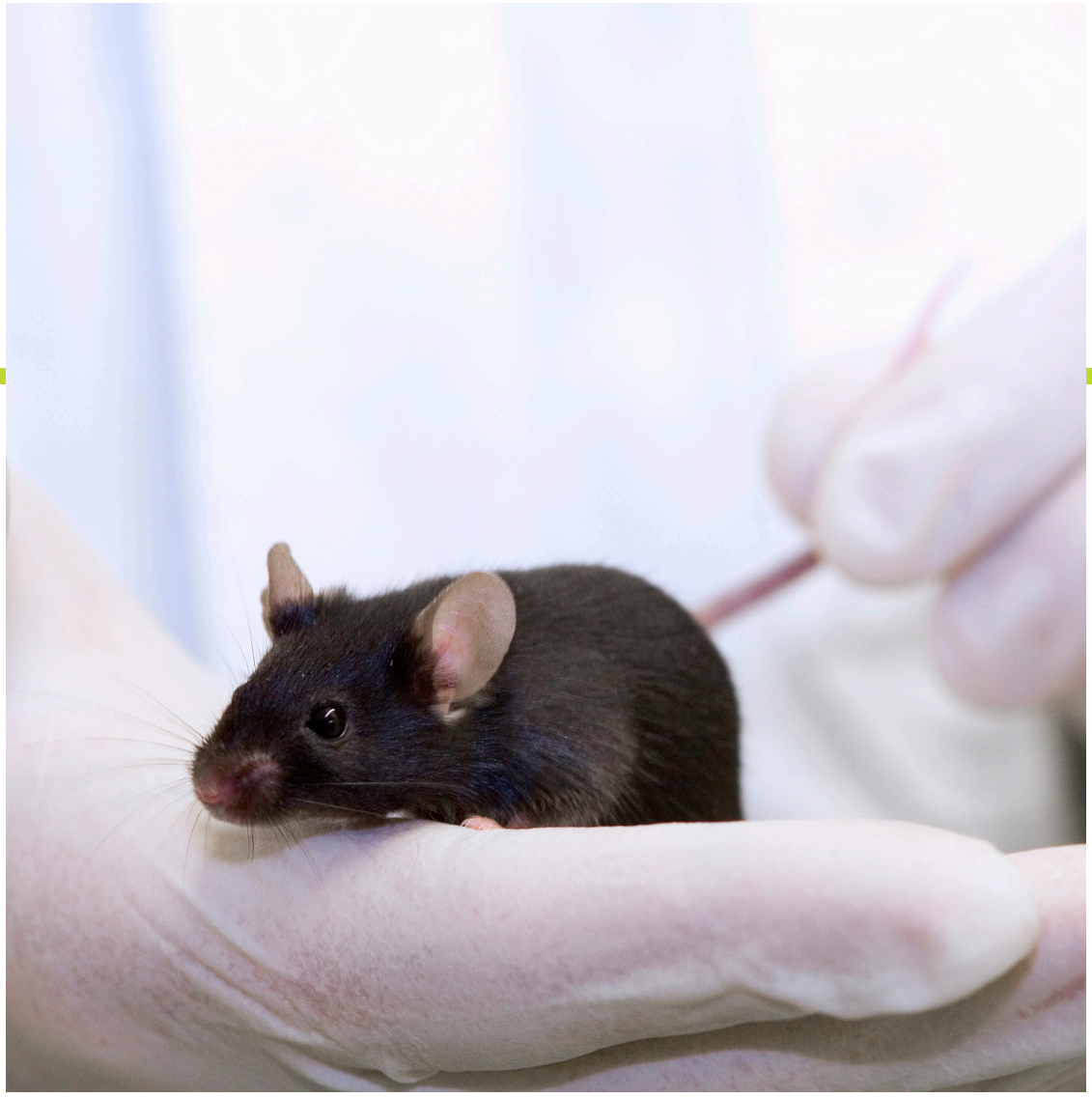


CatWalk XT & TBI mice

How to use CatWalk XT effectively for your own TBI models

By Marcella Cline and Dr. Donna Cross, PhD

A white paper by Noldus Information Technology

USING CATWALK XT EFFECTIVELY

The CatWalk XT gait analysis system can be an incredibly useful tool in assessing the subtle motor deficits exhibited by mice post-TBI, making it ideal for detecting deficits that might not be readily observable to researchers. But how can you use the CatWalk XT effectively for your own TBI models? We have a few tips!

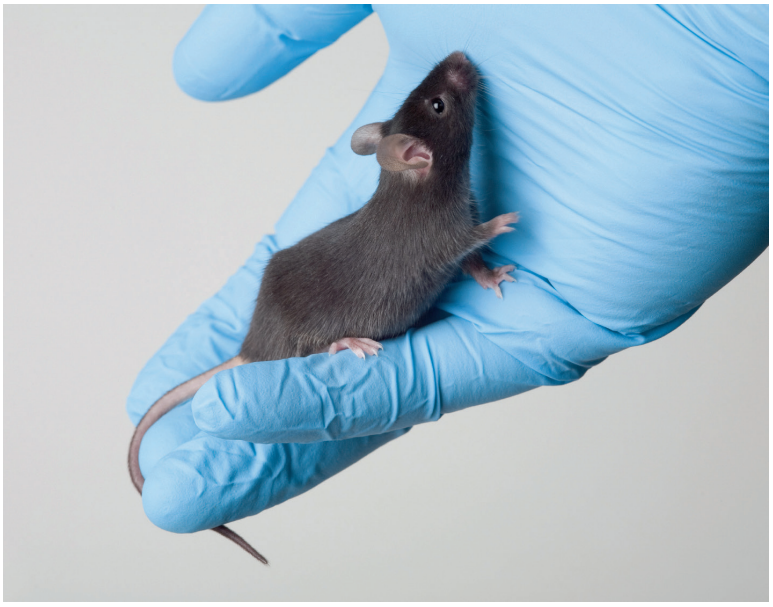
- Handle your mice often
- Practice makes perfect
- Keep the testing area free of smells and sounds
- Consistency is key
- No motivation necessary
- Use the CatWalk XT software auto classify option - but double check your results
- Timing makes all the difference in TBI

7 TIPS

HANDLE YOUR MICE OFTEN

As prey animals, mice are typically timid creatures, and any additional stress exposure has the potential to confound research results. We fully recommend that, prior to any behavioral testing or surgeries, you take the time to handle your mice in order to familiarize them with human contact. A mouse inured to being picked up, held, and simply being around researchers will be far less anxious during subsequent behavioral testing than a naïve, unhandled mouse.

It is also important to make a note of any anxiety-related behaviors observed in cages (fighting, barbering, excessive grooming), and take steps to intervene when necessary.



It is also important to make a note of any anxiety-related behaviors observed in cages (fighting, barbering, excessive grooming), and take steps to intervene when necessary. If fighting is observed, try to isolate and remove the aggressor from the cage.

PRACTICE MAKES PERFECT

Before conducting any testing using the CatWalk XT system, it's important that the researcher familiarize themselves with the equipment. The CatWalk XT software is fairly intuitive to use, and it shouldn't take more than a few trials to feel confident with the program. Take the time to run a few test runs with practice mice to ensure you have a decent understanding of how to operate the apparatus, how set up your testing protocols, and how to go through the menu screens.

KEEP THE TESTING AREA FREE OF SMELLS AND SOUNDS

Mice rely heavily on their olfactory sense and can find smells (especially those of unfamiliar mice) distracting during testing. Make sure you properly clean the CatWalk XT walkway between animals to minimize distracting odors. Lightly spray the walkway and walls of the apparatus with 70% ethanol and wipe down with a clean paper towel to ensure your subject doesn't get way laid by an unusual scent.

Keep talking and noises to a minimum during testing. Mice instinctually freeze when frightened, and might stop momentarily during a run if startled.

CONSISTENCY IS KEY

Find a testing protocol that works for your research group and stick with it! Making sure that each and every animal undergoes the same treatment during testing will help minimize inter-subject variability, and produce better results. This includes testing at similar times of the day across groups, as general locomotion in mice can be highly dependent on their day/night cycle.

Make a protocol template on the CatWalk XT software, and use this template for all of your testing to ensure software and acquisition settings are not inadvertently changed between groups.

> Collecting multiple consecutive runs per mouse, and averaging these runs for analysis, is another great way to help reduce potential variability.

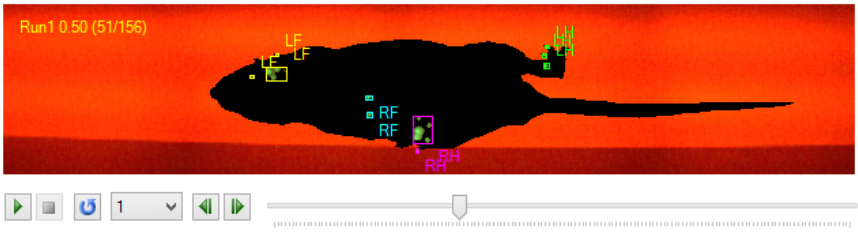
Collecting multiple consecutive runs per mouse, and averaging these runs for analysis, is another great way to help reduce potential variability. Acquiring three runs per subject takes just a few extra minutes, and can make a great deal of difference to getting an accurate picture of that subject's general gait.

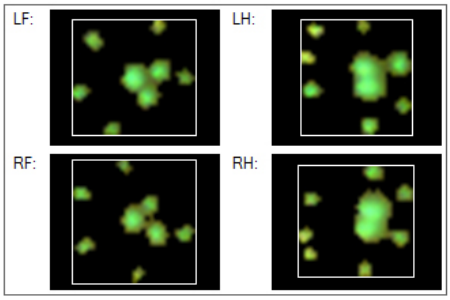
NO MOTIVATION NECESSARY

Food motivations, by in large, are not necessary to motivate mice to cross the walkway; all that is necessary is a bit of patience. Before placing your mouse on the glass top, block the ends of the walkway so your subject cannot escape (be sure these blocks are out of view of the camera, for easier data classification later). Place your mouse on the walkway, out of range of the camera, and start the acquisition. When a mouse crosses the walkway and reaches the dead end, it will instinctively turn and continue back down to the other side.

USE THE CATWALK XT SOFTWARE AUTO CLASSIFY OPTION – BUT DOUBLE CHECK YOUR RESULTS!

One of the advantages of using the CatWalk XT over other tests of motor function, is the relative degree of observer-independent assessment and analysis this system offers. Once your data has been acquired, the CatWalk XT software can take care of much of the data analysis on its own – including classifying individual paw prints.





However, while the auto classify option is a great time saver, be sure to review each run manually to ensure proper classification. Often, TBI mice step in such a manner that two paws might fall directly next to each other on the glass plane. This can sometimes confuse the classification software, resulting in an incorrect labeling of one or both paws. Another common source of confusion for the classification software is the appearance of fecal bolus on the glass walkway when a mouse defecates during a run. Always review the classified runs to ensure that no mistakes were made in the classification step.

TIMING MAKES ALL THE DIFFERENCE IN TBI

Timing is critical when assessing TBI symptomatology - testing mice at an inopportune time (too soon following TBI surgery, or waiting until deficits are no longer above the threshold for measurement) might result in missing a crucial timing window during which symptoms are detectable.

CatWalk XT has been used to successfully distinguishing between sham and TBI mice at 7 days post-injury (see recent publications). However, as motor deficits are highly injury dependent, you may need to adjust this time point to suit your injury model. Be sure to pilot test various time points post-TBI to ensure differences can be detected between sham and TBI subjects.

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