



Understanding serotonergic regulation of motor functions

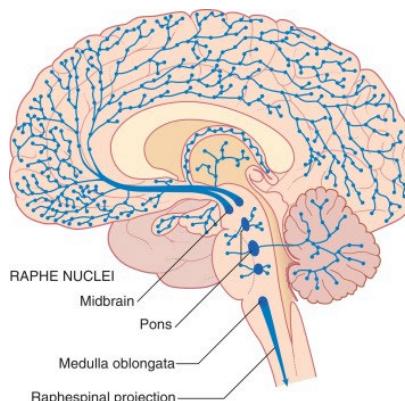
Yenah Bak

Department of Brain and Cognitive Sciences

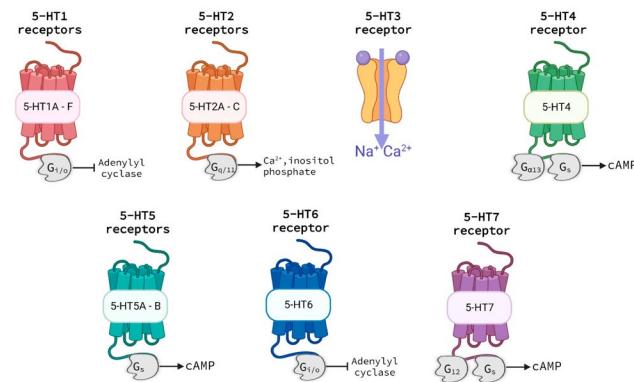
Serotonergic system:

It is a complex system with brain-wide projection, multiple receptors, volume transmission that affects various functions.

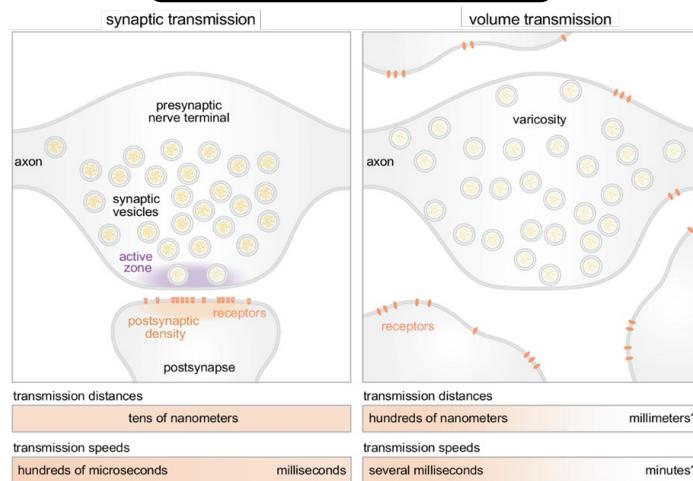
Projections



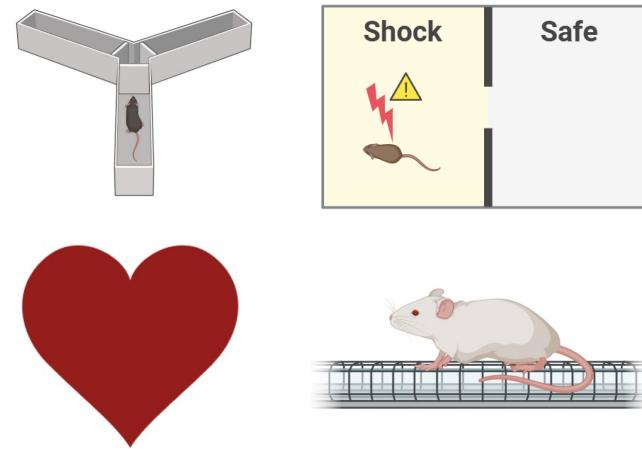
Receptors



Transmission

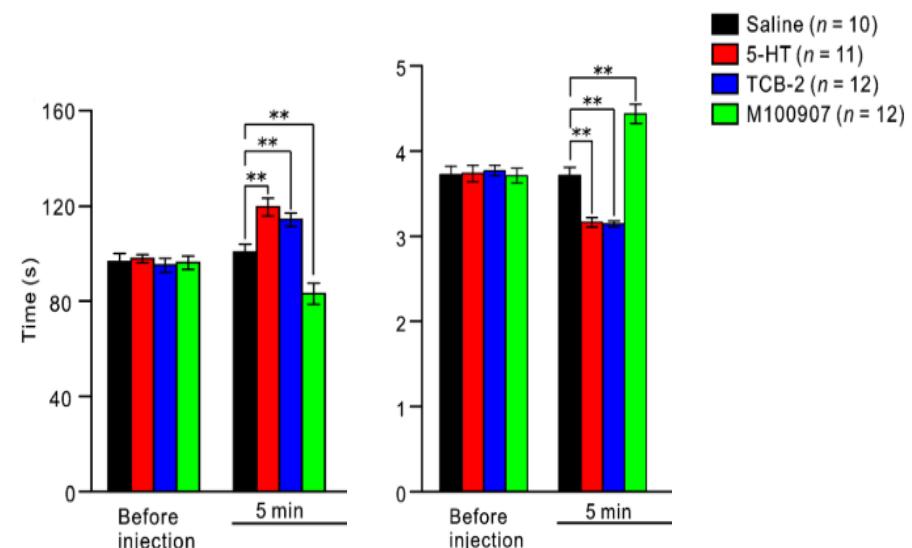


Functions



Serotonergic effect on Motor Control:

Serotonin (5-HT) regulates motor ability through Fastigial Nucleus (fDCN)



Balance beam

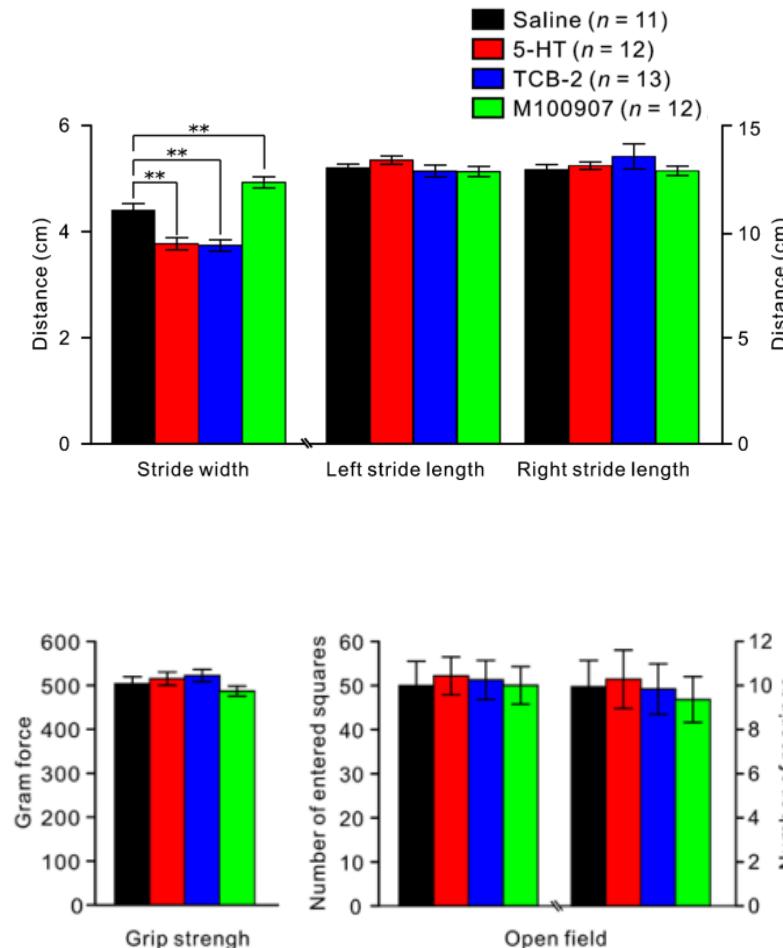
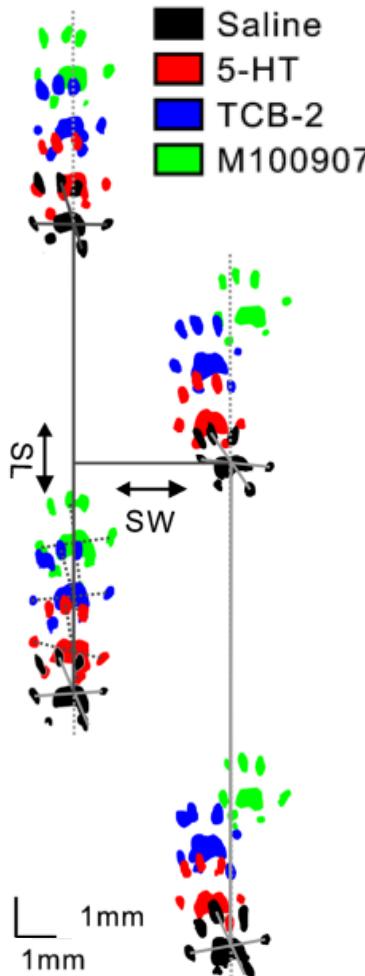


TCB-2: Serotonin 2A receptor Agonist

M100907: Serotonin 2A receptor Antagonist

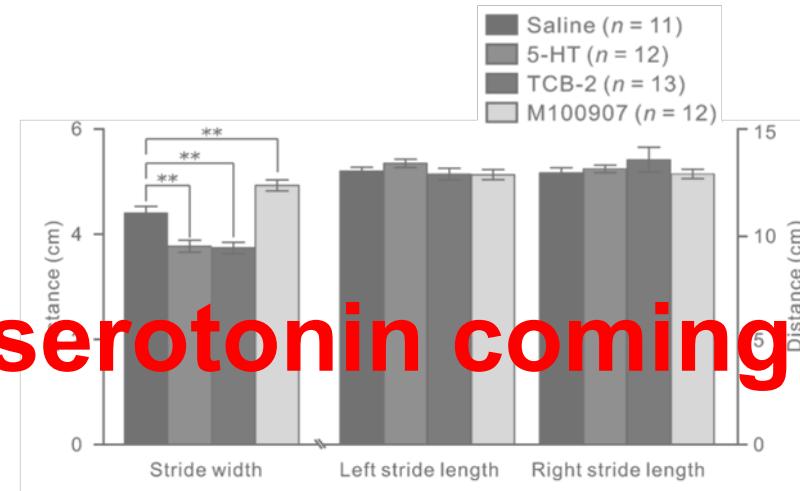
Serotonergic effect on Motor Control:

Serotonin promotes motor performance, especially motor balance and coordination through 2a receptor
in the cerebellar fastigial nucleus

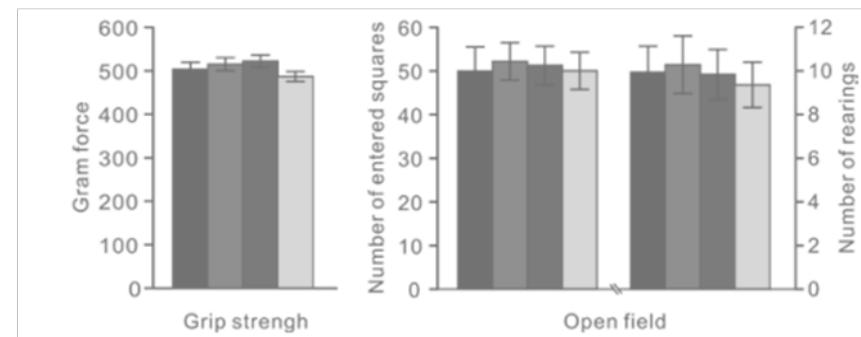


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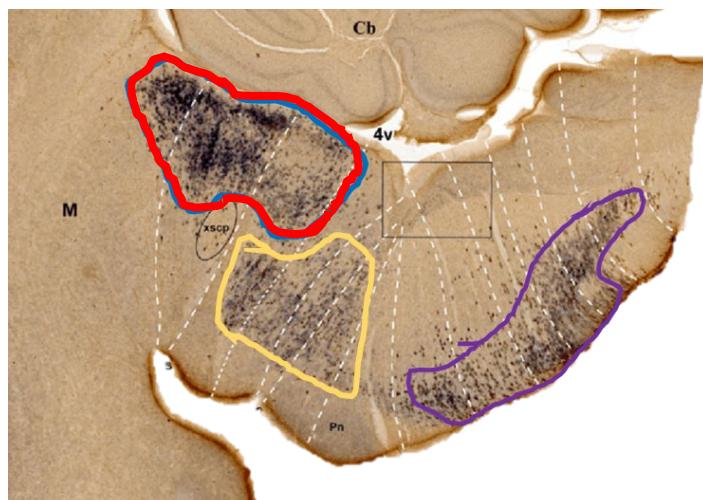
Where is the serotonin coming from?



Serotonergic effect on Motor Control:

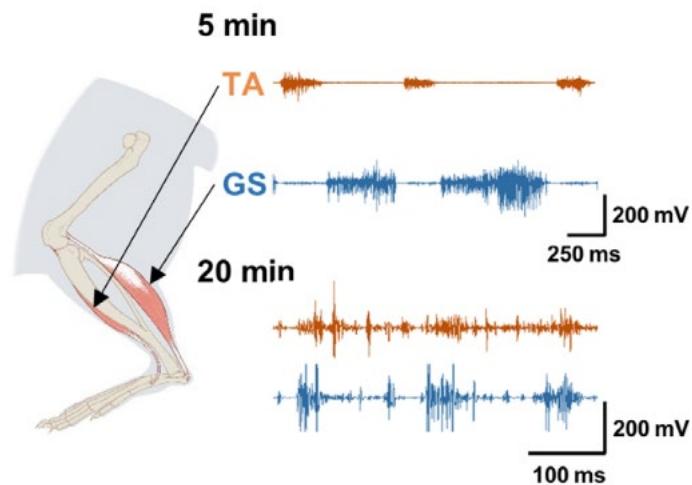
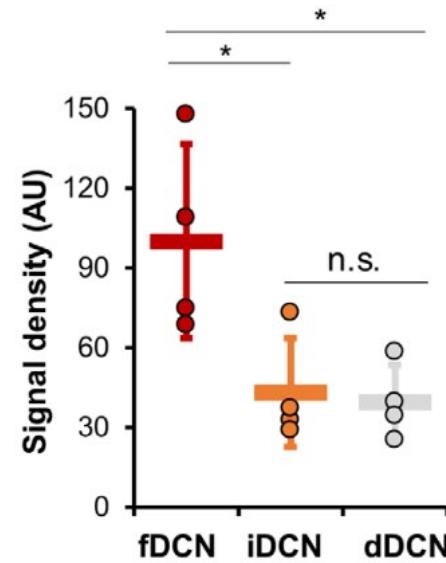
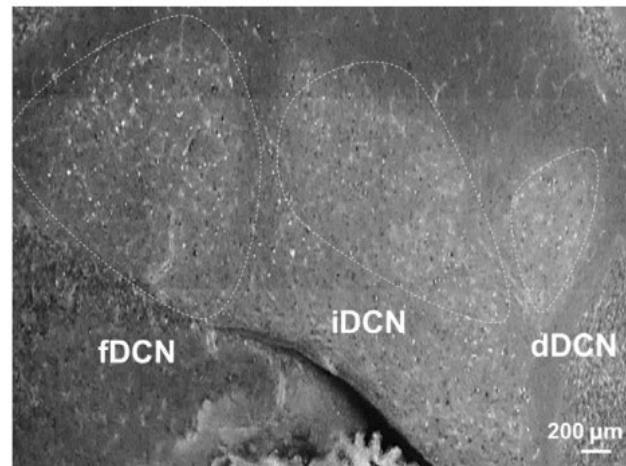
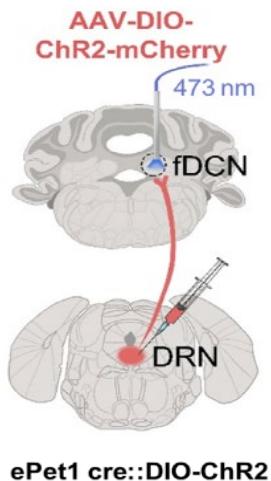
Serotonergic neurons are primarily located in the raphe nuclei, including the dorsal raphe nucleus, median raphe nucleus, and caudal raphe nucleus

Sagittal view of the raphe nucleus



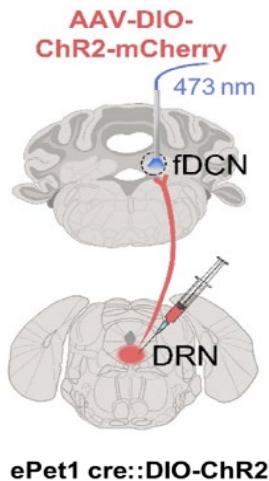
Serotonergic effect on Motor Control:

Hyperexcitation of serotonergic neurons at fastigial nucleus induces motor disorder, dystonia

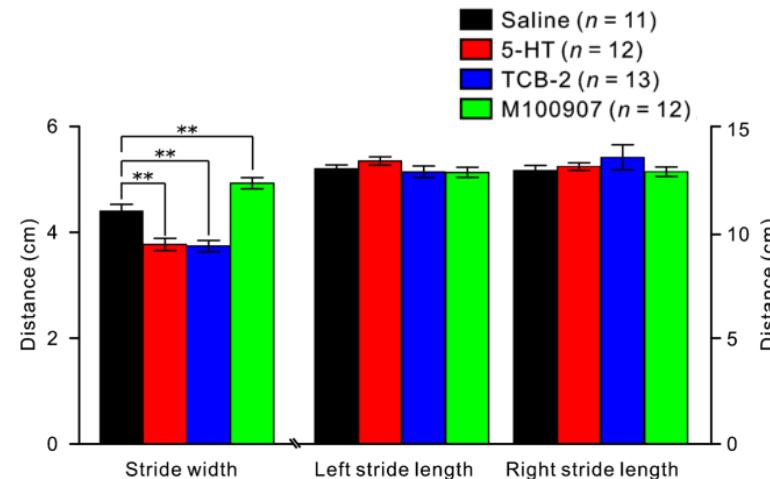


Serotonergic effect on Motor Control:

Previous studies show DRN serotonergic neurons might regulate motor ability via fastigial nucleus



+



Questions:

1. Could serotonin that regulates motor control from the DRN?
2. What information do these serotonergic neurons encode?



Outline

1. Conducted Experiments

1. Circuit tracing and cell population
2. Chemogenetic study
3. Fiber photometry study

2. Future Experiments

Questions:

1. Could serotonin that regulates motor control from the DRN?
2. What information do these serotonergic neurons encode?



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1. Circuit tracing and cell population
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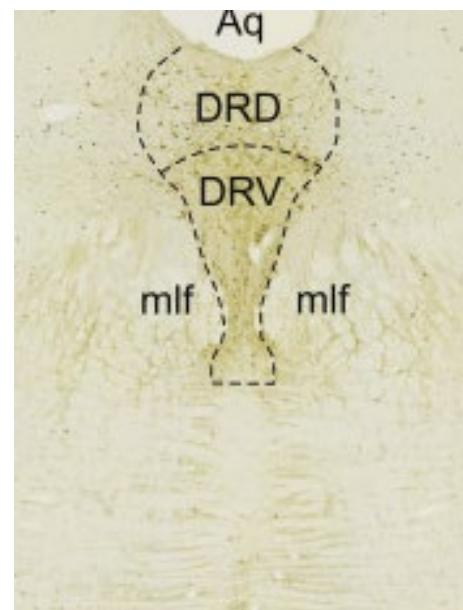
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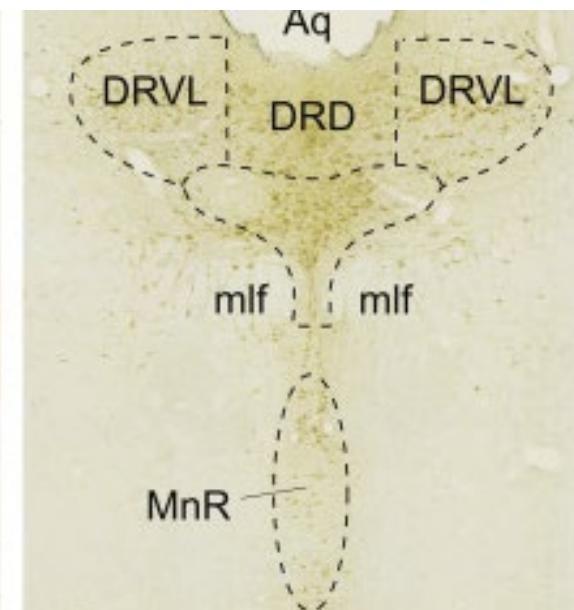
1. FN-projecting DRN population:

DRN can be divided input dorsal (DRD), ventral(DRV), and lateral (DRVL)



AP:

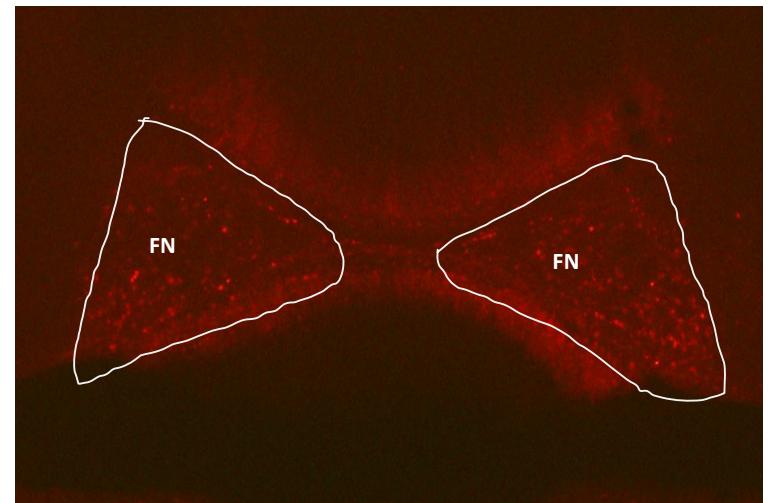
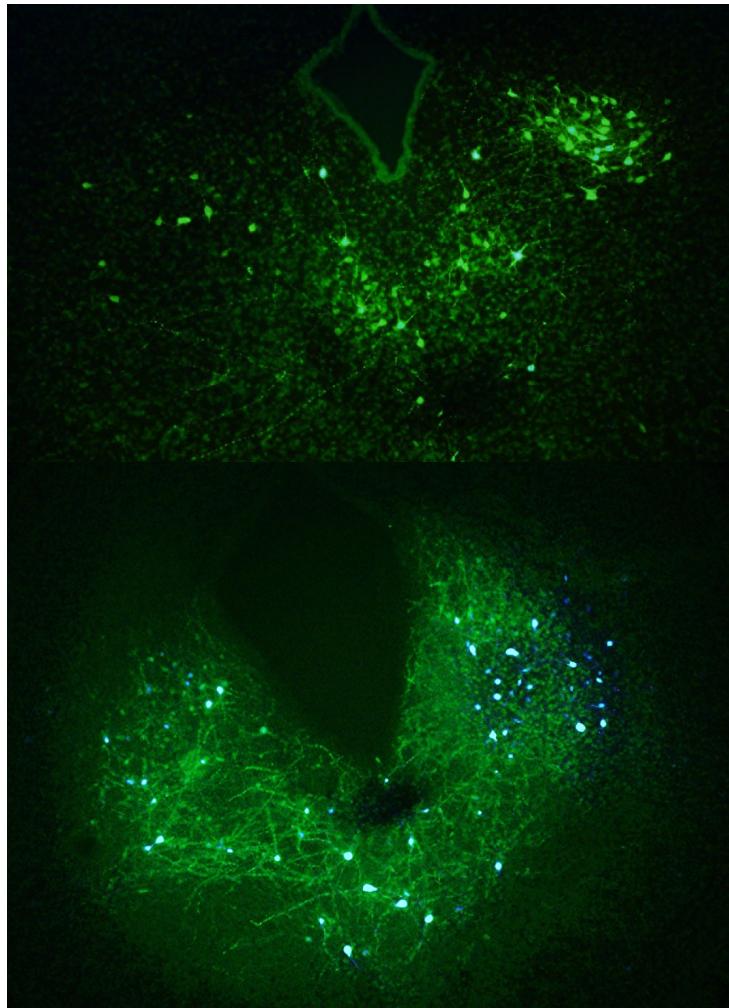
-4.2



-4.6

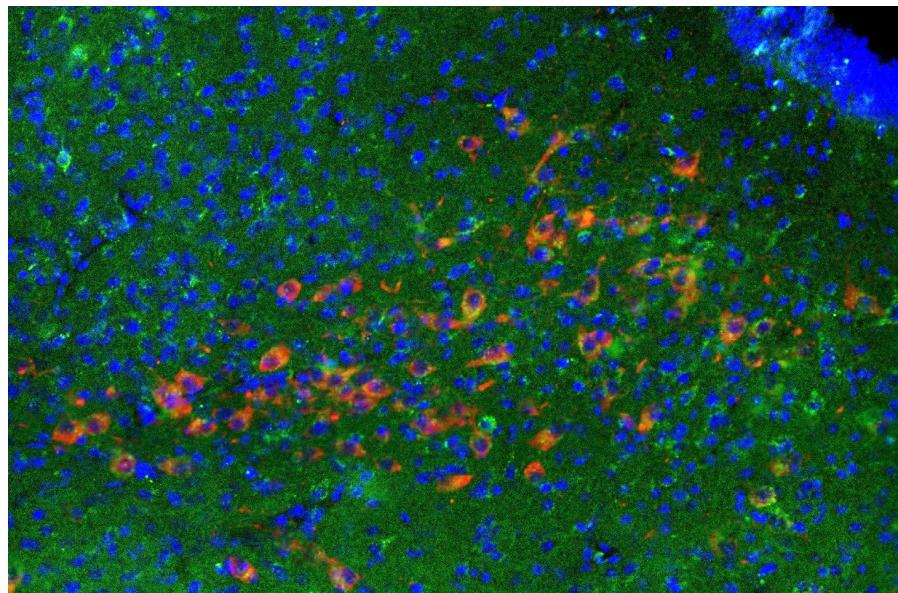
1. FN-projecting DRN population:

Retrograde experiment showed neurons in the dorsal raphe nucleus project to the fastigial nucleus



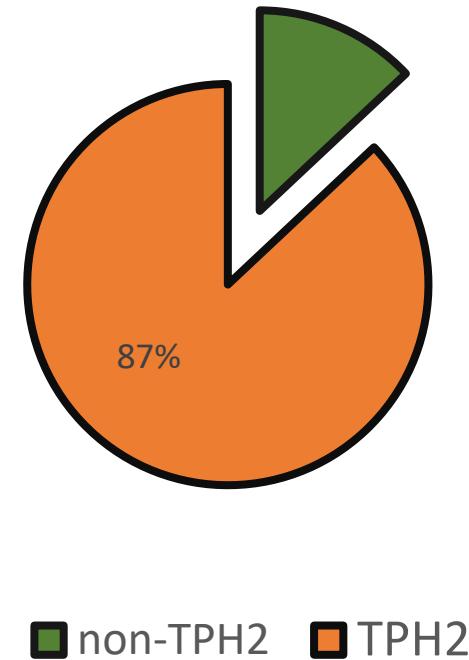
1. FN-projecting DRN population:

Anti-TPH2 staining showed majority of neurons projecting to the fastigial nucleus are serotonergic



Retrogradely labelled

TPH2-positive



Outline

1. Conducted Experiments

1. Circuit tracing and cell population

Result: Majority of dorsal DRN neurons projecting to the fastigial nucleus are serotonergic.

2. Chemogenetic study

3. Fiber photometry study

4. Electromyography study

2. Future Experiments

Questions:

1. Could serotonin that regulates motor control from the DRN?
2. What information do these serotonergic neurons encode?

Outline

1. Conducted Experiments

1. Circuit tracing and cell population
2. Chemogenetic study
3. Fiber photometry study
4. Electromyography study

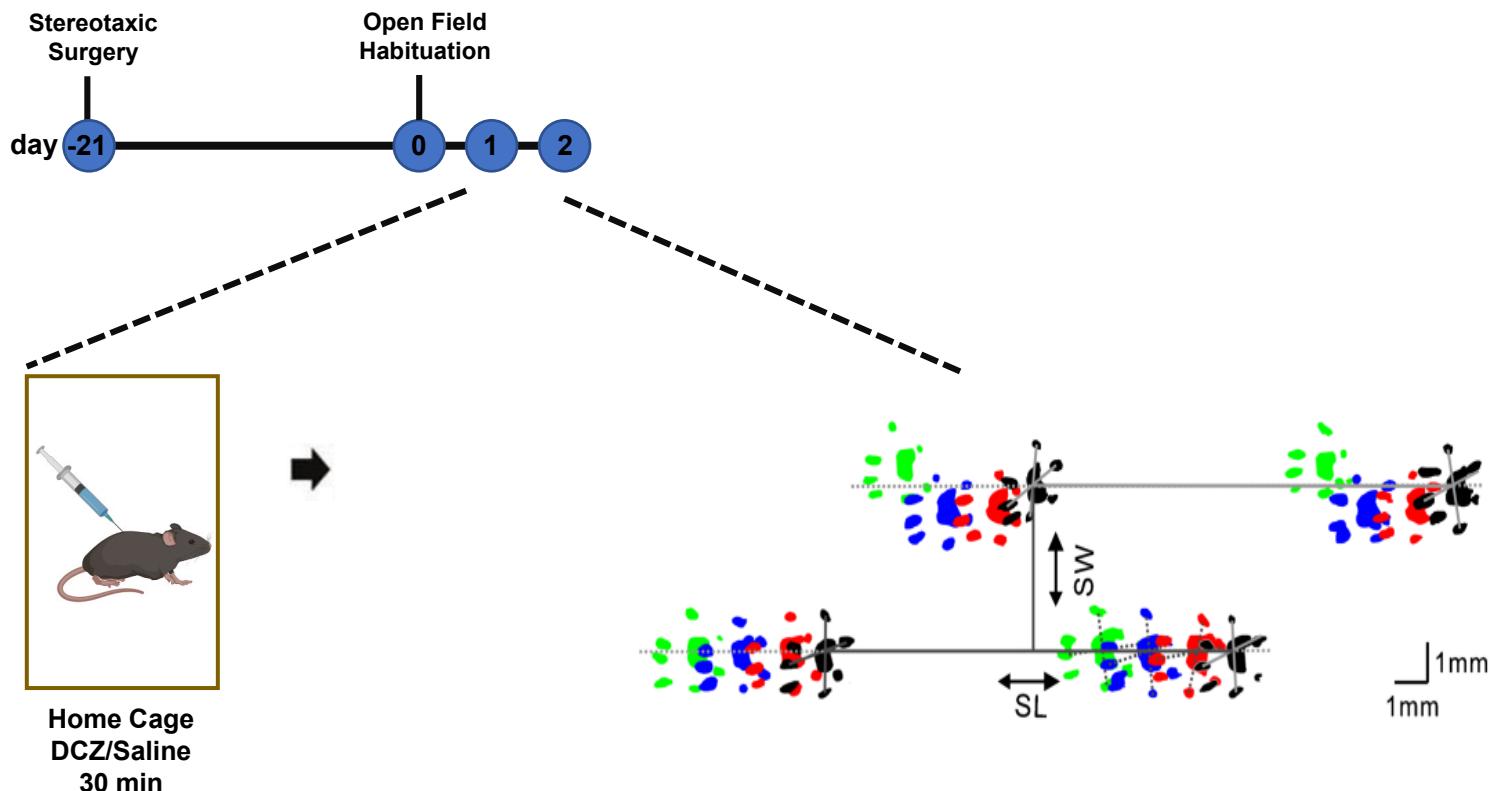
2. Future Experiments

Questions:

1. Could serotonin that regulates motor control from the DRN?
2. What information do these serotonergic neurons encode?

2. DRN-FN Chemogenetic experiments:

Gait analysis were conducted during chemogenetic manipulation

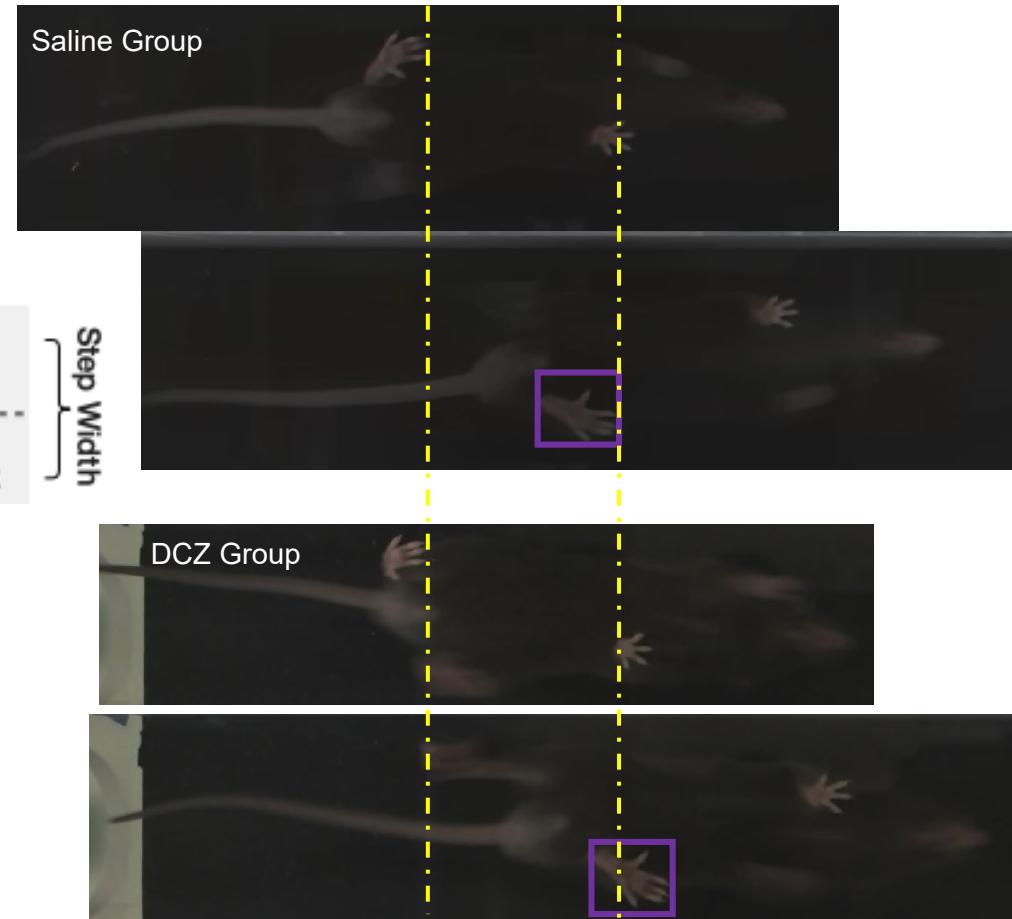
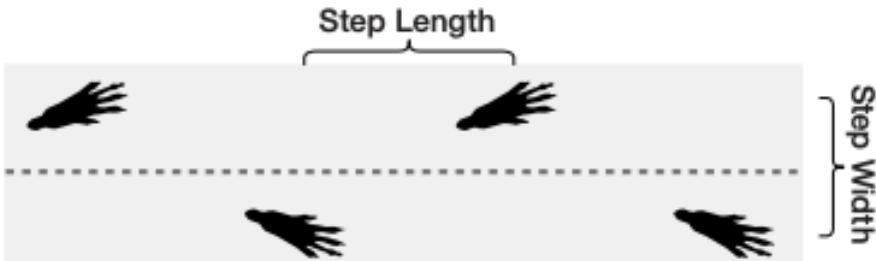


DCZ: 0.2mg/kg in 0.9% saline;
pre-experiment IP injection

2. DRN-FN Chemogenetic experiments:

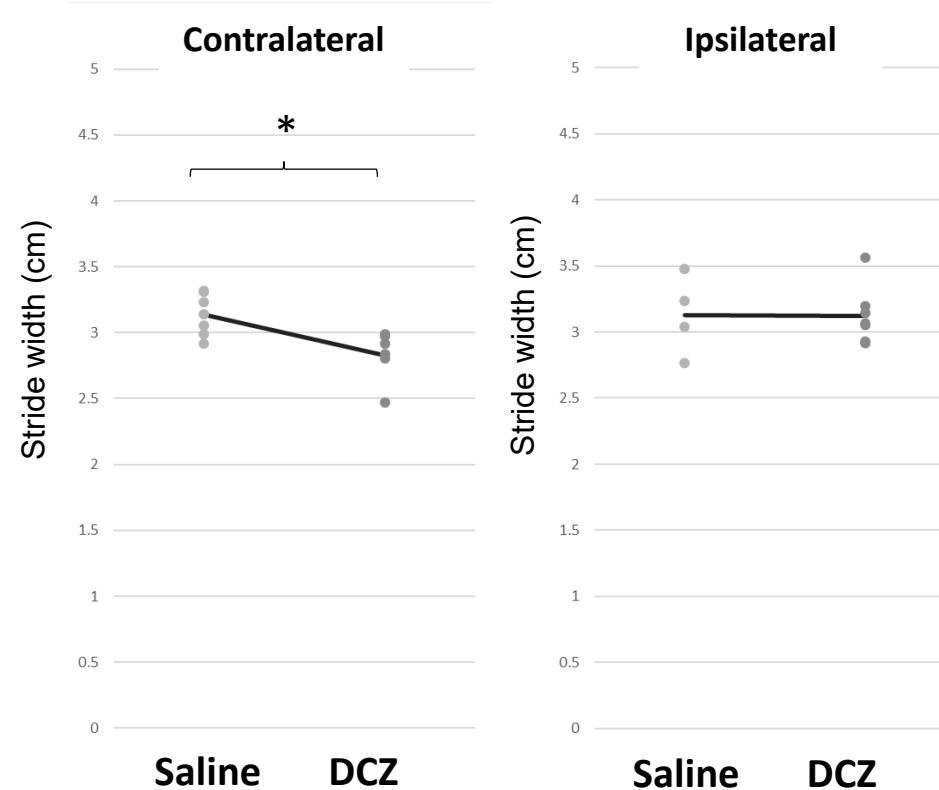
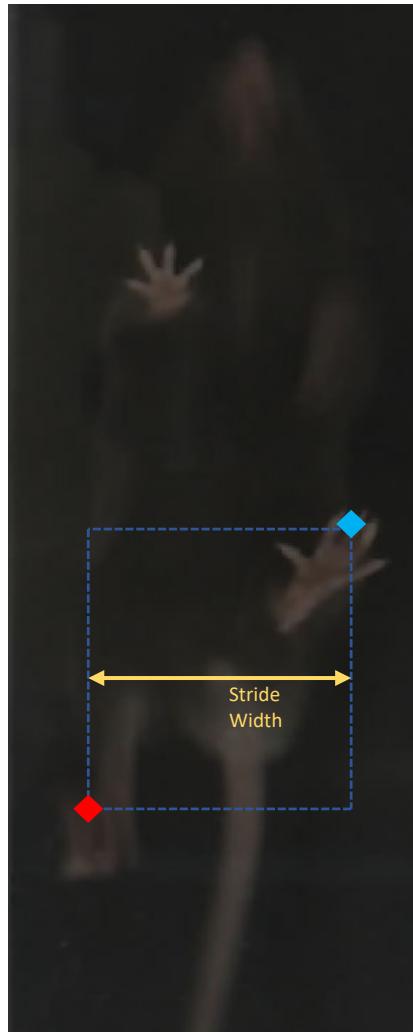
Catwalk: Unilateral DRN (Right) excitation

Spatial Characteristics



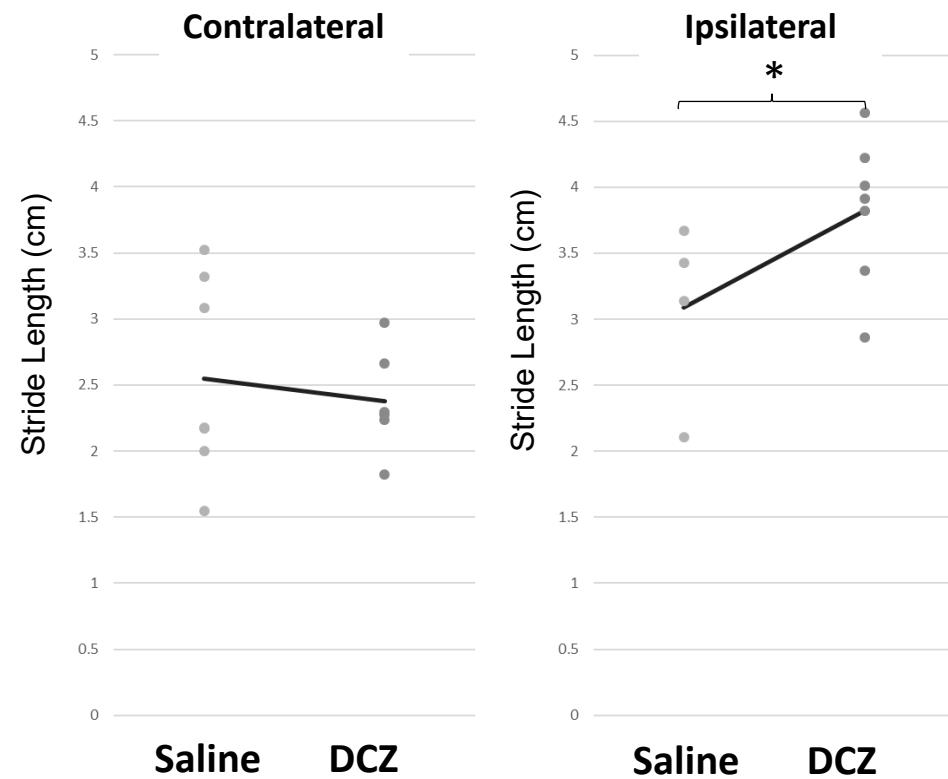
2. DRN-FN Chemogenetic experiments:

Unilateral DRN (Right) excitation induced narrower stride width on contralateral side



2. DRN-FN Chemogenetic experiments:

Unilateral DRN (Right) excitation induced longer stride length on ipsilateral side



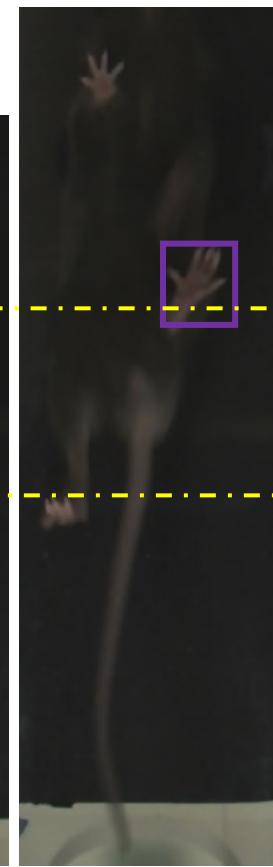
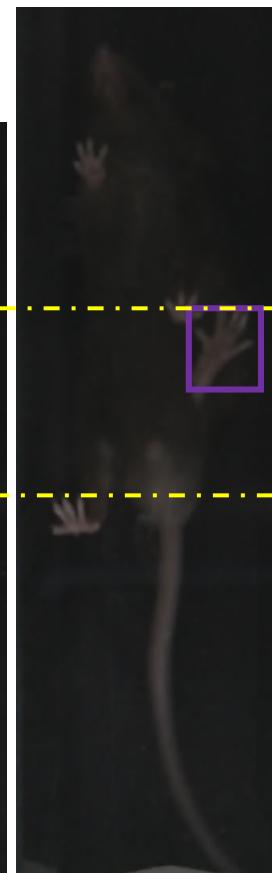
2. DRN-FN Chemogenetic experiments:

Unilateral FN excitation induced stride differences in the same pattern as DRN excitation

Saline Group

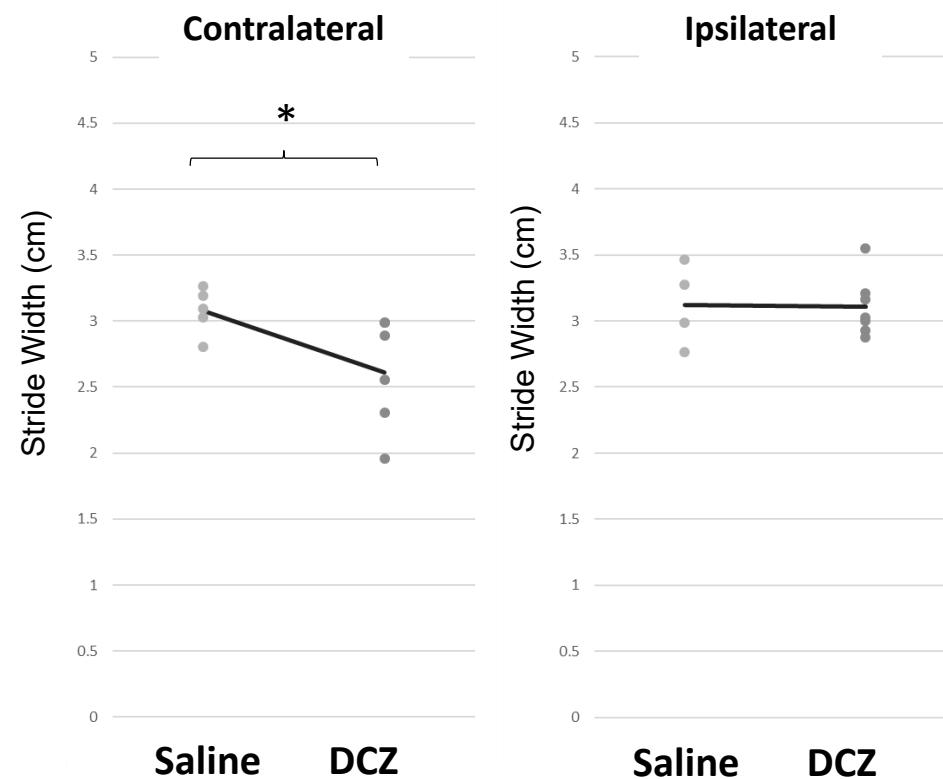
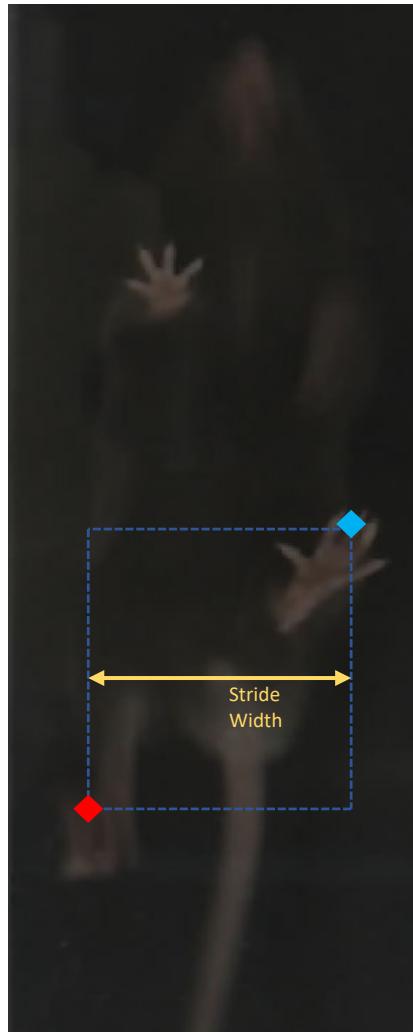


DCZ Group



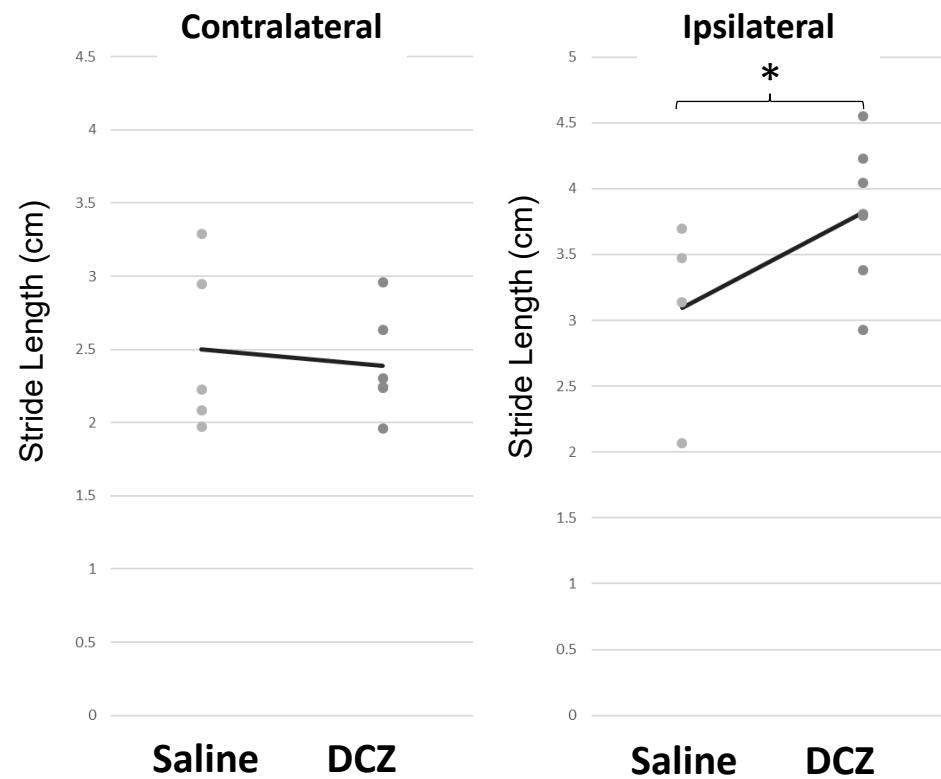
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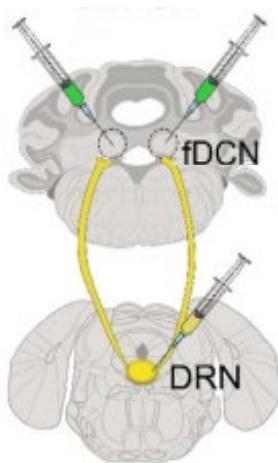
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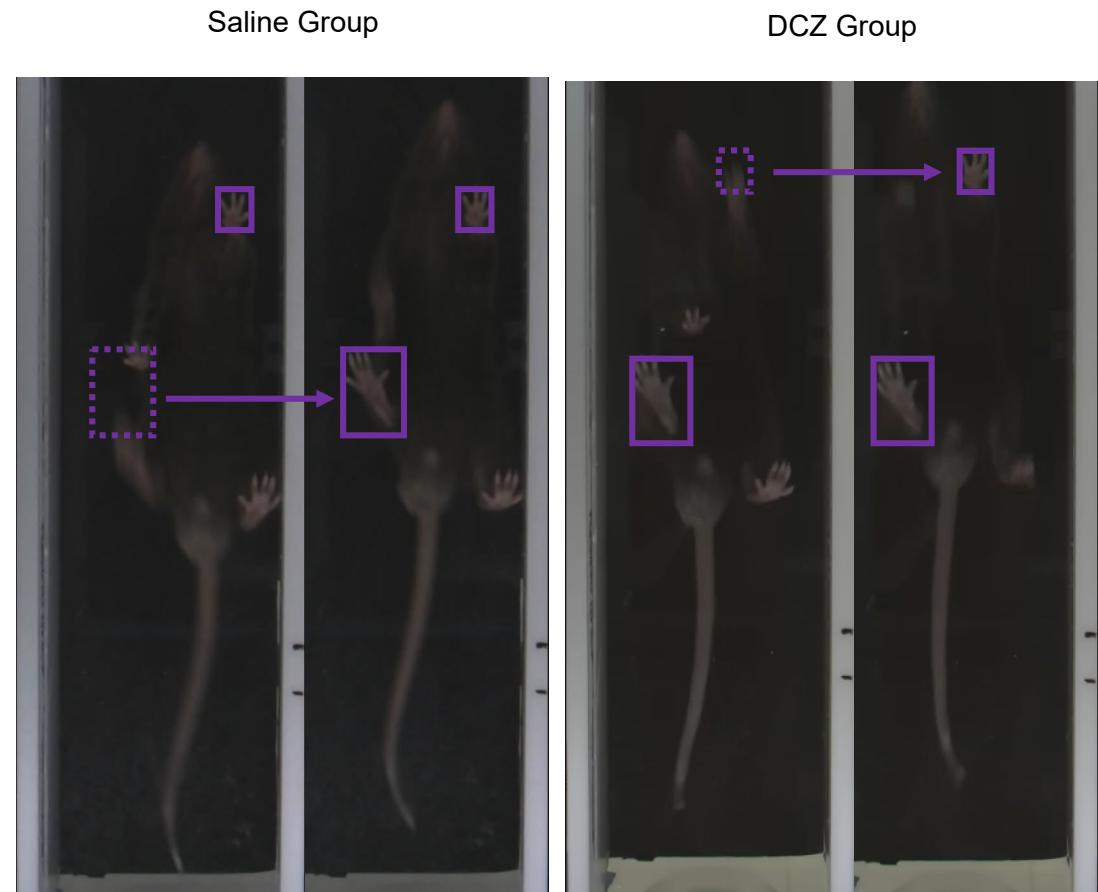
2. DRN-FN Chemogenetic experiments:

Bilateral inhibition of FN glutamatergic neurons results lagging of hindlimb but not different in gait pattern



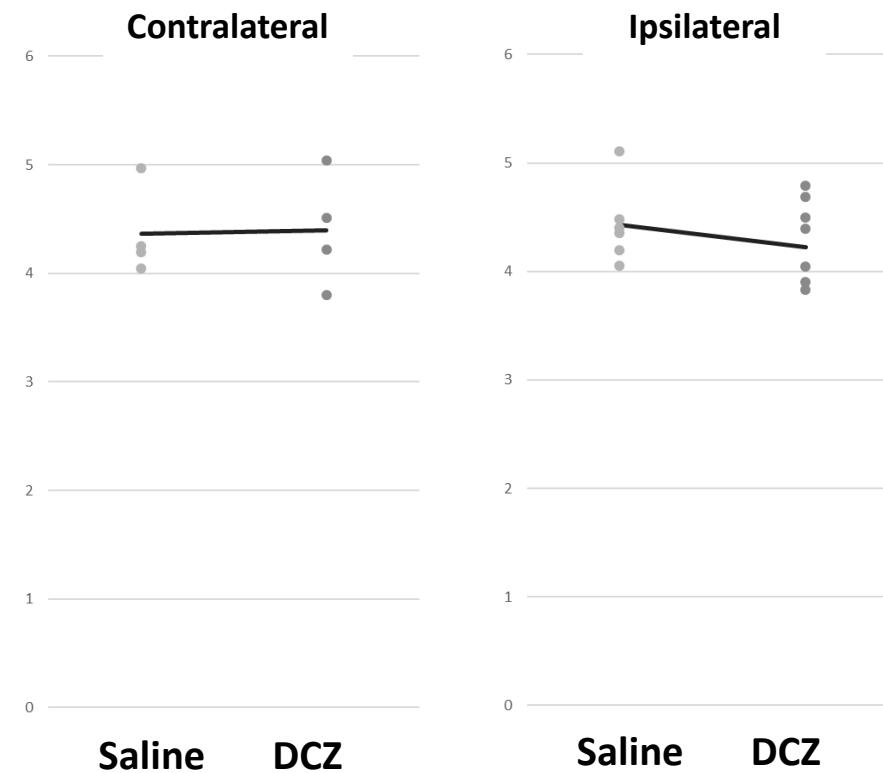
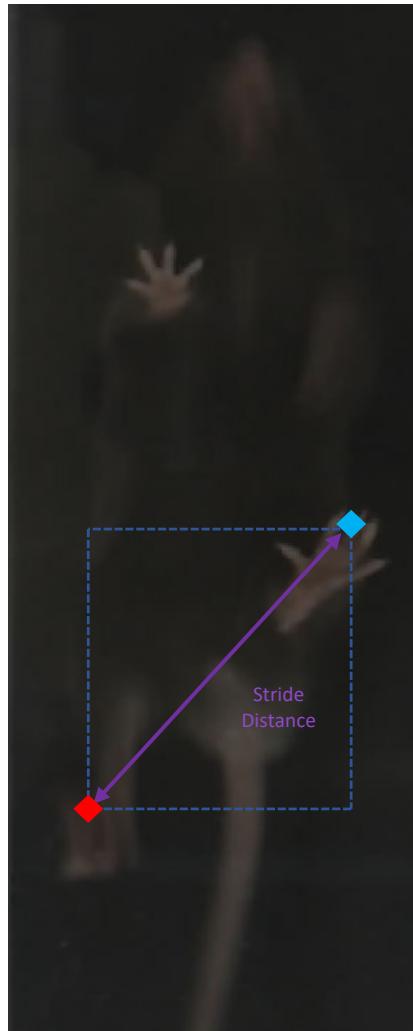
AAV5-CaMKII-DIO-hM4D(Gi)

AAV1-hSyn-Cre



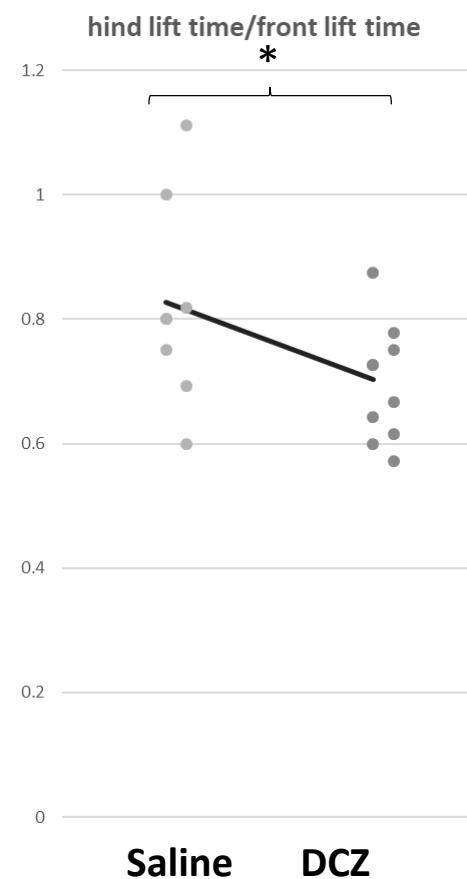
2. DRN-FN Chemogenetic experiments:

Bilateral inhibition of FN glutamatergic neurons did not induce different in stride distance



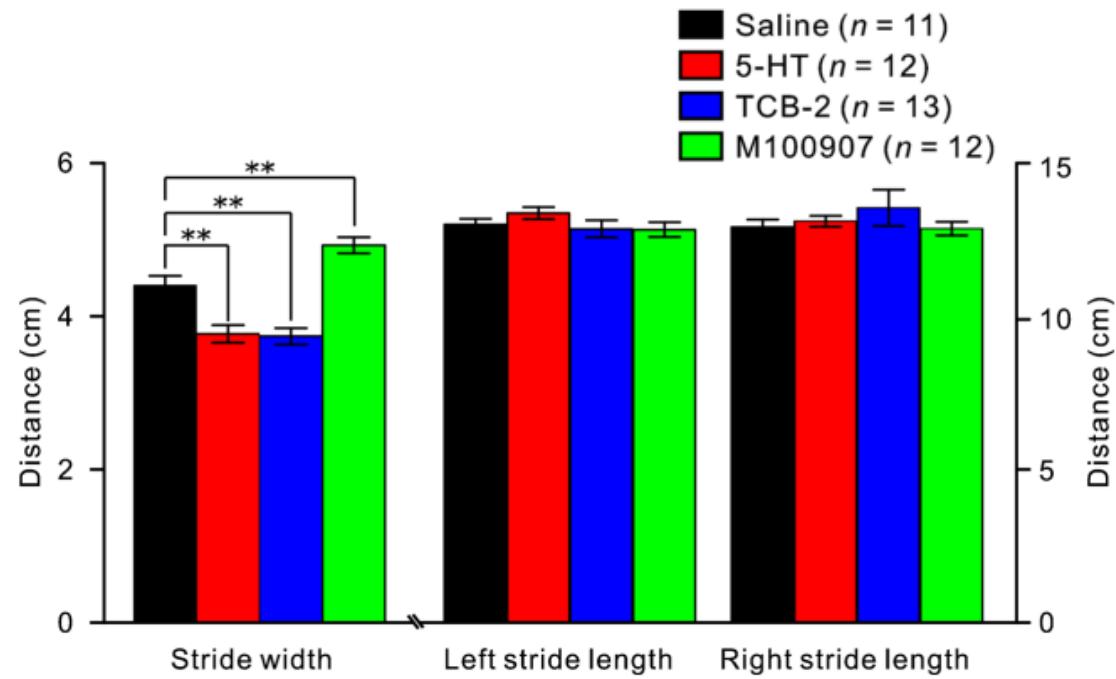
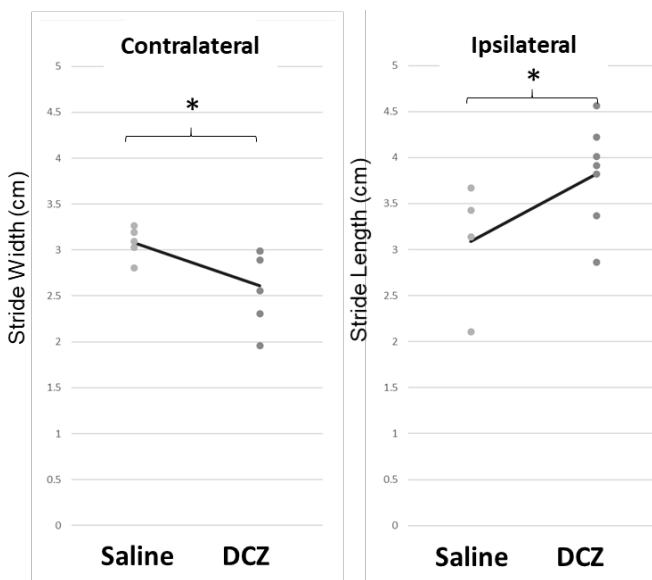
2. DRN-FN Chemogenetic experiments:

However, FN glutamatergic neuron inhibition decreased time that hindlimbs are lifted during strides



2. DRN-FN Chemogenetic experiments:

DRN (Right) excitation showed similar patterns to decrease stride width as previously reported
but not stride length





Outline

1. Conducted Experiments

1. Circuit tracing and cell population

Result: Majority of dorsal DRN neurons projecting to the raphe nucleus are serotonergic.

2. Chemogenetic study

Result: DRN neurons projecting to the fastigial nucleus changes gait pattern.

3. Fiber photometry study

2. Future Experiments

Questions:

1. Could serotonin that regulates motor control from the DRN? **Likely!**
2. What information do these serotonergic neurons encode?



Outline

1. Conducted Experiments

1. Circuit tracing and cell population
2. Chemogenetic study
3. Fiber photometry study

2. Future Experiments

Questions:

1. Could serotonin that regulates motor control from the DRN? **Likely!**
2. What information do these serotonergic neurons encode?

3. Fiber photometry experiments:

Serotonergic neurons show heterogeneous activities to different stimuli

novel mouse



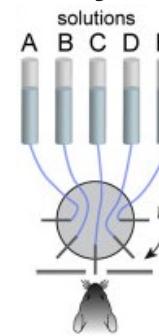
elevated plus maze



foot shock



gustatory stimuli

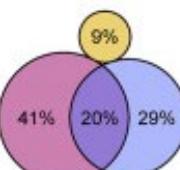


shock
and
sucrose



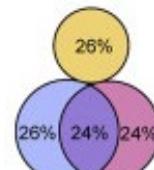
n = 63 cells

open arm
and
sucrose



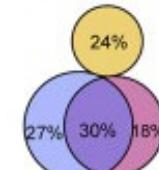
n = 95 cells

shock
and
first interaction



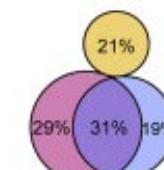
n = 76 cells

shock
and
open arm



n = 66 cells

open arm
and
first interaction



n = 146 cells

first interaction
and
sucrose



n = 94 cells

sucrose day 1
and
sucrose day 2



n = 79 cells

increasing overlap →

3. Fiber photometry experiments:

Five different stimuli given during fiberphotometry recording

Juvenile interaction



Chow food interaction



Novel mouse interaction



Tail picking



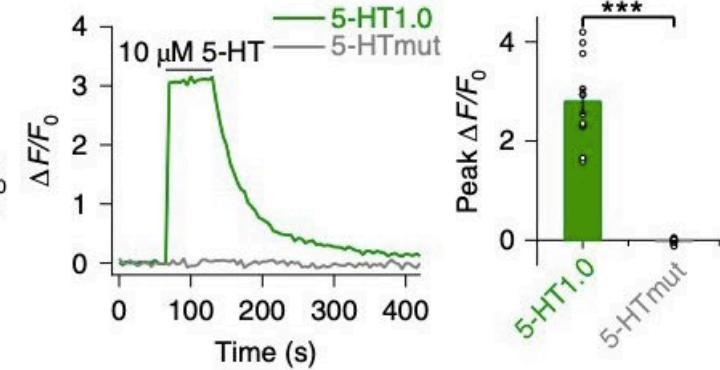
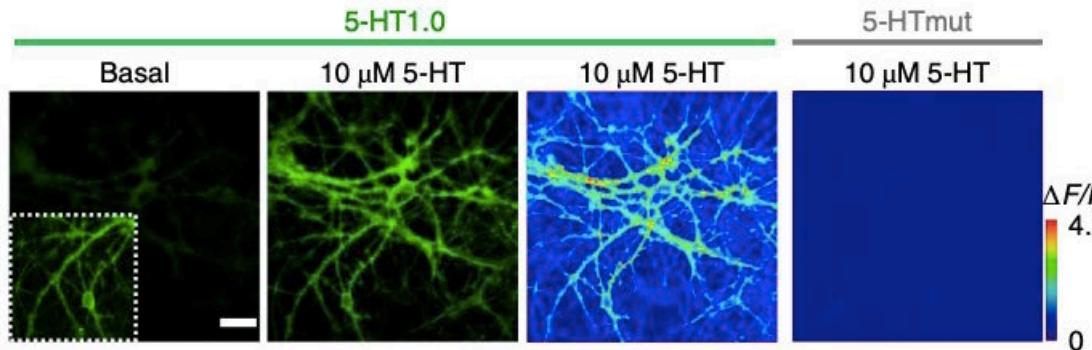
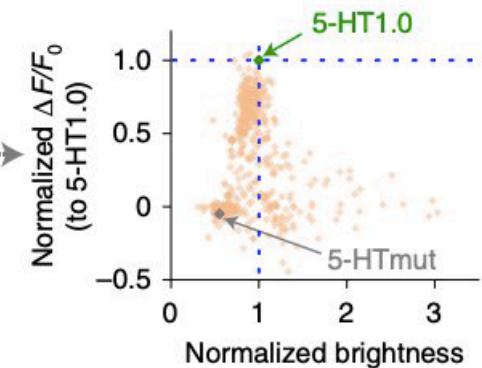
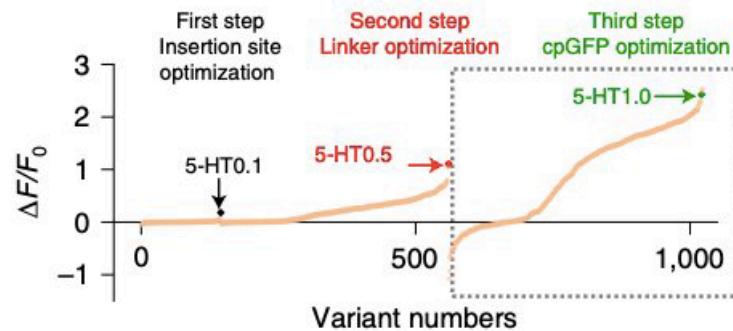
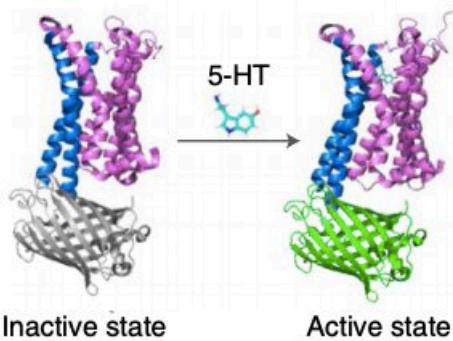
Novel object interaction



3. Fiber photometry experiments:

Use of GRAB 5-HT1.0 for alternative recording method due to low axon terminal expression of DRN axon terminal on FN

HTR2C receptor

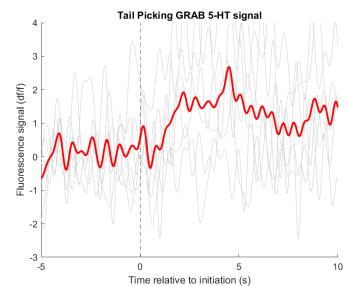
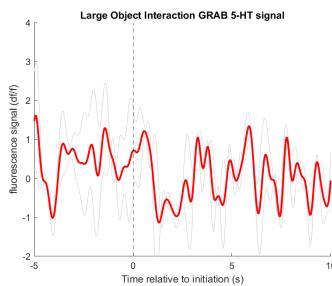
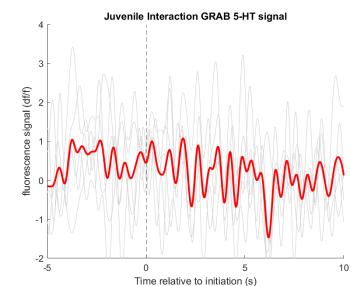
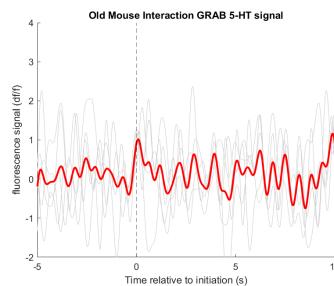
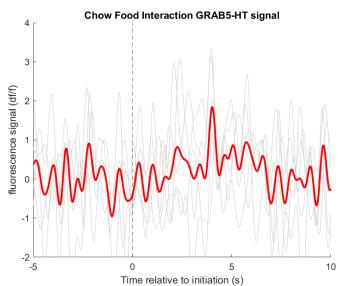
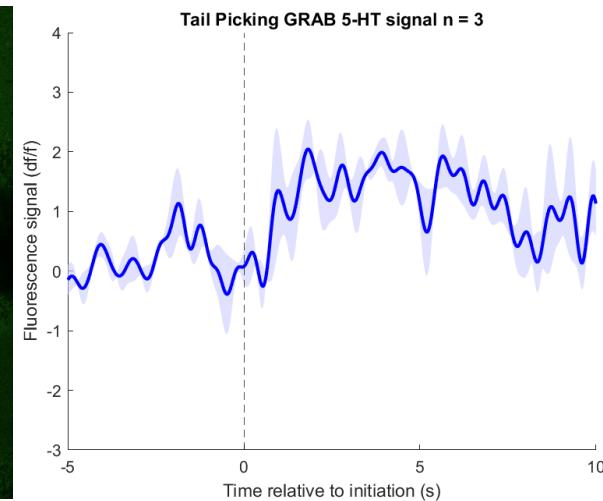
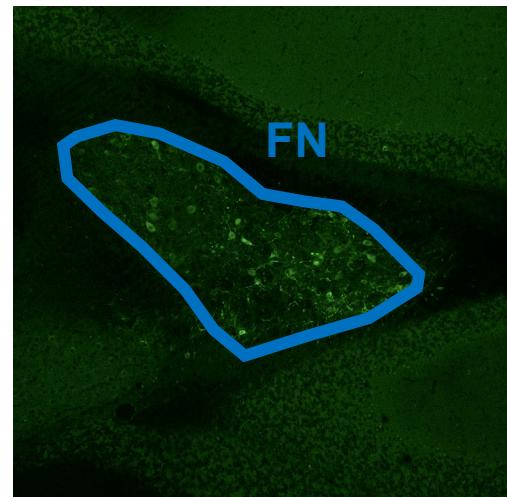


3. Fiber photometry experiments:

Use of GRAB 5-HT1.0 expressed on FN

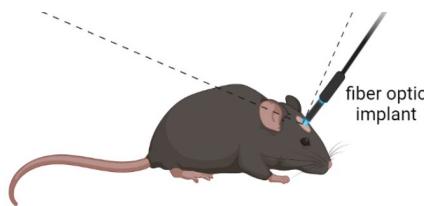


GRAB soma recording
from FN
(B6J wt)

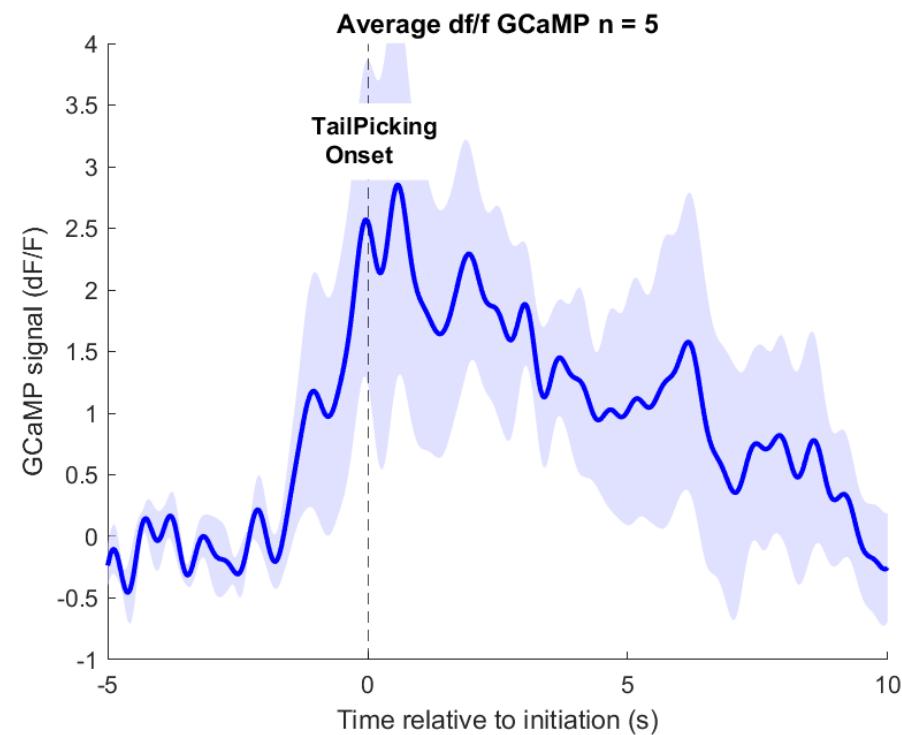


3. Fiber photometry experiments:

Soma recording from DRN by retrogradely expressing AAV9-DIO-GCaMPs6 from Fastigial Nucleus

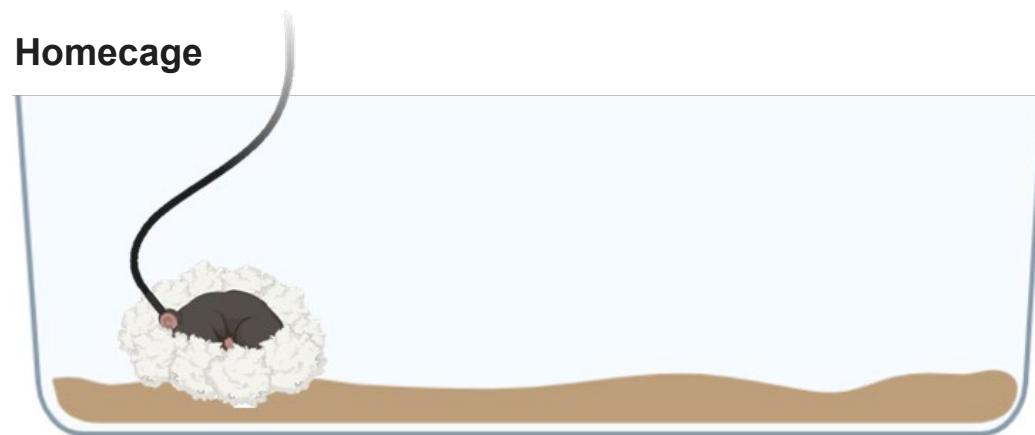


DRN neurons projecting to fastigial nucleus
(B6J wt)



3. Fiber photometry experiments:

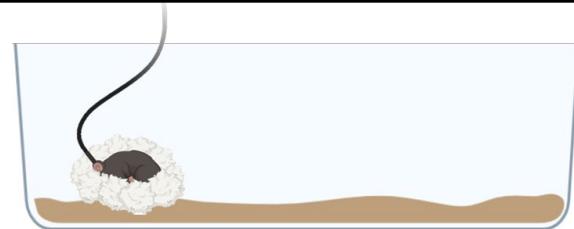
Serotonin activity was recorded home cage to reduce human interference and measure more “natural” behaviors



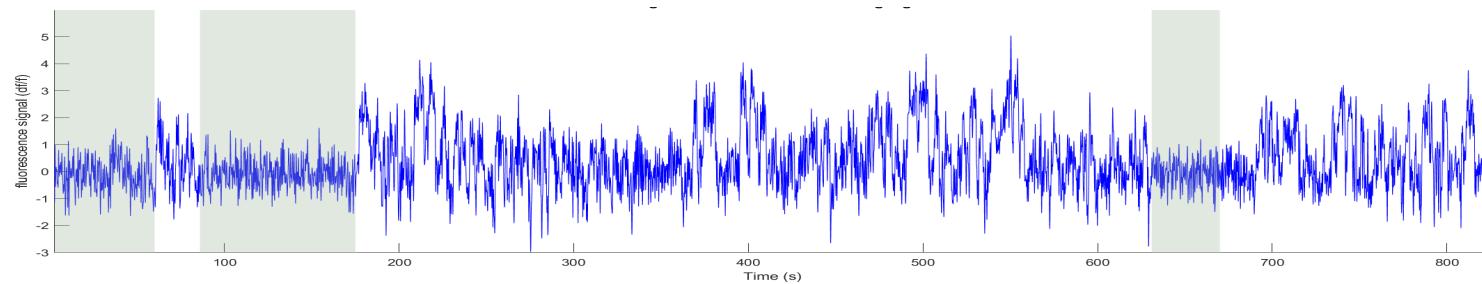
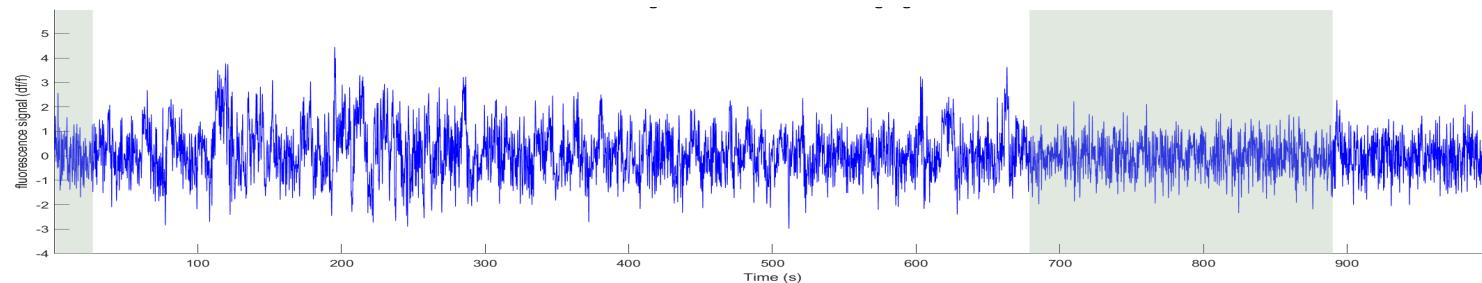
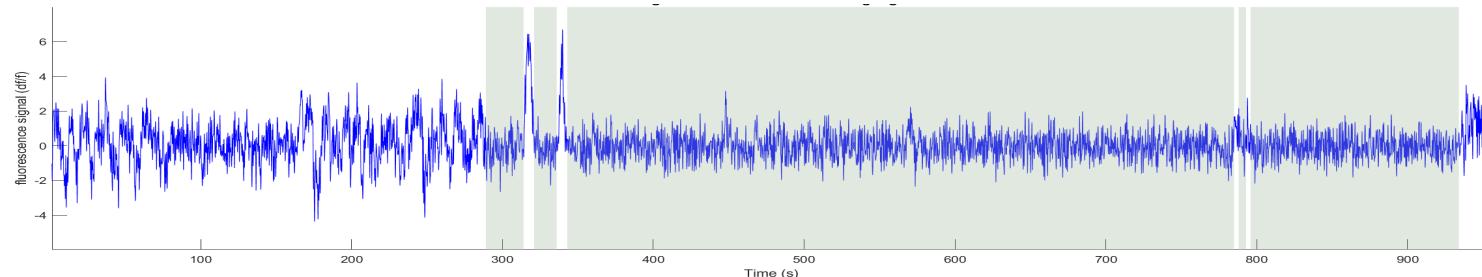
3. Fiber photometry experiments:

Serotonin levels decreased in the fastigial nucleus during sleep periods

Homecage



Sleeping periods in gray



Outline

1. Conducted Experiments

1. Circuit tracing and cell population
2. Chemogenetic study
3. Fiber photometry study

Result: Serotonin released in fastigial nucleus at tail picking session might come from DRN neurons.

2. Future Experiments

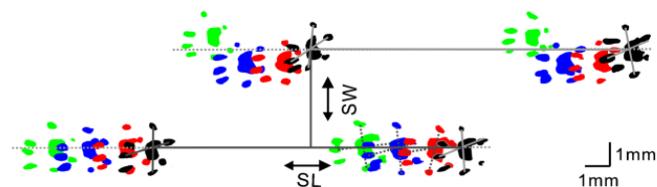
Questions:

1. Could serotonin that regulates motor control from the DRN? Likely!
2. What information do these serotonergic neurons encode? Valence? ☺

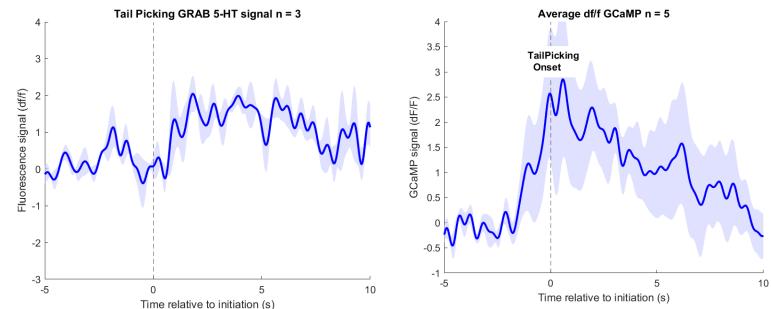
Summary:

DRN neurons release serotonin to the fastigial nucleus for motor control during awake period and tail picking session

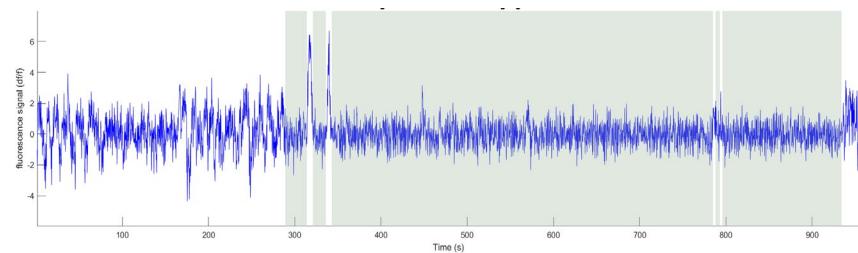
1. DRN activation regulates gait patterns in mice.



2. Serotonin release from the DRN increases in the fastigial nucleus at tail picking.

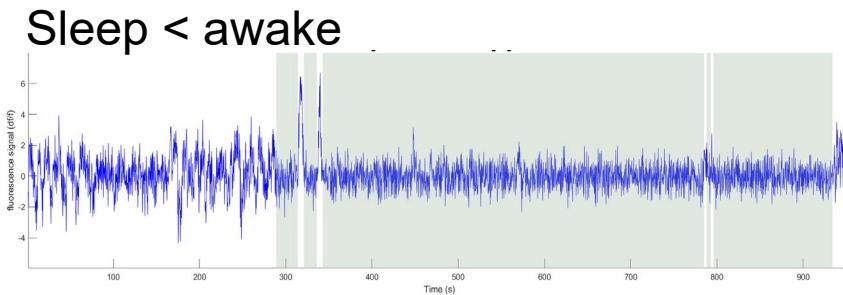
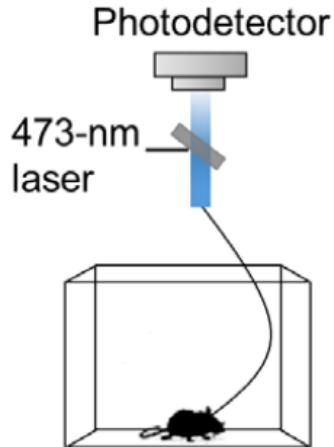


3. Serotonergic activities are silenced during sleep and increased when awake.

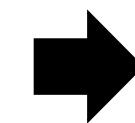
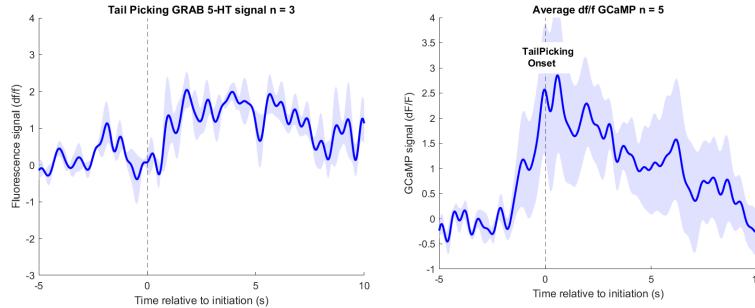


Future plan:

Further recordings of serotonin activities at the fastigial nucleus are necessary to decipher the information encoded via serotonin for motor control



awake < tail picking



Alert?
Arousal?
Threat?

?



Acknowledgement



All BI Lab members



Professor
Daesoo Kim

