Final Report for NENS Exchange Grant

Student: Violeta Maria Caragea

Period of training stay: 5 February - 29 April 2018

Host Programme: Donders Master in Cognitive Neuroscience - Radboud University, The Netherlands
Host Lab: Memory Dynamics lab, Department of Neuroinformatics, Donders Institute, Radboud University, The Netherlands
Host Supervisor: Assistant Professor Lisa Genzel

Home Programme: Master in Neurobiology - University of Bucharest, Romania
Home Lab: Research Center in Neurobiology and Molecular Physiology
Home Coordinator: Associate Professor Violeta Ristoiu

Project title of the proposed training stay: *Adapting the Object Space task for mice to explore the impact of G-Rh2 treatment on memory formation*

Overview:

The main purpose of my training stay was to learn how to use the *Object Space Task* (OS) (a multi-trial behavioral task testing memory accumulation in rodents) and find ways to adapt it for my thesis research plan in my home lab.

The OS task was developed in the Memory Dynamics lab with the intention to investigate cumulative memory in rodents (Genzel et al, 2017). The task measures the exploratory behavior of mice and rats while they are in an open field box where two identical objects are displayed in the corners following a predefined pattern. This pattern is established based on three distinct conditions: *stable* - both locations are constant during the training trials and one object is displaced during the test trial; *overlapping* - one location is stable and the other alternates, while the test configuration is the same as in the last training trial; and *random* - there is no spatial pattern of objects placement. The overlapping condition was previously demonstrated to measure the cumulative memory effect both in mice and rats. During my training stay I mainly focused on implementing the standard protocol for mice associated with a catastrophic interference - either extreme novelty or mPFC inhibition via an optogenetic intervention.

The training activities I was involved in during my three months as an exchange trainee in the Memory Dynamics lab were as follows:

1. Handling mice and rats in their first weeks in the animal facility;
2. Habituate the animals with the task specific setting (in the OS box and with the optogenetics experiment installation);
3. Train the animals according to the OS task protocol for various types of experiments: OS baseline with mice, catastrophic interference on memory consolidation with sleep deprivation and novelty conditions with rats and mice, and mPFC inhibition via optogenetics in mice;

4. Statistically analyze data from these experiments;
5. Design a plan for installing the OS task setting in my home lab and for conducting a pilot experiment with Ginsenoside Rh2 injected mice as part of my research thesis.

Moreover, during the stay, I had the chance to participate in rat brain slicing, slice mounting and immunostaining procedures and also to assist to a part of the optogenetics experiment preparation activities. Furthermore, I could also benefit from the rich informal learning activities in the lab, such as the journal club focused on the newest memory research literature, the various lectures held in the Donders Institute, and the presentations delivered by the Neuroinformatics department members, which all broadened up my perspective for future research plans.

After this experience, my short-term follow-up plans consist in:

- Disseminating my experience insights and results with my home lab members (7th of May 2018);
- Helding a presentation about my results at the student conference organized by the Faculty of Biology in my home university (18-19 May 2018);
- Developing the OS task setting in my home lab (planned for starting in September 2018), including the pilot experiment with the Ginsenoside Rh2 injected mice;
- Presenting a poster with an OS experiment results at the annual conference of the National Neuroscience Society of Romania (18-20 October 2018).

Overall, I can say that my training stay at the Memory Dynamics lab was a great learning experience which will probably have a great impact on my career start in cognitive neuroscience research. I am, therefore, grateful for the NENS Grant Stipend for making it happen and also to the two labs coordinators for supporting me.

References:

- Lisa Genzel, Evelien Schut, ..., Francesco Battaglia (2017). The object Space Task for mice and rats, in *bioRxiv 198382*; doi: https://doi.org/10.1101/198382, available online at https://www.biorxiv.org/content/early/2017/10/04/198382;
- Paul W. Frankland & Bruno Bontempi (2005). The organization of recent and remote memories, in *Nature Reviews Neuroscience* volume 6, pages 119–130 (2005) doi:10.1038/nrn1607;
- Larry R. Squire, Lisa Genzel, John T. Wixted, & Richard G. Morris. (2015). Memory Consolidation, in *Cold Spring Harbor Perspectives in Biology* 2015;7:a021766, doi: 10.1101/cshperspect.a021766.

Figure 1. *Object Space task apparatus* (Genzel et al, 2017)
A box with 75cmX75cm white walls and over 40 pairs of objects were used for the task. A special software provided the input for the training trials and offered support for scoring exploration behaviors and for exporting the experimental raw data. The operator was blinded from conditions of trials.

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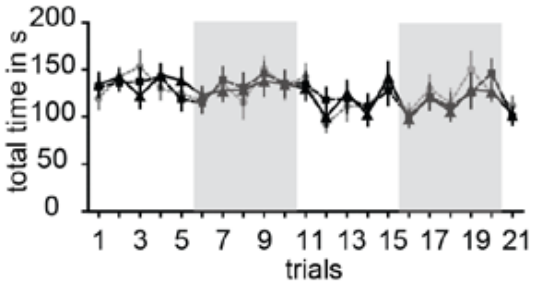

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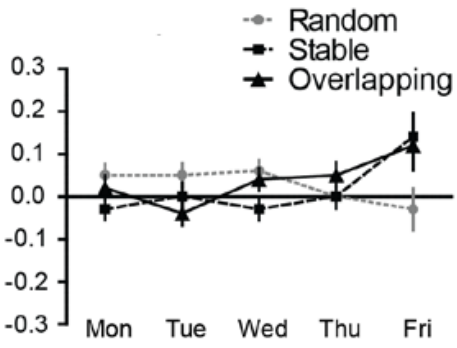

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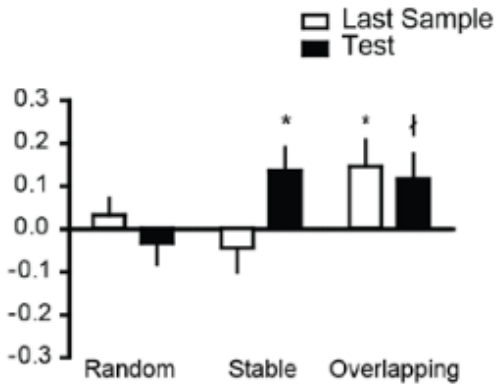

Figure 2. *Object Space Task baseline* (Genzel et al, 2017)

Data were collected from piloting the OS task protocol for mice with 5 training trials per day; N = 15, male C57Bl6/J mice, 7-8 weeks of age at the start of behavioral training (Charles River). **a.** *Exploration time* - no differences in total exploration time was found between conditions (condition $F_{2,28}=0.06$, $p=0.94$; trial $F_{20,280}=4.8$, $p<0.001$)**, b.** *Discrimination Index across days* - significant interaction effect present for condition x day (condition $F_{2,28}=0.2$, $p=0.81$, day $F_{4,56}=1.7$, $p=0.17$; condition x day $F_{8,112}=3.16$, $p=0.003$), **c.** *Discrimination Index test* - memory performance on trial 20 in the overlapping condition is significantly above chance ($t_{14}=2.4$, $p=0.034$) and a trend was observed in trial 21 ($t_{14}=2.0$, $p=0.07$).

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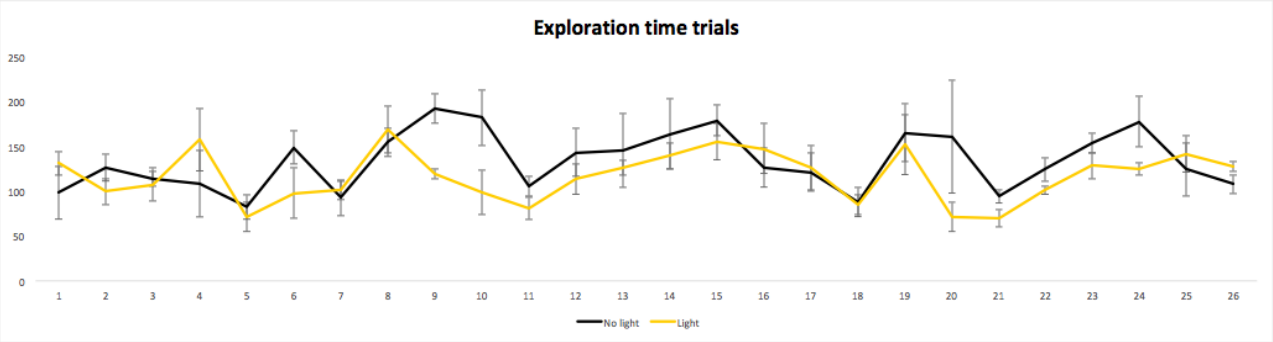

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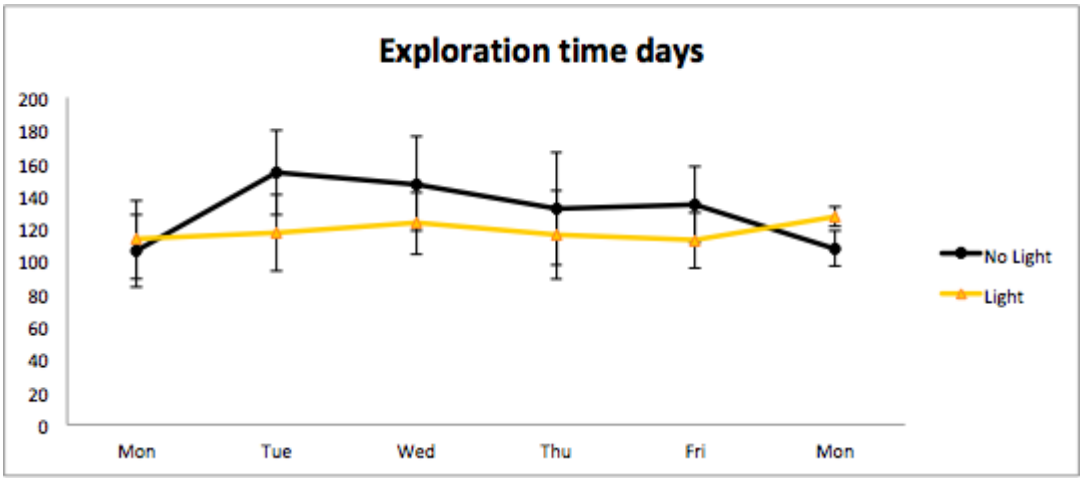

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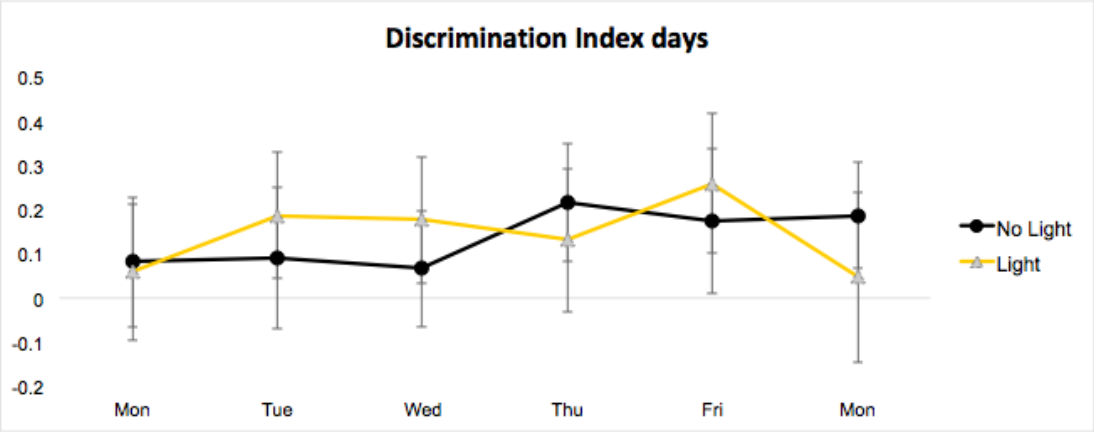

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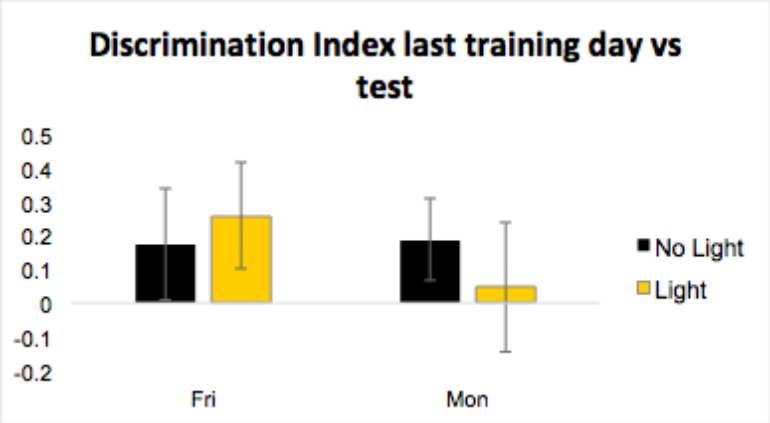

Figure 3. *OS pilot experiment with mPFC inhibition in mice* (preliminary results, work in progress, unpublished) - Data were collected for the overlapping condition for two rounds of the same experiment (N = 4, male C57Bl6/J mice) to identify any relevant statistical trends for the pilot (work in progress) **a.** *Exploration time average trials Light vs No light,* **b.** *Exploration time average days Light vs No light*, **c.** *Discrimination Index average days Light vs No light*, **d.** *Discrimination Index last training day vs test Light vs No light*

week 1	weeks 2-4	weeks 5-7
Handling & Habituation	G-Rh2 ip administration 10 mg/kg (n=16) saline ip admin (n=16)	OS training and test (n=32)

Figure 4. *OS task Ginsenoside Rh2 injected mice pilot experiment plan*