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科学研究費助成事業　　研究成果報告書

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研究課題名（和文）Pharmacological and Optogenetic Dissection of Inferentatial Decision Making

研究課題名（英文）Pharmacological and Optogenetic Dissection of Inferentatial Decision Making

研究代表者
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交付決定額（研究期間全体）：（直接経費）　　1,200,000円

研究成果の概要（和文）：この研究は、世界的なCovid19のパンデミック状況のために大幅に遅れていますが、私は、条件付けされた舐め抑制パラダイム（デバイスの設計と構築、およびタスクのプログラミング）に基づいた高次の条件付け手順を確立しました。私はすでに一次条件付け手順で手順を検証し、実験計画法でマウスの高次条件付けに関する有望な予備データをすでに取得し、それに取り組んでいます。 2021年以内に高次条件付けに関与する神経回路の操作を開始したいと思っています。

研究成果の学術的意義や社会的意義
A disruption in associative learning processes has been associated with psychiatric disorders. First-order conditioning does not reflect real-life situations. To better approximate real-life complex situations, investigating behavioral and neural mechanisms of higher-order conditioning is necessary.

研究成果の概要（英文）：While this research has been significantly delayed due to the worldwide Covid19 pandemic situation, I established a higher-order conditioning procedure based on conditioned lick suppression paradigm from scratch (design and building of the device, and programming of the task). I already validated the procedure in a first-order conditioning procedure and obtained already encouraging preliminary data on higher-order conditioning in mice in my experimental design, and keep working on it. I hope to start manipulating neural circuits involved in higher-order conditioning within 2021.

研究分野：Behavioral Neuroscience

キーワード：Associative learning　Pavlovian conditioning　Optogenetics　Memory engram　Fear conditioning

１．研究開始当初の背景

The simplest form of classical conditioning, where an event is associated with a pleasant or aversive event is well documented. However, real life situations are way more complex and also involve associations between sensory events. Higher-order conditioning extends associative processes to events that have never been physically experienced. In the sensory preconditioning procedure (Brogden, 1939), the presentation of a conditioned stimulus is followed by that of a second conditioned stimulus (CS1 → CS2) in a first phase. In a second phase of training, the CS2 is followed by an unconditioned stimulus, say a foot shock (CS2 → US). When CS1 is tested alone, a conditioned response of freezing is observed, even if it has never been paired with the US. Such phenomenon can be defined as inferential decision making and suggests that associations between events can also be formed mentally.

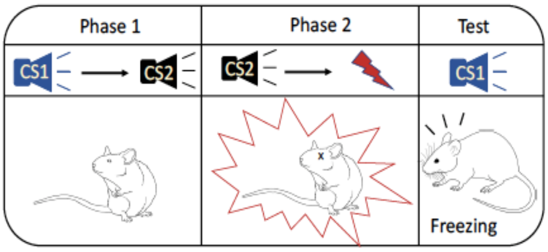


Figure 1. Sensory Preconditioning(SPC)

But so far, no mechanistic explanation has been proposed as to how this switch between SS and SR connectivity occurs. Moreover, the neural mechanism of memories integration is unknown.

２．研究の目的

M y project aims at unraveling the psychological and neural mechanisms of inferential decision making that occur during SPC, using a combination of fine behavioral assessment with cutting edge neuroscientific tools. A disruption in associative learning processes is found in many psychiatric diseases (Schizophrenia: Lubow et al., 1987; Obsessive-Compulsive Disorder: Gillan et al., 2014; Autism-Spectrum Disorder: Kosaki & Watanabe, 2016; Post-Traumatic Stress Disorder: Luo et al., 2018). The relapse of the symptoms is a major limitation suggesting that these therapies are based on an erroneous or at least incomplete model of associative learning and memory.

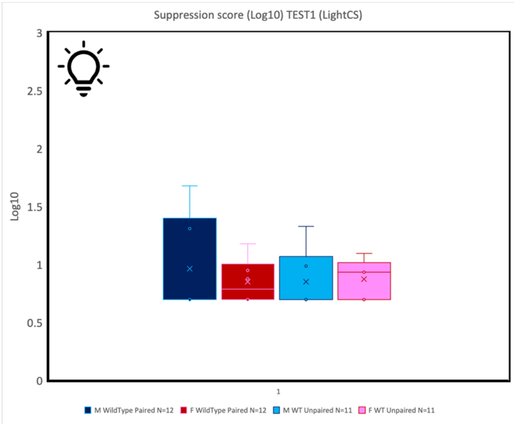
３．研究の方法

I propose a new strategy to dissect the associative structure of memory at the psychological level, while unraveling the underlying neural circuit involved. Previous studies used extinction of a specific association as a behavioral strategy to inhibit a subset of an integrated memory and see whether it is necessary for the CS1 to provoke a conditioned response. But there are numerous studies suggesting that the manipulation of the value of a stimulus (e.g., CS2) previously paired to another stimulus (e.g., CS1), will affect the later (see Miller and Witnauer, 2017 for a review). A particular feature of this project lies on the application of optogenetics to specifically target and neutralize a subset of an integrated memory, without thus involving the collateral effect of stimuli interaction. Another novelty of my approach is the complete reinterpretation of Molet et al. (2012) and Polack et al. (2013)

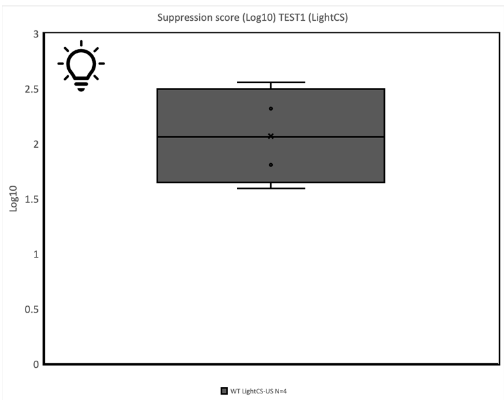
account in a simplified SR account, and the direct test of our model using neural manipulation, which would be impossible to achieve using behavioral assessment alone.

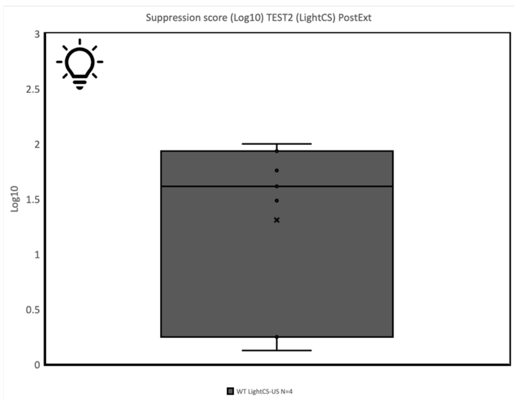
４．研究成果

After several tests of my self-made operant chambers, I tested male and female mice in a sensory- paired presentation of the same tone with a foot shock. For the unpaired groups, mice received paired presentation of the light followed by a tone during the first phase. They then received backward presentation of the foot shock and the tone, that is, the tone came after the foot shock. During the preconditioning procedure, in a conditioned lick suppression task. The paired groups received paired presentation of a light and a tone during the first phase, and test phase, mice of all groups were presented with the light conditioned stimulus (CS) when drinking water. I expected that the light would suppress their water consumption but did not observed significant differences between groups.



As an attempt to overcome this issue, I stepped back and increased the light CS intensity in a first-order conditioning procedure. The rationale behind, is to find optimal parameters and make sure that conditioning occurs with a light CS, which is known to be less efficient than with a tone. In this experiment, mice of the paired group received paired presentation of a light with a foot shock.





The results show that conditioning occurred as predicted. We then added an extinction phase where the light CS was presented 30 times. We predicted a decrease of suppression score (i.e., our index of fear response), and found a non-significant trend of decrease.

We used only a small number of subjects (N=4), and the number of extinction trials might be no sufficient for fully extinguish the CS-US association. These results inform that Light CS can be successfully conditioned to a foot shock, if the CS intensity is high enough, and the number of extinction trials should be increased to observe a complete extinction of the CS-US association. On future attempts, I will use again a sensory-preconditioning procedure with a light CS of higher intensity.

5．主な発表論文等

〔雑誌論文〕　計1件（うち査読付論文　1件／うち国際共著　1件／うちオープンアクセス　0件）

1．著者名	4．巻
Mao Nebuka, Yu Ohmura, Shuntaro Izawa, Youcef Bouchekioua, Naoya Nishitani, Takayuki Yoshida, Mitsuhiro Yoshioka	379
2．論文標題	**5．発行年**
Behavioral characteristics of 5-HT2Creceptor knockout mice: Locomotoractivity, anxiety-, and fear memory-related behaviors	2020年
3．雑誌名	**6．最初と最後の頁**
Behavioural Brain Research	112394
掲載論文のDOI（デジタルオブジェクト識別子）	**査読の有無**
10.1016/j.bbr.2019.112394	有
オープンアクセス	**国際共著**
オープンアクセスではない、又はオープンアクセスが困難	該当する

〔学会発表〕　計1件（うち招待講演　0件／うち国際学会　0件）

1．発表者名
Youcef Bouchekioua, Yuu Ohmura, Mitsuhiro Yoshioka
2．発表標題
Wireless Optogenetic Device Using μLED implants
3．学会等名
Optogenetics 2019
4．発表年
2019年

〔図書〕　計0件

〔産業財産権〕

〔その他〕

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6．研究組織

	氏名 （ローマ字氏名） （研究者番号）	所属研究機関・部局・職 （機関番号）	備考

7．科研費を使用して開催した国際研究集会

〔国際研究集会〕　計0件

8．本研究に関連して実施した国際共同研究の実施状況

共同研究相手国	相手方研究機関