

## Introduction

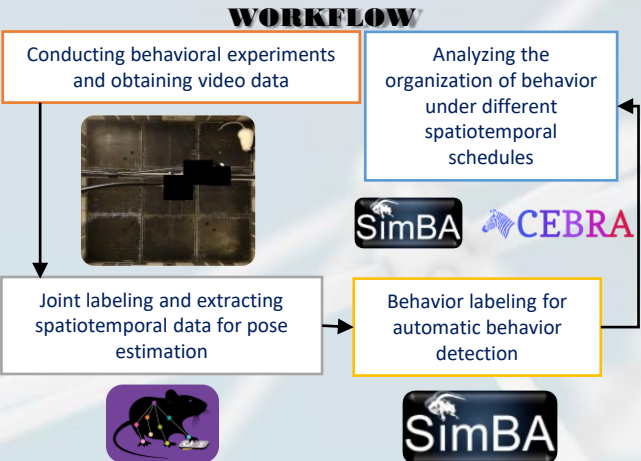
Traditionally, in the field of Experimental Analysis of Behavior, research has focused on discrete responses, but organisms exhibit a wide range of behaviors in natural settings (Skinner, 1966).

Spatiotemporal continuous features of behavior are sensitive to reinforcement contingencies in natural settings (León et al., 2020).

Supervised or unsupervised machine learning offers a method to detect the schedule a subject is following in uncontrolled environments using such data (Lanovaz et al., 2023). Previous studies have shown promise in using machine learning to identify reinforcement histories in animals, particularly in detecting concurrent reinforcement schedules in pigeons (Plessas et al., 2022).

## Objective

The primary objective of this paper is to employ machine learning techniques to analyze various behavioral organizations under spatiotemporal schedules in 12 rats, using pose estimation through deep learning algorithms.



## Method

### Subjects:

- 12 experimentally naïve Wistar rats, three months old at the beginning of the experiment, housed individually, and under a 23-hour water restriction with free 30-min access at the end of each session.
- Food was freely available in their home cages. Sessions were conducted daily, seven days per week.



### Apparatus:

- The experimental chamber was 92 cm wide, 92 cm long, and 33 cm high. Each wall had a liquid dipper (Coulbourn E14-05) 2 cm above the grid floor, providing 0.1 cm<sup>3</sup> of water for 3 s. Head entry detectors (MED ENV-254-CB) identified entries at the four dispensers.
- A buzzer in the upper central part of the chamber signaled water availability. MED PC IV software, connected to an external computer, recorded water deliveries, as well as head entries. A video camera (Topica TP-505D/3), 1 m above the chamber, recorded subjects real-time location.
- Softwares used are DeepLabCut, SimBA, and Cebra under Python (Goodwin et al., 2024; Lauer et al., 2021; Schneider et al., 2023).
- Computer components include an intel 12700h processor, 32gb of ram and a RTX 3060 6gb mobile GPU.



### Procedure:

- Subjects were divided into four groups and exposed to different schedules involving fixed time (FT) and variable time (VT) with fixed (FS) or variable space (VS) for water delivery. The rats experienced 30 sessions in the first phase and 10 sessions without a programmed schedule in the second phase.

### Training and model:

- The DeepLabCut model was trained from a pre-trained mouse model using 27 joints available to everyone for mouse or rat pose estimation (topview\_mouse).
- We refined the model using 4 rats, 36 videos of 20 minutes, and 218 labeled pose estimation frames.
- Following 4 training sessions of 170 000 iterations under ResNet 50 (Convolutional neural network CNN), the final model allows to reliably detect the rats in our videos as shown in Figure 1.

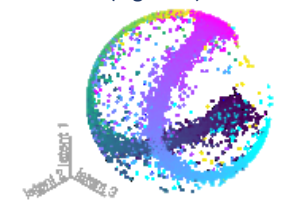
### Measures:

- The principal measure comes from DeepLabCut, which provides joint coordinates (X, Y) and spatial relationships at 14 Hz, as shown in Figure 1.
- SimBA adds measures like velocity, direction, and time spent in zones. For training, we labeled 84,000 frames from 4 rats across 5 videos lasting 20 minutes.
- Behavior classifiers include locomotion, rearing, head-dip, grooming, immobile sniffing, and immobility, with their frequency and spatiotemporal distribution recorded.

### Analysis:

- SimBA offers ROI analysis with metrics such as velocity, direction, time spent in each zone, location heatmaps, and path plots. It visualizes and analyzes behaviors under different reinforcement schedules using DeepLabCut (DLC) data (Figure 2).
- CEBRA provides robust machine learning analysis using supervised and self-supervised model training. It offers position-hypothesis-driven embedding and integrates with DeepLabCut pose estimation data for analysis under different reinforcement schedules (Figure 3).

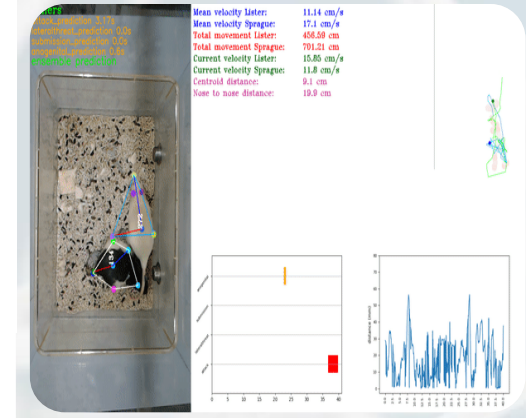
**Figure 3**  
 Latent behavior analysis using Cebra



**Figure 1**  
 Joints labeling on a video using DeepLabCut



**Figure 2**  
 SimBA position, directionality, behavior, velocity analysis over time and space



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