STANDARD OPERATING PROCEDURE

TITLE: Catwalk

CATEGORY: Behavioral Assay

Introduction

Goal: This document aims to provide the reader information on how to conduct the Catwalk Test (Noldus). The test provides quantitative gait analysis as an objective assessment of walking ability, and parameters collected include stride pattern, coordination, stance duration, and paw swing speed. As this is a general description of standard materials, test settings, and procedures, variations may be made to fit specific needs.

Materials

- **Subjects**: any strain of mice. Subjects require training and should be acclimated to testing environment and experimenter before testing.
- **Apparatus**: illuminated glass walkway on frame with camera mounted underneath walkway.
- **Software**: gait analysis software (ex. CatWalk by Noldus, license and dongle required).
- **Home cage**: placed directly next to entry of walkway, used to motivate animal towards end of walkway.
- **Step-down box**: object such as feeder container or ramp, placed inside home cage in front of walkway to facilitate entry into home cage.
- **Ethanol**: 70% or greater, used between trials to eliminate residue on glass walkway. (Virkon should not be used on glass walkway due to streaking that will interfere with tracking).

Test Settings

- **Lighting**: very dark. Computer screen should be dimmed as much as possible and all other lights should be turned off, including red lights.
Detailed Standard Operating Procedure

Before testing:
- **Acclimation**: subjects in home cage are placed in testing room for at least 1hr before testing to minimize effects of stress on behavior during testing.
- **Subject training**: subjects should be able to traverse walkway without hesitation, rearing, or reversing.
  - Animal is placed at edge of walkway closest to home cage and allowed to climb into cage with assistance of step-down box.
  - Animal is positioned on walkway farther and farther away from home cage until animal is able to walk toward cage when placed at opposite end of walkway.

Testing procedures:
- Animal is placed at far end of walkway and allowed to traverse walkway toward home cage.
- Animal is run until three compliant runs (defined by minimum of 4 complete sequence steps and no interruptions while traversing walkway) are recorded. Animals unable to complete three compliant runs are returned to home cage and attempted again at end of day.
- After three compliant runs, animal is returned to home cage and apparatus is cleaned with ethanol so no paw prints are visible.

Data Analysis
- The following parameters are collected for analysis:
  - Step sequence
  - Initial & max contact
  - Stride length
  - Print area
  - Base of support
  - Print position
  - Swing
  - Mean intensity
  - Swing speed