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研究課題名（和文）Dissecting the cortical and subcortical pathways of innate visual fear in the common marmoset

研究課題名（英文）Dissecting the cortical and subcortical pathways of innate visual fear in the common marmoset

研究代表者

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研究成果の概要（和文）：先天的な恐怖は、たとえそれまでに経験したりしたことがなくても、それを恐れる一種の感情です。霊長類におる先天的な恐怖の神経は、まだほとんどわかっていません。研費スタ助成金の助けを借りて、生来の恐怖の神経メカニズムを明らかにするために提案された一連の実験を実行しました。私は全半球の皮質電図を設計し、2匹のマーモセットに移植し、彼らが捕食者や他の動物の写真を見ている間、脳全体の活動と瞳孔の反応を同時に記録成功しました。また、マーモセットがタスクを実行している間に、血圧と心拍数を記録する可能性も検証しました。全体として、実験は順調に進んでおり、先天的な恐怖の神経基盤を理解できるようになるでしょう。

研究成果の学術的意義や社会的意義
Innate fear is a new research field. It is largely unknown in primate. The human extreme of it would be anxiety disorders like social phobia or panic attacks. Through studying innate fear, I hope I can advance our knowledge of fear processing and understand the cause of anxiety disorders.

研究成果の概要（英文）：Innate fear is a type of negative emotion that even if one did not learn or experience before, one is afraid of it. The neural mechanisms of innate fear in primate is still largely unknown. With the help of Kakenhi startup grant, I have performed a series of proposed experiments to uncover the neural mechanism of innate fear. I have designed a full-hemisphere electrocorticography (ECoG) and successfully implanted it in two marmosets. I have also started to record their brain-wide activity and pupil response simultaneously while they are viewing pictures of their predators or other animals. I have completed recording from 1 marmoset. We have also validated the possibility of remotely recording blood pressure and heart rate while the marmosets performing the task. Finally, I have tested several AAV vectors for inhibiting the brain areas with precise time control in rodents. Overall, the experiments are progressing smoothly and I will be able to understand the neural bases of innate fear.

研究分野：Neuroscience

キーワード：Innate fear　Marmoset　Electrophysiology　Optogenetics

１．研究開始当初の背景

Fear is a known negative emotion which acts as a signal of danger or threat, and to trigger adaptive action to overcome the current situation. Fear can be largely separated into two types:

(1). Conditioned fear: Animals learn through experiencing physical or psychological trauma, like pain or stress, and feel fear of certain situation. Most of neural pathways related to fear comes from this type.

(2). Innate fear: Animals feel fear to particular sets of stimuli that they have never experienced before. This type of fear is critical for survival in animals, because it can help them to escape from their predators right after their birth. However, for human this can cause anxiety disorders or specific phobia like fear of height or blood. The neural mechanism of innate fear in primate is still illusive.

In the current study, I showed the pictures of predators or neutral animals to a small new world monkey, the marmoset, while recording their brain-wide activities using full hemisphere electrocorticography (ECoG) to reveal the neural circuit of visual innate fear in primate.

Fig. 1 Marmosets performed 2 picture viewing with first center fixation (left most figure) and then two pictures showed (center figure). The marmosets clearly spent more time watching the snake (innate fear related predator) than the deer (non-fear neutral animal) (right most figure).

２．研究の目的

In primate, the knowledge of innate fear is minimal. One of the main difficulties is that primates adapt rapidly to most of the innate fear related stimulus if there is no physical consequence, like pain, associated with it. However, evidence suggests that the common marmosets do not adapt with repetitive exposure to pictures of their predators. This is in reminiscent of some of the human anxiety disorder. Through studying the neuronal mechanism of innate fear, I can advance our knowledge of fear processing. I can also acquire some hints of how phobia, one of the most common mental illness in the world, may occur. Furthermore, I am expecting to find novel functional brain pathways related to more general visual object processing.

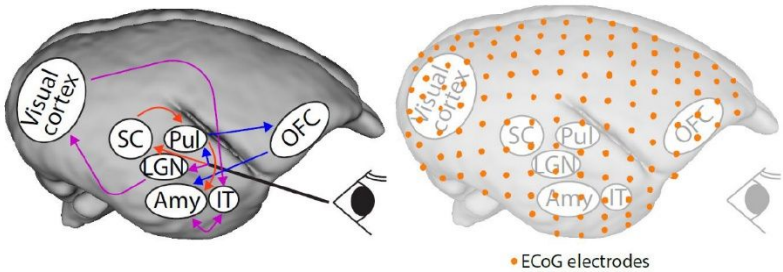

Fig. 2 At least three possible neural circuits proposed to be involved in innate fear (left figure). To disentangle which is the major pathway, a newly designed recording electrode array is needed. This array needs to cover all the dorsal surface of the brain from the most posterior to the anterior with high electrode density.

３．研究の方法

(1). Record the gaze location and the pupillary response of the marmoset while it is viewing pictures of predator or other neutral animals: Marmosets spent more time watching pictures of predator than other animals in my previous research (Fig. 1). To characterize if this behavior is related to fear or not, I am planning to use pupillary response to access the emotional condition of marmosets.

(2). Design and implant a brain-wide, full-hemisphere ECoG to record the brain activity while the marmoset is watching the pictures: The brain areas involving in innate fear processing is massive (Fig. 2). I will need to record the whole brain, starting from the primary visual cortex all the way to the prefrontal cortex, to analyze the potential neural pathway.

(3). Use optogenetics to manipulate the innate fear related brain areas to establish a causal relationship: After identifying the major circuit and brain areas by comparing the differential brain activation pattern of the marmoset watching

different pictures, I will optically stimulate those brain areas to establish the causal relationship between brain activities and innate fear.

4. 研究成果

(1). I have successfully adapted my previous behavior task for the marmosets into showing one picture on the screen only (Fig. 3). I have also successfully recorded the pupillary response while the marmosets were freely watching the picture, I found that the marmosets showed smaller pupil diameter while watching the pictures of predators (innate fear related) than neutral animals. In addition, they showed opposite pupillary response to the anti-bite glove, which is not innate fear related, but conditioned fear related. This behavioral response further supports that the innate and the conditioned fear may not share completely the same neural pathway.

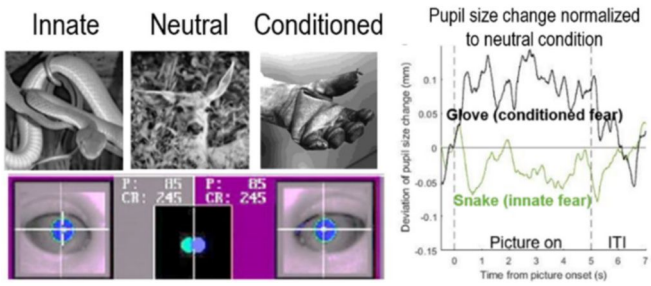

Fig. 3 Marmosets showed differential pupillary responses of innate fear, neutral, or conditioned fear related pictures.

(2). For developing the novel ECoG array covering a whole hemisphere of the brain, I asked for help from a company called CirTech. With several back-and-forth discussion, I have finally completed the design. The ECoG array was produced and implanted successfully in two marmosets. (Fig. 4). I have collected significant amount of brain activity data while the marmosets performing the picture viewing task describe above. What I found was that at the first 50 ms, the marmosets did not show significant differential brain activity in the gamma frequency range between pictures of predators to neutral animals. But 1500 ms later, a very strong difference showed where predominantly in the parietal and frontal cortex (Fig. 5). Further analysis is needed to pin down the information flow difference with time between these two conditions.

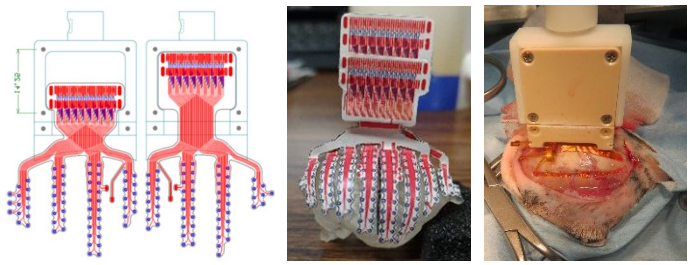

Fig. 4 (Left) Design of the ECoG array containing 128 electrode channels. (Middle) Dummy implanting on a 3D printed brain surface showing the coverage of the ECoG array. (Right) Array successfully implanted in 1 marmoset. For all figures, left is occipital (posterior) direction.

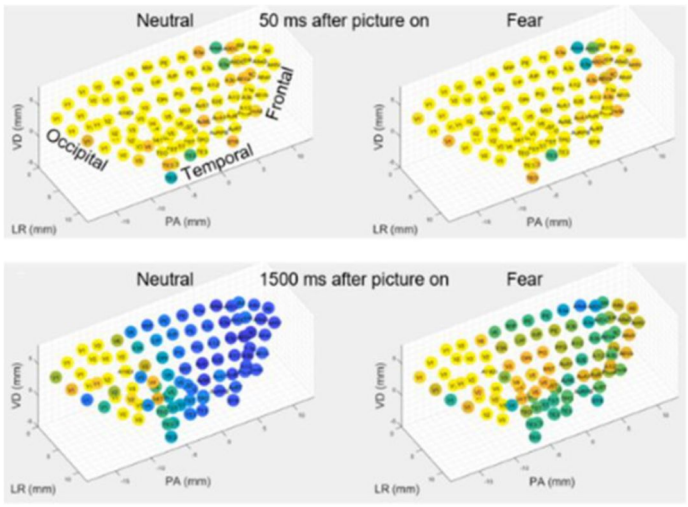

Fig. 5 Differential gamma band response in fear to neutral pictures 1500 ms after pictures showed, but not 50 ms.

(3). The optogenetic modulation for the marmosets was first tested on rats (Fig. 6). I first constructed two virus vectors, AAV-Thy1-stTA and AAV-TRE-ChrimsonR-mCherry. The former one is for amplifying the latter opsin carrying vector. Both vectors were wrapped by recombinant AAV, AAV2.1, for better expression in primate brain. The results showed promising and clear reporter protein expression spanning 3.84 mm in the rat cortex. When the analysis finished, I can pin down the brain area and perform the next step.

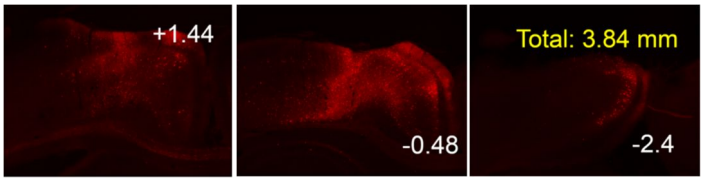

Fig. 6 Optogenetic tests on rat cortex showed promising wide infection of the reporter protein

５．主な発表論文等

〔雑誌論文〕　計5件（うち査読付論文　5件／うち国際共著　5件／うちオープンアクセス　0件）

１．著者名 Chen Chih-Yang、Matrov Denis、Veale Richard、Onoe Hirotaka、Yoshida Masatoshi、Miura Kenichiro、Isa Tadashi	４．巻 125
２．論文標題 Properties of visually guided saccadic behavior and bottom-up attention in marmoset, macaque, and human	５．発行年 2021年
３．雑誌名 Journal of Neurophysiology	６．最初と最後の頁 437～457
掲載論文のDOI（デジタルオブジェクト識別子） 10.1152/jn.00312.2020	査読の有無 有
オープンアクセス 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	国際共著 該当する

１．著者名 Veale Richard、Chen Chih-yang、Isa Tadashi	４．巻 2021
２．論文標題 Marmoset Monkeys Model Human Infant Gaze?	５．発行年 2021年
３．雑誌名 2021 IEEE International Conference on Development and Learning (ICDL)	６．最初と最後の頁 1
掲載論文のDOI（デジタルオブジェクト識別子） 10.1109/ICDL49984.2021.9515602	査読の有無 有
オープンアクセス 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	国際共著 該当する

１．著者名 Hafed Ziad M.、Chen Chih-Yang、Tian Xiaoguang、Baumann Matthias P.、Zhang Tong	４．巻 125
２．論文標題 Active vision at the foveal scale in the primate superior colliculus	５．発行年 2021年
３．雑誌名 Journal of Neurophysiology	６．最初と最後の頁 1121～1138
掲載論文のDOI（デジタルオブジェクト識別子） 10.1152/jn.00724.2020	査読の有無 有
オープンアクセス 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	国際共著 該当する

１．著者名 Hafed Ziad M.、Hoffmann Klaus-Peter、Chen Chih-Yang、Bogadhi Amarender R.	４．巻 9
２．論文標題 Visual Functions of the Primate Superior Colliculus	５．発行年 2023年
３．雑誌名 Annual Review of Vision Science	６．最初と最後の頁 1
掲載論文のDOI（デジタルオブジェクト識別子） 10.1146/annurev-vision-111022-123817	査読の有無 有
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1．著者名 Hafed Ziad M.、Hoffmann Klaus-Peter、Chen Chih-Yang、Bogadhi Amarender R.	4．巻 9
2．論文標題 Visual Functions of the Primate Superior Colliculus	5．発行年 2023年
3．雑誌名 Annual Review of Vision Science	6．最初と最後の頁 11.1-11.23
掲載論文のDOI（デジタルオブジェクト識別子） 10.1146/annurev-vision-111022-123817	査読の有無 有
オープンアクセス 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	国際共著 該当する

〔学会発表〕　計6件（うち招待講演　1件／うち国際学会　1件）

1．発表者名 Chen Chih-Yang
2．発表標題 The ventral part of dorsorostral area 6 (6DR) in common marmoset exhibits saccade-related activity
3．学会等名 Annual Meeting of the Japan Society of Marmoset Research
4．発表年 2022年

1．発表者名 Chen Chih-Yang
2．発表標題 Functional mapping and anatomical tracing of the saccade related region in dorsal frontal cortex of common marmoset
3．学会等名 Annual Meeting of the Japan Society of Marmoset Research
4．発表年 2023年

1．発表者名 Chen Chih-Yang
2．発表標題 Mapping saccade representation in the frontal cortex of common marmoset
3．学会等名 Annual Meeting of the Japan Neuroscience Society
4．発表年 2020年

１．発表者名
Chen Chih-Yang

２．発表標題
Mapping saccade representation in the frontal cortex of common marmoset

３．学会等名
Taiwan Society for Neuroscience Meeting（招待講演）（国際学会）

４．発表年
2020年

１．発表者名
Chih-Yang Chen

２．発表標題
Functional mapping and anatomical tracing of the saccade related region in dorsal frontal cortex of common marmoset

３．学会等名
Annual Meeting of The Physiological Society of Japan

４．発表年
2023年

１．発表者名
Chih-Yang Chen

２．発表標題
Functional mapping and anatomical tracing of the saccade related region in dorsal frontal cortex of common marmoset

３．学会等名
日本マーモセット研究会大会

４．発表年
2023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