WHITE PAPER

Acquiring simultaneous *in vivo* calcium imaging and behavioral recordings

Powerful partners: syncing nVista™ and Noldus PhenoTyper®

With nVista and PhenoTyper you can:

- Produce synchronized population-level neural activity data with behavioral videos during active behavioral sessions.
- Obtain longitudinal neural circuit and behavioral recordings of the same animal.
- Filter data for specific behaviors, locations, and times.
- Adapt the synced behavioral and neural activity system setup to address diverse research questions.

Introduction

The ability to acquire *in vivo* neural activity data simultaneously with behavioral data is vital to truly understanding the brain in action. The nVista miniature microscope system from Inscopix enables researchers to conduct genetically targeted calcium imaging in hundreds of cells over time in awake, freely behaving rodents, and has provided insights from multiple brain areas, from the prefrontal cortex all the way to the hypothalamus. nVista's compact, sleek plug-and-play interface makes it easy to add powerful new neural circuit insights to any laboratory's behavioral research arsenal.

The Noldus PhenoTyper system is a proven behavioral observation platform which obtains behavioral data during multiple types of experimental paradigms, integrating the animal test environment with video equipment and automated animal tracking. nVista is fully compatible with the PhenoTyper system, allowing you to harness these two data powerhouses in the service of your research questions.

There are multiple ways to sync the nVista system with the Noldus PhenoTyper depending on the behavioral paradigm that best fits your research question. Here, we detail an example of triggering nVista recordings in an implanted animal when the animal crosses into a target region of the arena, as defined in the EthoVision® XT software.

Setting up nVista

Hardware

To use the Noldus PhenoTyper to trigger the nVista system, attach a male BNC cable to the nVista Trig port (*Figure 1*). The other end will be connected to the RJ45 port of the PhenoTyper system.

Figure 1. Back of nVista box

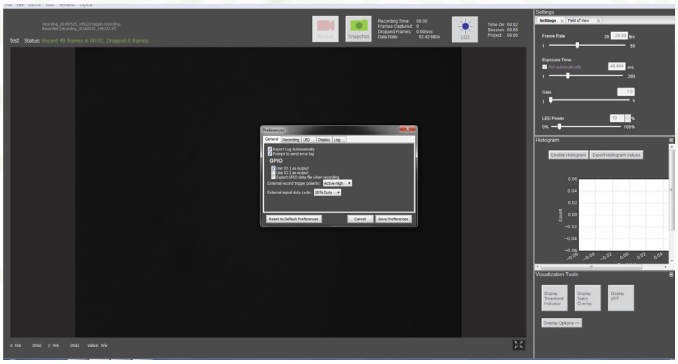


Figure 2. Image shows nVista Preferences and Settings.

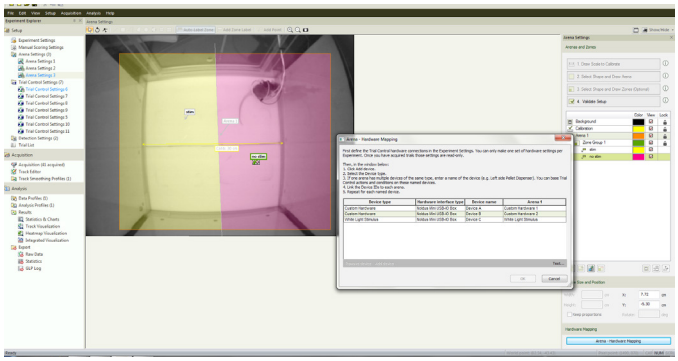


Figure 3. Image shows Arena Settings box bound in EthoVision XT software.

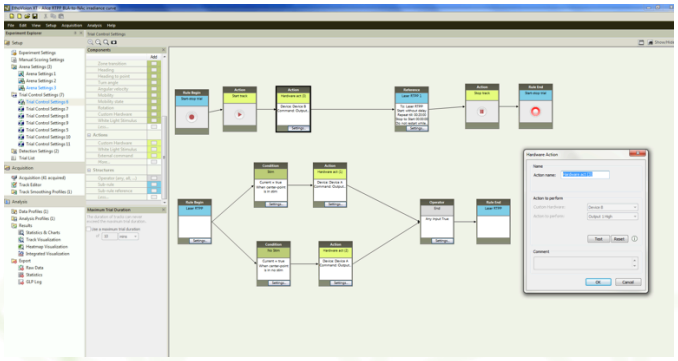


Figure 4. Image showing Trial Control Settings in EthoVision XT software.

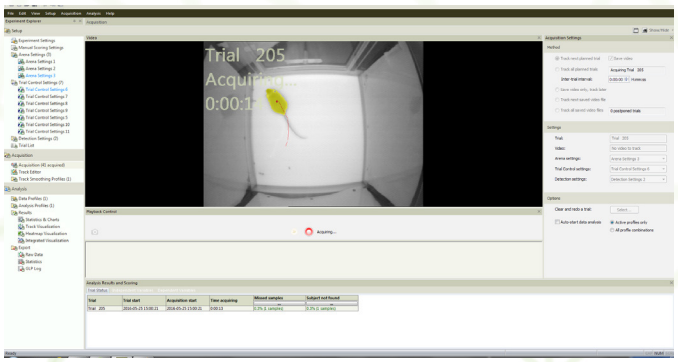


Figure 5. Snapshot of a trial acquired with EthoVision XT software.

Software

Open the nVista Acquisition software. Under the Device menu at the top of the software interface, select **Trigger from external hardware**.

Configure your other nVista settings, such as image acquisition format, GPIO status, LED power, etc, as needed based on your experimental needs (*Figure 2*). The EthoVision software will control when the recordings start and stop.

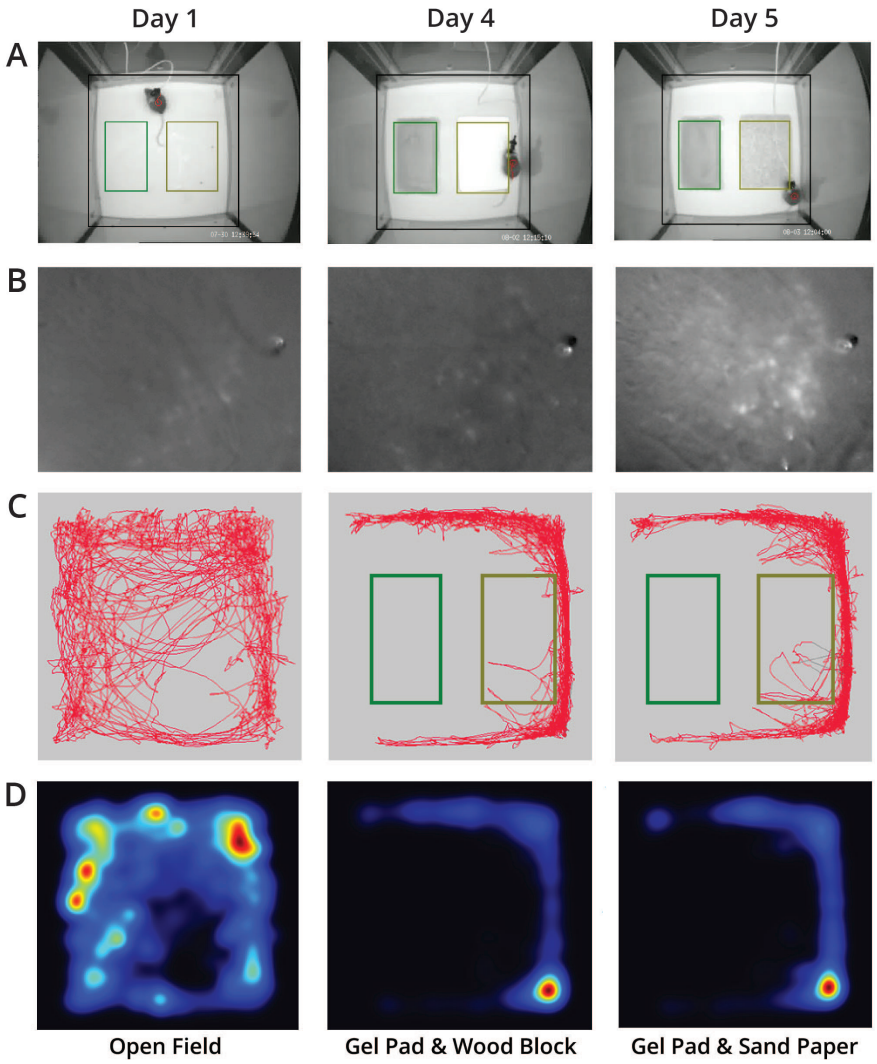
Under Preferences, select whether or not you would like the GPIO port to be toggled On or Off, and whether or not it should be an input or an output.

To set the nVista system to record only when the animal has entered a specific part of the behavioral box arena, select **Arena Settings** in the Experiment Explorer.

Set up the arena to satisfy your experimental needs. To ensure the correct hardware device will be used during the experiment, scroll down to Hardware mapping from the Arena Settings window on the right, and make sure the Arena Hardware Device type, interface and name are linked to the proper Arena (please refer to the EthoVision XT manual for additional instruction).

Set up the Trial Control Settings (*Figure 4*) to dictate when the Noldus system will communicate with nVista and trigger a recording to begin or end. Shown here is the visual experimental trial controls for triggering an nVista recording every time the animal approaches a target region in the behavioral box (*Figure 5*).

To start the experiment, select the **Arena Settings > Trial Control Settings > Acquisition**, and begin acquiring data under the Playback Control window.



A. Cortical activity recording and behavioral tracking of the animals was performed on animals across 3 days for 20 minutes each day using nVista and shown through the EthoVision XT software interface. The animals were placed in an arena devoid of any objects on Day 1. Two texturally different objects (Gel Pad and Wood Block) were placed in the arena for 3 consecutive days and the animals were imaged on last day of object familiarization (Day 4). The following day (Day 5) one of the objects was replaced with a novel object (Sandpaper) and the animals' activity was recorded.

B. nVista snapshots from three days at different time points show mouse somatosensory cortical activity.

C. Mouse tracks over the three days of imaging are shown here computed using EthoVision XT.

D. Respective heat maps of the mouse's location extracted using EthoVision XT.

Figure 6. This representative figure combines nVista data with EthoVision XT data where the animals were imaged across 3 days for 20 minutes each day.

Summary

The ability to gather population level neural activity data in the context of awake behavior is proving essential for gathering meaningful insights in an increasing number of innovative neuroscience applications. The nVista miniature microscope system provides new insights into different cell types in a variety of brain regions; used in concert with the Noldus PhenoTyper system, this powerful pairing empowers you to bring firing rates into the context of behavior, and take your neural circuits research projects to the next level.

iNSCOPIX

Our solutions are designed to help you map neural circuit dynamics and ultimately advance the understanding of the human brain.

We care about the quality of your data, the impact of your research, and the direction of your field, because we're right there with you in the pursuit of neuroscientific knowledge.

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2462 Embarcadero Way
Palo Alto, CA 94303
888.874.6458
info@inscopix.com

www.inscopix.com