the fate of active substances (such as nootropic drugs) in the environment and determining the metabolic pathways as well as stable, persistent metabolites (which may enter the ecosystem and cause potential ecotoxicological risk) is of key importance.

The aim of this project is to determine the microbiological metabolic pathways of nootropic drugs based on piracetam and ritalinic acid (main human metabolite of methylphenidate). The compounds which are stable and do not undergo any transformation in the environment, such as biodegradation, photo-degradation and abiotic transformation, can be used as biomarkers to evaluate the human consumption of abused drugs. Moreover, such biomarkers also allow to monitor the environment and its response to potentially dangerous xenobiotic substances. The research hypothesis of this study assumes that ***piracetam and ritalinic acid undergo biotransformation in the environment to metabolites with lower potential for microbiological degradation.*** Therefore the purpose of this project is to also identify stable bacterial metabolites for piracetam and ritalinic acid, which might be applied as biomarkers in sewage epidemiology. The project is divided into two parts: **determination of metabolic pathway of piracetam and ritalinic acid** and **biodegradation of nootropic drugs in activated sludge**.

For the first time, metabolic pathways and stable metabolites produced during the degradation of piracetam and ritalinic acid will be determined. In order to accomplish this goal, the *resting cells* experiments will be performed, which allow to deactivate the cellular division processes and subsequently focus bacterial metabolism only on degradation of nootropic drugs. The qualitative and semi-quantitative kinetics of metabolites production in experiments with *resting cells* will be determined. Based on chromatographic and spectroscopic methods, the structure of intermediates as subsequently the metabolic pathways of nootropic drugs will be described. Moreover the toxicity of the identified metabolites will be also evaluated.

The biodegradation potential of piracetam and ritalinic acid in activated sludges (ASs) will be determined by means of respirometric and chromatographic methods. This analysis will allow to classify both compounds as readily or not readily biodegradable according to the OECD 301 test. It is significant for the success of the project to possess axenic strains effectively degrading piracetam and ritalinic acid and apply them in the bioaugmentation treatments with ASs to improve the decomposition efficiency. The innovative approach of this project is to monitor the qualitative and quantitative changes within subpopulations of ASs exposed to the presence of nootropic drugs by means of next generation sequencing. Using the state-of-art molecular techniques, the necessary information regarding the subpopulations, which are responsible for the biodegradation of nootropic drugs and the determination of bioaugmentation efficiency will be provided. Mathematical modeling methods will be used to compare the biodegradation efficiency of nootropic drugs at different stages of the experiments with and without bioaugmentation approaches. Finally, the obtained results will be used to correlate the stability and changes within subpopulations of activated sludges with regard to the biodegradation efficiency of tested compounds.

The key aspect is to **determine the stable metabolites of piracetam and ritalinic acid, which will serve as biomarkers in *sewage epidemiology* for the monitoring of nootropic abuse**. This research will address the principles of sustainable development to a wide extent, enabling us to understand and, in the further stages, **eliminate pharmaceutical micropollutants from the environment**.