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## 🌐 Single-limb fast FR4 operant task (Mendonça et al 2024)

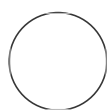
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### ABSTRACT

Single-limb fast FR4 operant task methods from Mendonça et al 2024.

### GUIDELINES

A behavioral experiment control system built around the Micropython microcontroller, was used to control and detect events and supply rewards. Licks were detected using an infrared beam and through a side camera, mouse position in the box was monitored through a camera placed on the top of the box.

### MATERIALS

-14x16 cm custom-built operant chambers placed inside sound attenuating boxes. The custom-built boxes had in their design a retractable lever.

-Sucrose solution (10%) was delivered through the opening of a solenoid (LHDA1231515H, Lee Company). Sucrose solution was delivered through a tube into the magazine (5µl per reward).

### BEFORE START INSTRUCTIONS

Mice were placed on food restriction throughout training, and fed daily after the training sessions with approximately 1.5 -2.5g of regular food to allow them to maintain a body weight of around 85% of their baseline weight.

**Protocol status:** Working  
We use this protocol and it's working

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## Single-limb fast FR4 operant task behavior

- 1 At the beginning of each session there was the onset of a light, and the animals were required to perform a sequence of presses at a minimum frequency in order to obtain a sucrose reward.
- 2 Session 1: To facilitate learning, animal are initially exposed to one session of magazine training where sucrose would be available on a random time schedule.
- 3 There are then 3-4 sessions of continuous reinforcement schedule (CRF) before training, where single lever presses would be reinforced.
- 4 In the following sessions animals were reinforced if they performed a sequence of 4 consecutive presses (Fixed Ratio 4, FR4) in a particular time window (FR4/Xs, fixed-ratio four within X seconds).
- 4.1 The duration of time required to perform the four lever presses was reduced across sessions from 100 seconds to 20 s, 8 s, 4 s, 2 s and finally 1s. To shape animals to use only one of the forelimbs the lever was progressively retracted and the slit through which the forelimb accessed the lever was reduced with a custom-built piece.

## Single-limb fast FR4 operant task (Imaging group)

- 5 Animals perform the task 2 times/session - one with the lever in the left side of the box and the other with the lever in the right - that were randomized throughout the training. Task ended after 30 minutes on

each side or when the animals obtained 30 rewards.

- 5.1** The lever was equipped with a digital 9-axis inertial sensor with a sampling rate of 200 Hz (MPU-9150, Invensense) assembled on a custom-made PCB and connected to a computer via a custom-made USB interface PCB (Champalimaud Foundation Hardware Platform). Lever velocity was extracted from this sensor. Timestamps from the behavioral task were synchronized with calcium imaging data using TTL pulses sent from the behavioral chambers to the Inscopix data acquisition system via a BNC cable.