Wireless cell-type specific magnetic control of neural activities in live animals

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Introduction

Neuromodulation is an important method for the neuro-behavioral and brain circuit study. Magnetogenetics is one of the neuromodulation tool using magnetism which is wireless and able to modulate deep brain areas. There are three elements for using magnetogenetics, uniform magnetic field, magnetic nanoparticle, and mechanosensitive ion channel. The mechanosensitive ion channel was expressed in the mouse's brain and a magnetic nanoparticle, m-Torquer bound with ion channel. When the mouse was in the magnetic field, the torque force generated by m-Torquer opened the mechanosensitive ion channel and induced the ion influx. In previous research, the mechanosensitive calcium channel Piezo1 was targeted for the application of magnetogenetics (Lee, Ju., Shin, W., Lim, Y. et al, 2021). The calcium cation ion influx occurs the neuron excitation. For the next step of the research, mechanosensitive chloride channel DmFlyC1 was targeted by the reason that chloride ion influx causes neuron inhibition. To check the behavioral change when stimulating the neuron by magnetogenetics, we set the rotating magnetic field with the food zone.

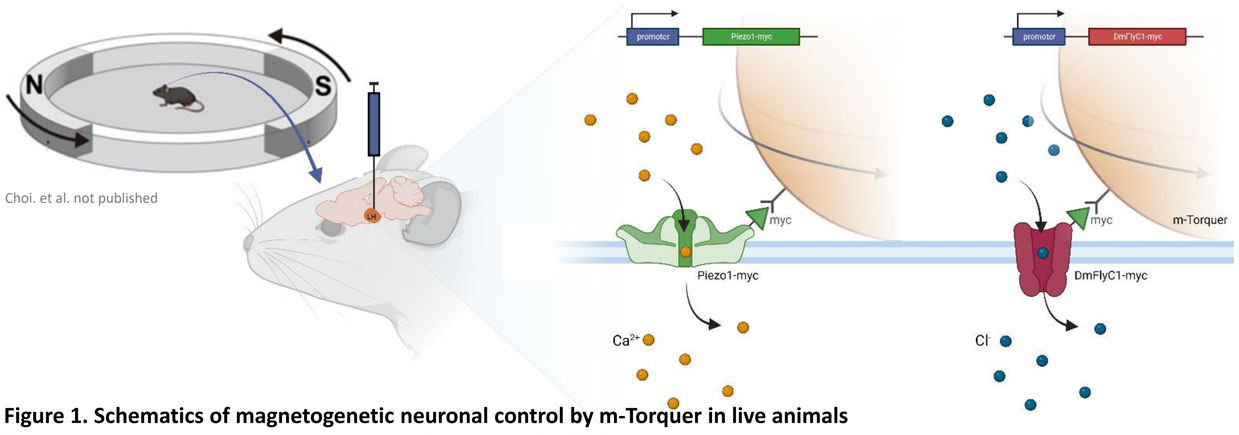

Figure 1. Schematics of magnetogenetic neuronal control by m-Torquer in live animals
The mechanosensitive ion channel, such as Piezo1 and DmFLYC1, engineered with myc-tag is expressed in the deep brain using viral transduction. Rotating magnetic field induces the torque force by magnetic nanoparticle, m-Torquer. Since m-Torquer bind with myc, the ion influx occurs. When the Piezo1 expressed, cation ion diffused into the cell and neuron excitation will be occurred. On the other hand, when DmFlyC1 expressed, chloride ion diffused into the cell and neuron inhibition will be occurred.

Vgat neuron-specific stimulation increases feeding

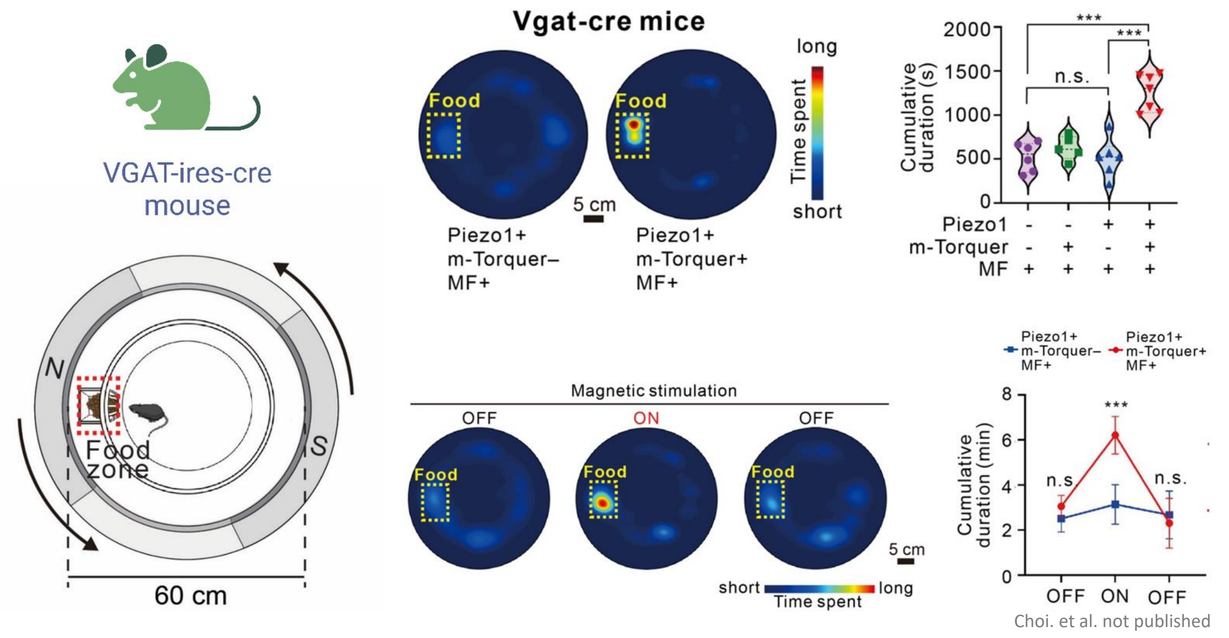

Figure3. in vivo experiment of Vgat-cre-Piezo1
Piezo1-myc was specifically expressed in Vgat neuron, LH region by cre recombinase. Since the Vgat neuron in LH region excitation induced the feeding, mouse in the rotating magnetic field shows the increase of time spent in the food zone. The mice with Piezo1 and m-Torquer show increased feeding behavior in response to magnetic field. When MF is off, the feeding behavior is reduced. This shows that magnetogenetics by m-Torquer is reversible.

Cell-type specific magnetogenetics

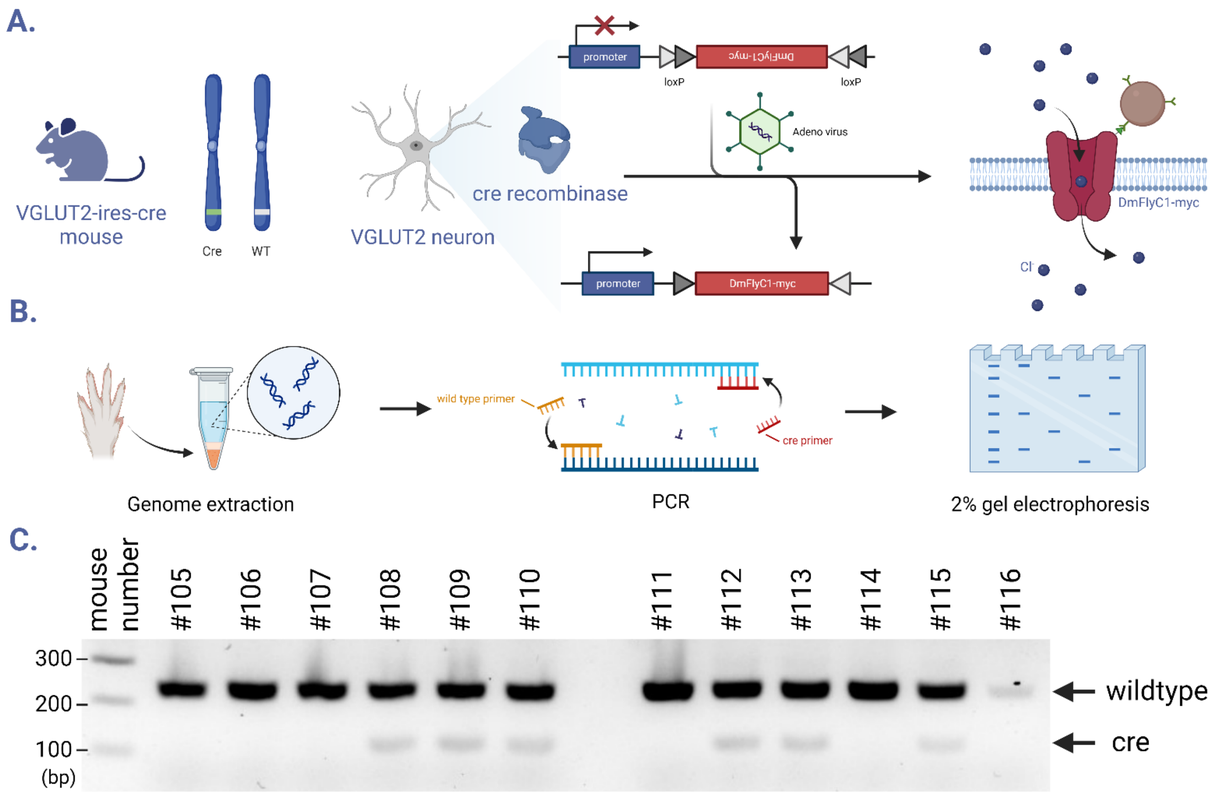

Figure2. Cell-type specific Magnetogenetics
(A) VGLUT2-ires-cre mouse expresses the cre recombinase and cre recombinase inverted the sequence transduced by adeno virus. Since cre recombinase only expressed in VLGUT2 neuron, the specific binding of nanoparticle will be able.
(B) Process of genotyping. Extract the genome from the toes and amplified the DNA with mutant VGLUT2 and WT primer. After PCR, progress 2% agarose gel electrophoresis
(C) Genotyping result. The Cre recombinase activity is detected in Vglut2 positive. Since the cre mutant DNA band is identified at 124bp and the wildtype 245bp, hetero type mouse shows two bands. #108, #109, #110, #112, #113, #115 shows two band while the other number shows just wild type band. This indicated that the 6 samples are cre hetero type.

In vivo expression of DmFLYC1 for magnetogenetics

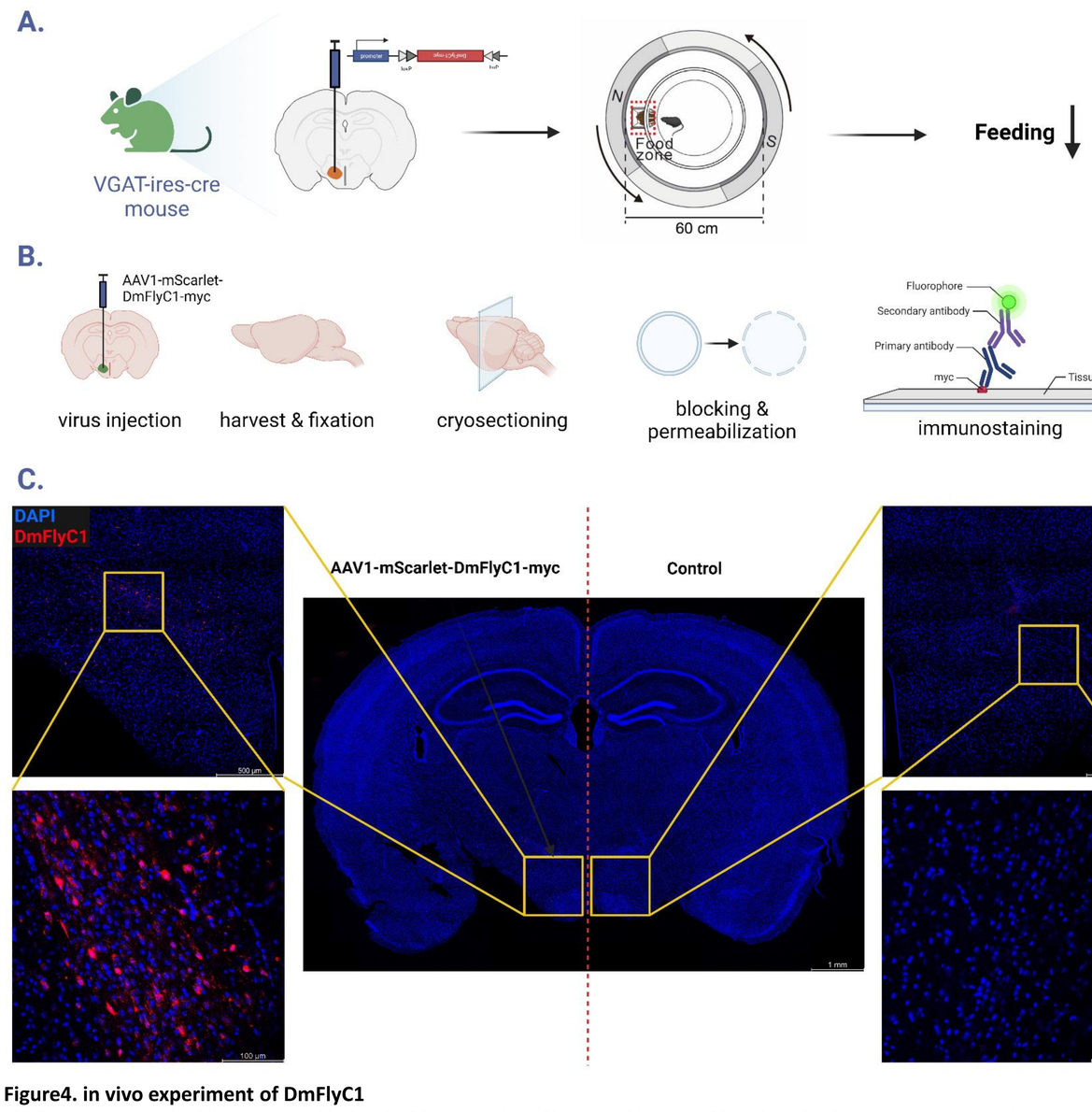

Figure4. in vivo experiment of DmFlyC1
(A) Expected data for Vgat-cre specific DmFlyC1 expression. Since the Vgat-cre-Piezo1 excitation the neuron and increase the feeding, Vgat-DmFlyC1 will inhibit the neuron and decrease the feeding.
(B) Process of Immunohistochemistry. After 3 weeks of virus injection, harvest the brain and post-fixation for 24 hours. Freeze the brain tissue, and cryosection coronal. Select the brain slicing with LH region and progress the blocking and permeabilization. The primary antibody is myc tag, rabbit and the secondary antibody is rabbit 488nm for immunostaining. Dapi was diluted as 2,500x.
(C) 20x, 25x image of microscopy. LH region of left side was injected DmFlyC1 whereas LH region of right side was control. AAV1-mScarlet-DmFlyC1-myc expression was identified by mScarlet fluorescence signal(red).

Future Study

Checking Social Behavior

Magnetogentics using m-Torquer is wireless so that mouse can freely move in the magnetic field. In this respect, the multiple mice can be in the magnetic field with no tethered. So, researchers can check the social behavior with neuro modulation.

Searching Brain circuits

The brain tissues are so heterogeneity and have million of neurons with mutual organic connection. By this reason, excitation and inhibition can be powerful tools for researching and finding brain circuits.

Treatment of Neurodegenerative disease

Neurodegenerative disease (e.g. Parkinson disease, post-stroke) is occurred by damaging of specific neuron modulation. Applying the magnetogenetics can be approached to the treatment.

Deep Brain neuromodulation

Whereas optogenetics cannot modulate the deep brain region causes by short delivery distance, the magnetogenetics using m-Torquer can apply to the deep brain region. This can extend the research area for the large animals or deep region of brain in the neuroscience.

Reference

Lee, Ju., Shin, W., Lim, Y. et al. Non-contact long-range magnetic stimulation of mechanosensitive ion channels in freely moving animals. Nat. Mater. 20, 1029–1036 (2021). 10.1038/s41563-020-00896-y

J.S. Wiegert, M. Mahn, M. Prigge, Y. Printz, O. Yizhar. Silencing neurons: tools, applications, and experimental constraints. Neuron. 95, 504-529 (2017). 10.1016/j.neuron.2017.06.050

Mattis J, Tye KM, Ferenczi EA, Ramakrishnan C, O'Shea DJ, Prakash R, Gunaydin LA, Hyun M, Fenno LE, Gradinaru V, et al. Principles for applying optogenetic tools derived from direct comparative analysis of microbial opsins. Nat Methods. 2011;9:159–172.