Campagne 2020 Contrats Doctoraux Instituts/Initiatives

Proposition de Projet de Recherche Doctoral (PRD)

Appel à projet ISVI - Intitiative Sces du vivant ses interfaces 2020

Intitulé du Projet de Recherche Doctoral : How Cerebello-Navigation structures projection(s) process sensory-motor signals required to drive subsequent adapted behaviours?

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Unité de Recherche :
Intitulé : Neuroscience Paris Seine
Code *(ex. UMR xxxx)* : UMR 8246

ED158-Cerveau, cognition, comportem

Ecole Doctorale de rattachement de l'équipe & d'inscription du doctorant :

Doctorants actuellement encadrés par le directeur de thèse (préciser le nombre de doctorants, leur année de 1ere inscription et la quotité d'encadrement) : 1, 2019, 50%

Co-encadrant :

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Unité de Recherche :
Intitulé : Neuroscience Paris Seine
Code *(ex. UMR xxxx)* : UMR 8246

Choisissez un élément :
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Cotutelle internationale : X Non Oui, précisez Pays et Université :

Description du projet de recherche doctoral *(en français ou en anglais)*

3 pages maximum – interligne simple – Ce texte sera diffusé en ligne

Détailler le contexte, l'objectif scientifique, la justification de l'approche scientifique ainsi que l'adéquation à l'initiative/l'Institut.
Le cas échéant, préciser le rôle de chaque encadrant ainsi que les compétences scientifiques apportées. Indiquer les publications/productions des encadrants en lien avec le projet.
Préciser le profil d'étudiant(e) recherché.

How Cerebello-Navigation structures projection(s) process sensory-motor signals required to drive subsequent adapted behaviours?

This project fits with the following I-bio objective:

Adaptation of biological processes, their plasticity and learning mechanisms

Biology, Engineering and informatics Interface

The link between perception, mental representation and accurate behaviour is still a fundamental and unresolved question in Neuroscience. How can we adapt and learn the appropriate behavioural actions during navigation, a reward-directed behaviour? This project aims at characterizing the mechanisms that influence the dynamical link between the cerebellum a dedicated sensory-motor structure and navigation-related structures, namely the hippocampus and the medial septum (MS), two major navigation structures hosting space-coding neurons (i.e. place and grid cells). Importantly, we recently described that the MS is a potential single-relay pathway from the cerebellum to the hippocampus (Watson et al., 2019). Overall, this study will contribute on our understanding of the brain mechanisms that ensure accurate coding of sensory-motor signals required to drive subsequent adapted behaviours. Such a project requires strong skills in electrophysiology in freely-behaving mice, programing with non-linear GLM model development, and biomechanical and electronic engineering.

Introduction

The cerebellum plays a major role in the control and learning of skilled movements at the heart of motor coordination (see review in De Zeeuw and Tenbricke, 2015). During the last decades, however, its role in cognitive function (see review in Rondi-Reig and Burguière, 2005; Buckner, 2013; Schmahmann et al., 2019) and in the control of emotional states (Carta et al., 2019) has been increasingly recognized. This idea is supported by anatomical, physiological and clinical studies which have described its connections with different brain areas engaged in cognition, including cortices such as the prefrontal or the parietal cortex and subcortical ones such as the hippocampus or the medial septum (MS), structures known to be critically involved in spatial memory and navigation (Kelly and Strick, 2003; Gianetti and Molinari, 2002; Rowland et al., 2016; Watson et al., 2019).

Along those lines, we demonstrated that the cerebellum is crucial for hippocampal place cell stability (Rochefort et al., 2011, Lefort et al., 2019), a physiological proxy for spatial memory (O'Keefe and Dostrovsky, 1971), and efficient navigation behaviors, including spatio-temporal navigation (Burguière et al., 2005; Igloi et al., 2015, Babayan et al., 2017). We recently proposed that the cerebellum may monitor multimodal sensory information in particular originating from self-motion (vestibular, proprioceptive, optic flow or motor command efferent copy) and generate an output signal influencing navigation structures to stabilize and/or update the mental representation of space (Rondi-Reig et al., 2014). However, the mechanisms by which such influence is exerted and the consequences on learning and memory remain unknown.

The MS is known to send direct projections to the hippocampus and the medial entorhinal cortex (MEC, part of the hippocampal formation) (Raisman, 1966; Risold and Swanson, 1996), whereas its connections with the cerebellum are still to be defined. It projects to the hippocampal formation through cholinergic, GABAergic and glutamatergic neurons (Amaral and Kurz, 1985; Freund, 1989; Manseau et al., 2005) and these septo-hippocampal formation projections are reported to play an important role in cognitive function (Winson, 1978; Mitchell et al., 1982). Most neurons in MS are theta-rhythmic and are considered to be pacemakers of the hippocampal formation theta oscillations (Lawson and Bland, 1993; Dragoi et al., 1999). In addition to temporal processing, MS also contributes to the animal's spatial and navigation abilities. Inactivation of septal activity impairs self-motion based linear distance estimation (Jacob et al., 2017), similar to the effect induced by MEC lesion; abolishes the distance-specific firing of place cells on a running wheel (Wang et al., 2015); disrupts the formation of grid cells in the MEC, cells part of the brain's metric for representation of space (Brandon et al., 2011).

Theta is a brain rhythm present during ongoing locomotion, anaesthesia and REM sleep. MS, hippocampal and MEC neuronal firing and theta are positively correlated with running speed (King et

al., 1998; Slawinska and Kasicki, 1998; *Jeewajee et al*., *2008*). MS is reported to convey running speed information to hippocampus and MEC (Fuhrmann et al., 2015; Hinman et al., 2016). Interestingly, the slope component of theta frequency to running speed relationship is considered to be linked to distance estimation (McNaughton, 2006; see review in Korotkova et al., 2018). As cerebellum monitors multimodal sensory information originating from self-motion and MS rhythmically integrates processed sensory and motor signals necessary for navigation, we question the role of the cerebellar projections to MS and how they may affect the MS and hippocampal formation neuronal firing and theta rhythm. Behaviourally, we question the role of cerebello-septal projections in self-motion based linear distance estimation.

1- Anatomical characterization of the cerebellar projections to MS

We will perform a neuronal tracing study by stereotaxically injecting an anterograde virus into the deep cerebellar and vestibular nuclei (DCN and VN). After confirming the presence and the origin (i.e.DCN and/or VN) of the direct cerebello-septal projection, we will determine the functional connections between cerebellar fibers and septal neurons, by modulating the cerebellar axons projecting to MS while recording the MS activity, using *in vivo* optogenetic coupled to *in vivo* electrophysiology. To do so, we will stereotaxically inject an adeno-associated viral vector carrying NpHR, a protein able to inhibit neuronal activity under light-stimulation, into the DCN (or VN). Then, we will implant a silicon probe with integrated optical fiber (optrode) in MS to record neuronal firing and local field potential (LFP, necessary to determine theta) while inhibiting cerebellar afferents in MS, during a navigation task enabling to also test the mice ability to estimate distance.

2- Functional characterization of the cerebellar projections to MS during a navigation task that enables to test the ability of mice to estimate distance.

Another neuronal tracing study will take place to precisely identify the region of the MS that support the cerebellar projections to the hippocampal formation via MS. We will stereotaxically co-inject an anterograde virus into DCN (or VN) and a retrograde tracer into the hippocampus or MEC. In addition, we will identify which type of neurons in MS are connected to both, the cerebellum and the hippocampal formation, using immunochemical techniques. Then, we will determine the role of cerebellum on the septo-hippocampal formation projections, by modulating the cerebellar axons projecting to MS using *in vivo* optogenetic while recording the hippocampal and MEC activity using *in vivo* electrophysiology. To do so, we will stereotaxically inject an adeno-associated viral vector carrying NpHR into DCN (or VN) and implant an optical fiber in MS. Then, we will implant a tetrode Microdrive in hippocampus and MEC in order to record simultaneously neuronal firing and LFP while inhibiting cerebellar afferents in MS, during a navigation task enabling to also test the mice ability to estimate distance. The engineering part described thereafter is necessary for this second part of the project.

3- Development of 64-channels microdrives and electronic boards for mice

Unlike primary sensory processing, most of higher brain functions are distributed over multiple brain areas. Neuronal cells are the fundamental building blocks of brain whose firing activity leads to brain functions. To study neuronal underlay of cognitive functions such as space-coding during navigation, not only localized ensemble neuronal recordings but also multi-structure recordings can lead to better understanding of how cognitive functions are processed in the brain. However, the available technologies in this regard, are very costly, have size or weight issue or cannot target multiple brain regions. For example, multi sites silicon probes with a channel count of 32 cost approximately 1000 euro, and it cannot be reused in a live animal experimentation which required a large budget and limits the possibility of pilot experiments. Recently developed miniature 2-photon imaging endoscopes in live animals are advantageous for their reusability and recording from large number of cells within a structure, however due to their size it is impossible to implant multiple of such endoscope in small animals like mice. Plus, recording from deep brain structures requires removal of superficial brain tissues which can cause systemic brain interventions. Yet, a conventionally popular method has been the use of twisted ultra-fine wires and performing wire-tip recordings. Such wires can be twisted

together to generate sterotrodes, tetrodes, or octrodes to facilitate triangulation of neuronal activity for separation of activity of nearby neurons (Steenland & McNaughton, 2015).

Employing this technique, we aim to design inexpensive and rapidly-constructible microdrives with electronic interface boards in the lab to be able to perform both ensemble and structure brain recording in mice. We will design electronic interface boards (EIBs) using state of the art of printed circuit board (PCB) technologies to generate tiny, cheap (~2 euro) and high channel count PCB boards (2x 32channels). Further, we will design and fabricate miniature and cheap (1-3 euro using 3D printers) microdrives with multiple screws which allows us to advance the tetrode wires in the brain to record from different layers of a brain structure in live freely moving animals. The aim is to get tetrode-drive design incorporating a total of 64 channel simultaneous recordings in freely behaving mice (using tetrodes in published articles, highest number of channel count in mouse is 48) and in four separate brain structures (highest number of simultaneously recorded separate brain structures performed in mouse is two). Therefore, we will be able to examine the causal influence of inactivated structure on other targeted structures. With this method, we will not only monitor the simultaneous dynamics of multiple brain structures under a certain behavior, but also reduce the necessity of more animal usage which in turn is in line with the 3Rs principle of animal welfare.

This PhD project will beneficiate from my expertise in complex behavioral development and analysis as well as the knowledge of Julien Fournier in electrophysiology and program development and of Mehdi Fallahnezhad who has a strong bio-engineering background. The student will ideally have a biological background with strong abilities to develop skills in bio-engineering and programing.

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