

Electronic von Frey and filament testing of mice

Dos and don'ts, from Topcat Metrology Ltd

These notes have been compiled by Dr Michael Dixon and Dr Polly Taylor, the inventors of the MouseMet EvF system (www.mousemet.co.uk). They are a summary of experience gained over 25 years with many species and are applicable to most types of mechanical threshold testing equipment (and to all species!).

Get Comfortable

The environment is crucial, for the mice and for you. You are going to test many times. If your back aches from crouching because the runs are at the wrong height, or your arms tremble from holding an awkward piece of equipment, your data will be poor. Spend time getting the bench, the runs and your chair at the right height. We've worked in rodent facilities, and we know this is easier said than done, but trust us; it's worth it.

Then get your mice as comfortable as possible. Cage floors vary, and if possible, choose one that the mice seem comfortable standing on, while still leaving the plantar surface accessible. Some cages have grids as a floor, some have bars. Some have a thick frame round the edge which means you can't look straight at the foot from the side. This is not helpful. Ideally you want transparent sides with no obstructions to your view.

Remove all distractions... or as many as possible. Busy labs, with people walking past, doors slamming, phones ringing etc are really bad. Ideally you want a quiet room with a low but constant noise level. Playing a radio in the background is ideal as this will cover up any clicks etc as you operate the equipment or your computer. Similarly, if you have someone in the lab working with you, make sure they don't stop talking every time you start a test. Your mice will learn! The more mice you can test at one time the better, as you will then have more choice as to which to test next, and the fidgets will have more time to settle. But try to test away from other colonies, to reduce the distraction from cage rattling, etc. And don't even try to test while animals nearby are being fed or cleaned out!

Pick your Moment

Timing is important. Allow sufficient acclimation time before starting to test. A mouse in a new environment will explore. There's no point in testing while he is doing this. Similarly, mice that have been in the same place for too long will start to doze off. This will raise their thresholds.

So pick your moment, and test when the mouse is calm but attentive. Most testing cages don't have food or water, but other distractions as listed above will tend to raise the thresholds. Don't get too hung up on the theoretical time points. A good test 10 minutes early is better than a bad one at exactly the right time. And, as we said above, the more mice you can test as a group, the better. That way you can always choose the mouse that's in the right mood.

Try to be consistent about the rate of application of the stimulus (if you are applying it manually), but don't worry too much about it. A good reaction at a slightly high or low rate is better than a poor or missed reaction because you were concentrating on some guide lights. Watch the mouse during the test and nothing else. With practice, your rate will become consistent anyway. The ideal rate is a compromise between a number of factors; too fast and the reaction point is blurred by the time it takes both you and the mouse to react. Too slow and there is more chance of the mouse walking off or being distracted during the test. We recommend applying the stimulus at the minimum level for 1-2 seconds and then a ramp at 1gf/sec.

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Your mice are all different

Mice are individuals so their reactions will be different. Don't expect a whole cohort to react in the same way. Keep a record of reaction behaviour (leg lifts, leg shakes, attacking the probe tip or filament) so you can reject outliers later with some logic. You won't remember afterwards that someone opened the door during a particular test.

Animals learn. Don't worry if your first baselines are scattery. Keep going, as far as you can within the protocol, and they will get tighter. It's a new sensation for the mice and they don't know what to do with it at first. We have found, with all species (right up to camels!), that baselines get tighter quite quickly. It's very common, with naïve subjects, to get a high proportion of "touch-on" responses where they respond as soon as they feel the application of the filament or tip. Soon, they will learn to ignore that, and then only respond as the stimulus becomes noxious. Choosing a testing system with a "soft" transducer that absorbs hand tremor and allows you to start with a very low force, will go a long way to solving the "touch-on" problem.

Consider abandoning really difficult mice. There are usually a few who just won't play and seem to take a perverse delight in subverting your tests. You might as well take them out of the study as their numbers are not going to be useful.

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