

## **Involvement of astrocytes in the neuroprotective mechanism evoked by enhancement of noradrenergic transmission in transgenic mouse model of progressive parkinsonism.**

**Parkinson's disease (PD)**, the second most common neurodegenerative disorder, is characterized by progressive, inevitable loss of the **dopaminergic neurons**, directly responsible for the observed clinical manifestation. Currently available pharmacotherapies are based on disease symptomatology and they never restore neuronal function nor prevent neuronal loss. An important factor considerably diminishing the therapeutic efforts is that the neural loss begins long before the first clinical symptoms appear, and even a prompt diagnosis provides very little opportunity for further effective treatment as most of the neurons are already gone. Although historically the hallmark of the disease has been focused on the degeneration of the brain regions called substantia nigra and ventral tegmental area (SN/VTA), recently it become widely accepted that the neurodegeneration occurs in subsequent stages and damage in other brain areas may affect i.e. **noradrenergic neurotransmitter system**, which may be regarded as a prodromal phase of PD.

There is a lot of evidence regarding the integrity of the noradrenergic and dopaminergic systems in the pharmacological and transgenic mice models of PD. These data are concomitant with the hypothesis that **noradrenergic neurodegeneration may be regarded as an early pre-symptomatic phase of PD**. The potency of noradrenergic enhancement as a neuroprotective strategy to compensate (at least to some extent) for dopaminergic cell loss was recently **confirmed in our studies with reboxetine** (a highly selective noradrenergic antidepressant). Namely, in the mice model of progressive PD, reboxetine treated animals revealed slowed down neurodegeneration of SN neurons compared to non-treated littermates.

It is widely known, that our brain consists not only with neurons, but also with **glia**, which – among many functions – are important marker of inflammatory processes. Emerging lines of evidence have shown that features of neuroinflammation and glial reactivity (i.e. changes in the **astrocytes** morphology and function) associate with reduced level of noradrenaline in the brain are distinctly observed during early stages of neurodegeneration. Therefore, the purpose of the project is to **verify strategies proposed to prevent neurodegeneration by astrocytic activation** by applying drugs that elevate or normalize noradrenaline and agonists acting via different adrenergic receptors.

The project will be divided into two parts: *in vitro* screening of noradrenergic agents and *in-vivo* validation of the hypothesis of astrocytes playing pivotal role in the mechanism of noradrenergic neuroprotection. Regarding the first issue, using the *in vitro* reactive astrocyte model, we intend to check whether the selected **noradrenergic compounds** will have a direct **impact on reducing astrocyte reactivity and maintaining their physiological function**, which is expected to diminish the inflammatory transmission. In the second part of the project, to **dissect the role of astrocytes in the putative mechanism of neuroprotective effects observed after reboxetine treatment**, the hypothesis will be evaluated on our mice model of progressive PD, where the animals will be additionally devoid of astrocytes, selectively depleted in the region of SN/VTA. This will be achieved by **CRISPR/Cas9 gene editing**, the powerful and relatively simple methodology, acknowledged in 2015 as a top scientific breakthrough of the year in the life sciences. The CRISPR/Cas9 construct “wiping out” the astrocytes from SN/VTA will be delivered by direct injections to the brain by ultrathin needle on a special microsurgery device (stereotactic table).

We believe that studying this topic is an important aspect that will bring us new knowledge regarding the process of developing PD and the impact of the microenvironment, especially astrocytes on neuronal degeneration. As mentioned earlier, it is suggested that one of the markers of the prodromal phase of PD is reduced level of noradrenaline in the brain. Therefore, it is worth to dissect this mechanism in context of potential neuroprotective therapies that could target the noradrenergic system and delay or even prevent symptoms of PD. What is more, if the hypothesis will become true, there are available already existing drugs targeting noradrenergic system (i.e. antidepressants), that could be effectively used for this purpose.