Glutamic acid is the main excitatory transmitter in the mammalian central nervous system, and plays an essential role in controlling synaptic plasticity underlying learning and memory processes. This neurotransmitter is responsible for passing information between nerve cells by interacting with specialized proteins called glutamate receptors. The activation of the receptor is evoked by binding between the protein and the glutamic acid molecule, what, consequently, sends the nerve impulse further. Some of the glutamate receptors are associated with ion channels (so-called NMDA, AMPA and kainate receptors) and mediate the flow of positive ions through nerve cells.

Too high concentration of glutamic acid is the main reason for an excessive influx of positive ions into the cell, which can initiate neurotoxic intracellular processes and, as a consequence, neuronal cell death. This type of damage of nerve tissue is observed during many neurological diseases, such as ischemic stroke or epilepsy, and neurodegenerative diseases, such as Parkinson's disease or multiple sclerosis. For this reason, research focused on compounds regulating glutamate ionotropic receptor activity is currently one of the more important and intensively developing directions in the search for new potential drugs for therapies related to neurological and neurodegenerative diseases.

Compounds inhibiting the action of kainate receptors (kainate receptor antagonists) in multiple research demonstrated a broad-spectrum of neuroprotective and anticonvulsant activity, as well as analgesic activity in models of chronic, postoperative pain and migraine. Unfortunately, such compounds often act non-selectively, interacting at the same time with other receptors. A precise understanding of significance and pathophysiological role of kainate receptors depends, therefore, on finding appropriate, highly selective (acting only at one receptor) substances regulating their action. Such compounds are called pharmacological tools.

The ultimate goal of this project is to find appropriate pharmacological tools that selectively block the activity of kainate receptors. Scientific research will be conducted within a series of compounds from one chemical group with a structure based on the quinoxaline-2,3-dione core. The work plan will be realized in the following steps:

Step 1. **Design of compounds** and computer research using modern, highly specialized computer software.

Step 2. **Chemical synthesis** of a series of compounds designed in Step 1.

Step 3. **Pharmacological characterization *in vitro*** of the obtained compounds will be performed in collaboration with scientists from the Department of Drug Design and Pharmacology University of Copenhagen - a leading research center in the field of study on glutamic acid receptors. The tests will include: radioligand binding assays to the native NMDA, AMPA and kainate receptors in rat brain membranes; affinity studies of the most active compounds to selected recombinant rat glutamate receptors. The most active compounds will be additionally characterized *in vitro* in more advanced assays including TEVC and patch-clamp electrophysiology of selected homo- and heteromeric receptors conducted for the most active compounds.

Step 4. **Solubility tests** in water and other solvents (methanol). The condition for effective interaction of a chemical compound with a biological target in a living organism is its good solubility. Unfortunately, a large part of the substances resulting from the research of the pharmaceutical industry do not possess this feature. For this reason, during the design step we have assumed increased solubility of final compounds (which can be obtained through appropriate modifications of the structure). This physicochemical property should be confirmed for real compounds by the experimental method.

Step 5. **Studies on permeability through biological membranes**. These studies will be carried out for a selected group of compounds, intended for further animal testing. Only compounds with an adequate degree of membrane permeability will proceed to the next stage.

Step 6. **Behavioral studies** for 1 or two the most potent and selective compounds selected in Steps 3-5. At this stage, anticonvulsant activity in mice models of seizures will be determined.

Step 7. **Crystallographic research**. As part of the project, attempts will be made to co-crystallize selected compounds in complex with appropriate receptors. Obtaining such crystal structures will allow analysis of protein-antagonist interactions and further effective design of compounds with expected activity.

As a result of the above tests, chemical compounds with the best physicochemical properties, with the highest affinity and selectivity at kainate receptors will be selected as candidates for selective kainate pharmacological tools. The obtained scientific results will be published in international scientific journals and submitted at conferences in the field of medical chemistry.