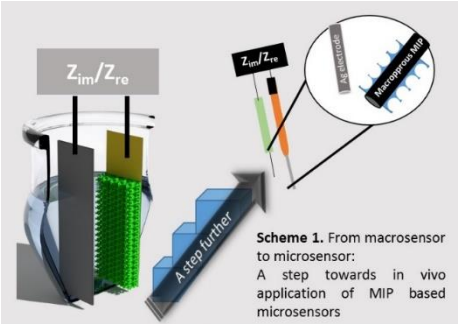
Project goal. Herein, we propose developing **conducting polymer-based molecularly imprinted polymer-modified electrochemical microsensors for different mediators of neuroinflammation**. We plan to design **microsensors for Interleukin 1β, and interleukin 6**. The small physical size of microelectrodes will allow the dimension of the electrochemical cell to reduce, permitting electrochemical measurements to be performed in small volumes (Scheme 1). We will attempt to **implant these microsensors in the mice brain parenchyma**. This will be first step towards measuring MIPs based microsensor performance in detecting neuroinflammation proteins in the living brain. **Such approach is not yet described.**

Description of research. Selective synthetic receptors for neuroinflammation proteins will be prepared following the molecular imprinting approach. This class of polymers, i.e., molecularly imprinted polymers (MIPs), are represented as a synthetic analog to natural antibody-antigen systems. Porosity in such polymers will be introduced by following a hard molding approach to enhance the sensitivity of the final microsensor. Moreover, these molds will provide the surface to immobilize proteins (Interleukin 1β and interleukin 6). These immobilized proteins will be later covalently imprinted in the conducting polymer matrix. In the final step, we will generate macroporous imprinted polymer after silica mold removal. Such a porous structure will have molecular cavities located at the surface of the macroporous polymer for the non-restrictive diffusion of bulky proteins. The recognition of proteins by the macroporous MIP films will be transduced with the help of capacitive impedimetry (CI). Successful compilation of project will result in synthetic receptor-based microsensors suitable to be apply for monitoring neuroinflammation in vivo. This transducer requires low potential for functioning, at such potential interferences from neurotransmitter will be minimum. Microelectrode with robust and selective recognition film will perform detection of protein in the picomolar/femtomolar concentration range the presence of complex matrixes.



Scheme 1. From macrosensor to microsensor: A step towards in vivo application of MIP based microsensors

Reasons for attempting a particular research topic. The development of high-performance in vivo electrochemical biosensors still faces challenges due to the complex biological environments. Therefore, we propose to develop selective electrochemical microsensors. Devising of such microsensor will be beneficial for its application in an in vivo measurement. For instance, ability to monitor the chemical composition of brain interstitial fluid remains an important challenge in bioanalytical chemistry. MIP modified microelectrode can do this task effectively. Moreover, it will permit electrochemical measurements to be performed in drops of undiluted real samples. Additionally, by directly implanting microelectrodes in specific brain regions, in combination with suitable electrochemical output signals, for instance, capacitance, direct measurement of proteins and biomolecule will be possible in short term.

To better understand the role of neuroinflammation in brain diseases, there is a need for efficient tools for monitoring neuroinflammation in vivo. Brain imaging techniques are poorly adapted to monitoring brain cytokines because specific markers for PET or MRI are lacking. The development of high-performance in vivo electrochemical biosensors faces challenges due to the complex biological environments in the brain. The microelectrode proposed to be used in the current project will be small (7-8 µm diameter) and far less than the average distance between blood vessels. This type of measurement will provide local concentrations in specific brain areas, which is more helpful in understanding brain function than global measurements like CSF. CSF sampling is a one-shot technique that does not provide kinetic information on when and how fast cytokines are released in the brain. **Moreover, it is not possible to sample CSF several times in the same animal** unless a long interval of several days.

Substantial results expected. The synthetic receptor based microsensor technology will significantly impact the field of neuroinflammation and neuroscience in general and especially the understanding of neurological diseases in which a neuroinflammatory component is suspected, like Parkinson's or Alzheimer's disease. There is currently no available method for monitoring neuroinflammation in vivo, neither in humans nor animals. If project is successful, **in long term, synthetic receptor (MIP) based-microsensors can be a popular tool in neuroscience**, as they can improve the selectivity and sensitivity issues of microelectrode and provide new knowledge about brain signaling.