

NENS Exchange Grant

The Functional Role of Top-Down Inputs onto the Hypothalamus

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Host Lab: ‘Neural Circuits and Behaviors Group’ - Max Plank Institute for Metabolism Research, Cologne, Germany

Host Lab Supervisor: Dr. Tatiana Korotkova

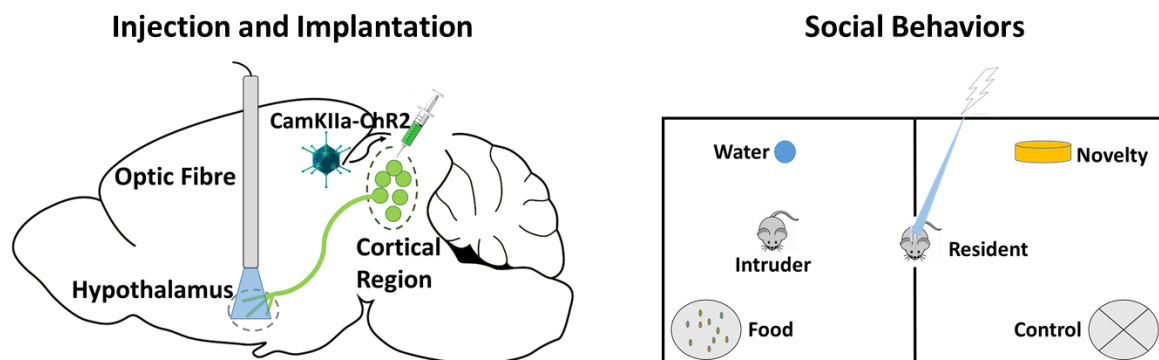
Overview:

The aim of my training stay was to acquire the technical skills necessary for performing optogenetics experiments in behaving mice, by applying them to study the role of the projections from a cortical structure to the hypothalamus. For being able to perform this technique, I learned to handle small rodents (mice), I performed injection of viral vectors for expressing excitatory opsins and implantation of optic fibers for optogenetic stimulation. In addition, I learned how to carry out behavioral experiments with mice and analyze the data obtained in behavioral tests.

The hypothalamus is a brain structure with highly conserved neuronal circuits closely related, both in anatomy and function, to several telencephalic, diencephalic and brainstem regions. It is located in an ideal position to modulate many behavioral functions, including emotion, motivation, feeding, social behaviors and neuroendocrine-related responses (Saper and Lowell, 2014). The hypothalamus constitutes the ventral diencephalon and it is situated between the midbrain and the thalamus. It receives cortical projections from different cortical areas including the hippocampus, subiculum and medial prefrontal cortex and it is bilaterally connected with many subcortical structures, e.g. the lateral septum and ventral tegmental area (Puelles et al., 2012). However, despite its privileged neuroanatomical position for regulating many behaviors, the precise functional significance of different hypothalamic nuclei remains to be determined. In addition, it is unclear the control that the different cortical areas exert over it. In this study, we applied optogenetics to specifically stimulate glutamatergic projections coming from a cortical region to a hypothalamic nucleus in mice performing a social behavior test to understand the influence of this pathway over different innate behaviors.

To study the role of these projections, wild-type C57BL6 mice were injected in a cortical region projecting to the hypothalamus with an adeno-associated virus type 2 under the promoter CaMKIIa to express channelrhodopsin 2, a light-gated ion channel, specifically in

glutamatergic neurons of this region. For the injections, bilateral craniotomies were performed over the cortical structure and the virus was infused by introducing a bevelled metal needle in the desired coordinates, connected via a tube with a microsyringe pump. After 5-6 weeks, when channelrhodopsin was expressed in the axon terminals, I implanted fiber optic cables coupled to a ceramic stick ferrule in the hypothalamus, and I fixed them to the skull using dental cement. After a recovery period, I habituated the mice to a custom two chamber enclosure with food, water, novel object and control designated zones were mice were tested for the social behavior test. In this test, the implanted mice were optogenetically stimulated while they were video-monitored for the analysis of social, feeding and other different behaviors. This way, channelrhodopsin channels in the glutamatergic terminals coming from the cortical region to the hypothalamus were activated, depolarizing the axon terminals and stimulating hypothalamic neurons, producing different effects in behavior that were posteriorly manually scored and analyzed.



Perspectives

My training stay at the “Neural Circuits and Behaviors Group” was a great experience that allowed me to acquire the necessary skills for performing optogenetics in behaving mice and applying it to a specific brain circuit. In the future, I plan to continue my career in the field of systems neuroscience, trying to understand how different brain circuits are involved in different innate behaviors. For this, optogenetics is one of the most useful tools due to the specificity when manipulating neural activity and time precision. It is my intention to continue using this technique during my PhD and combine it with electrophysiology and calcium imaging to monitor neural activity. Therefore, I would like to thank NENS and my host supervisor, Dr. Korotkova, for granting me the opportunity of performing this training stay and successfully acquiring these skills.

References

- Saper CB, Lowell BB (2014) The hypothalamus. *Curr Biol* 24:R111–R116.
- Puelles L, Martinez-de-la-Torre M, Bardet S (2012) Hypothalamus. In: *The Mouse Nervous System* (Watson C, Paxinos G, Puelles L, ed), pp221-312. London: Academic Press.