Webinar Q&A Report:

Turn Away from Traditional Tethering and Towards a Better Method for Data Collection

Q: Can you perform behavior tasks in the Raturn? Which ones?

A: It is possible to perform some behavior tasks in the system. Some researchers have used it for novel object recognition. However, this is not a behavioral monitoring system, per se. The system is capable of capturing activity when used with the <u>Animal Activity Analysis</u> system. This is primarily used for looking at changes in activity levels. For example, differences between nocturnal and diurnal activity or differences in activity following administration of a compound. The system should never be used for studies that require two animals in a single cage.

Q: Does the Raturn record its own movement?

A: When used with the <u>Animal Activity Analysis</u> system or the <u>Culex Automated Blood Collection</u> <u>System</u>, the left rotation, right rotation and rearing activity can be collected. Because of the way the system works, the animal has 270 degrees to move where no cage activation occurs, and therefore no activity data can be collected. This means that it is not a tool for all behavioral studies, but it can be helpful for determining overall activity levels of the animal.

Q: Can you house multiple mice?

A: No. Each system is for a single animal. Because the animals are tethered and have implanted catheters or probes, it would be dangerous to house more than one animal in the system unless under conditions of strict supervision for very short periods.

Q: How long can animals stay in the system?

A: This depends on the protocol and individual IACUC oversight. Generally speaking, animals do not stay in the system for more than 2 weeks continuously. For some studies, a few days is all that is needed for completion of the study. For longer term studies (several weeks or months) it is recommended that the animals are connected during sampling periods and returned to a home cage during non-sampling periods. However, there is no technical reason that mice could not remain in these cages indefinitely.

Q: How is the animal exactly tethered to or connected to the system?

A: The animal is connected via a tether that is mounted to the balance arm. This tether has a spring mechanism that ejects a hook when pressed by the researcher and hides the hook when not being pressed. The hook can be connected to a simple collar, a loop on a head mount, or a harness. BASi is also working on a modified tether/balance arm that will work with a spring coil tether for use with implantable buttons, such as these. These attachments are made with easy magnetic fittings.

Q: If you can have a second 'companion' mouse in the cage, would that mean you could potentially evaluate social interaction behavioral paradigms? e.g. novel/familiar individuals, anxiety indicators.

A: It is not recommended to use this system for any studies that require social interaction. As noted above, animals are tethered and have implanted catheters or probes. Placing a second animal in the cage creates a high likelihood that these implants will be damaged.

Q: Do the electronics of the cage create electrical interference/noise in EEG recordings?

A: John answered this in more detail during the webinar, but this does not seem to be an issue for most types of electrical recordings. If the room has substantial electrical noise, or if the electrical current you measure is very small, you can wrap the Raturn and lid in copper mesh to create a self-enclosed faraday cage. In addition to wired recordings, the system has been used for telemetry recordings and no issues were uncovered with regards to noise or cross-talk between systems.

Q: How would one utilize this technology at scale? ie. multiple animals being studied in parallel?

A: The systems can be configured for multiple lab spaces depending on your needs. There is an open benchtop system, as well as a <u>cube system</u> that keeps instruments closer to the animal. The cube system can be customized with various <u>shelves</u> and <u>wheeled carts</u>. These options make it possible to fit the systems into a variety of spaces. In Dr.Cirrito's lab, the systems are mounted on wire shelving carts. In other labs, we have provided <u>4-station carts</u>. Most customers have 4-12 units to run their studies. In BASi's Contract Research Lab, we have 48 stations that can be run simultaneously with 3-4 technicians. In Dr. Cirrito's lab, he mentioned that they have multiple units, but some are designated for acclimation while others are actively running studies. The number you can run at any time is a product of the number of technical staff you have as well as the complexity of your studies. For simple blood collection 2 people could run a 48-animal study without much trouble. With something more complicated, you may wish to have a 1 person per study group ratio. When Dr. Cirrito's group is performing microdialysis, they find that running 1-4 mice at a time is comparable effort, but that starting more than 4 mice at a time is much more difficult.

Q: In general, are you seeing an increase requirement for hands-free data collection and a higher importance on reduced animal handling, intervention, etc. Is this an important factor that may influence data acceptance and success in publication?

A: I have not seen any indication of an impact on success in publication, but this system does make it possible to uncover data that gets masked by human involvement. Additionally, getting better data or more data from an individual animal does provide an opportunity to uncover something new that is worth publishing. But outside of that, there is a changing tide when it comes to animal research, affecting both the public opinion and has also had an impact on research approaches. For public opinion in general, any time that researchers can share a story about how we're getting better data with less stress then we're helping stem the flow of PETA-inspired bad-mouthing of the animal research community. For the effect on research overall, I would recommend a quick review of this article: https://www.nature.com/articles/laban.1224 and some of the accompanying articles from that lab animal issue as a case in point.

Q: Is there a reasonable way to monitor timing of food intake when a mouse is in the system?

A: It is possible to monitor food intake, but it isn't always easy. For studies with a head-mount, we recommend our <u>Stackable Cage</u> because it has no corners/accessories for the mount to get stuck on. For this cage, food must be placed on the floor and that makes it extremely difficult to measure with any accuracy. However, we do provide a <u>Universal Cage</u> that has food hoppers on the outside (for rats). This cage makes it much easier to measure food consumption. Neither of these options is as accurate as a true metabolic cage which often uses weight sensors to measure consumption of food.

Q: What habituation time is recommended when using rats?

A: This depends on the type of study being run. For a short-term blood sampling study, such as for pharmacokinetics, it is common to place the animal in the system on day 1 and sample on day 2. For a study that is stress sensitive, it is common to acclimate the animal for several days. When it comes to stress hormone release in blood or dialysates, most animals return to a baseline level within 2-4 hours. With activity levels, most animals return to baseline levels within 2-4 hours, however, it can be helpful to give them a full overnight (active) period before starting the study. For our clients that have performed extremely stress sensitive studies (heart rate, blood pressure, body temperature) it was found that 3-5 days of acclimation is required, but that this was the same whether they were put in a clean shoebox cage or into the Raturn cage. See this source for details.

If you have additional questions for <u>BASi</u> regarding content from their webinar or wish to receive additional information about their products and laboratory services, please contact them by phone or email:



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