STANDARD OPERATING PROCEDURE

TITLE: Garcia Neurological Test

CATEGORY: Behavioral Assay

Introduction

Goal: This document aims to provide the reader information on how to conduct the Garcia Neurological Test. The test can be run simultaneously with the 28-point Neuroscore Test. Scores for the following six tests are summed to give a maximum total score of 18 points:

1. Spontaneous activity (max score 3)
2. Symmetry in forelimb movement (max score 3)
3. Forepaw outstretching (max score 3)
4. Climbing (max score 3)
5. Body proprioception (max score 3)
6. Response to vibrissae touch (max score 3)

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 rats. No prior training is required, though subjects should be acclimated to testing environment and experimenter before testing.
- *Wire sheet*: used in climbing test.
- *Stick*: used in body proprioception and response to vibrissae touch tests.
- *10% ethanol*: used to clean between trials.

Test Settings

- *Test area*: clean table top with sufficient space for animal and two home cages. No additional people in the room during testing.
- *Lighting*: well-lit.

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*: none required.

Testing procedures:

- Animal is subjected to a number of tests, with sequence of testing unimportant. Behavior is scored for each test according to score systems presented in Appendix A.
- Animal is returned to home cage and test area is cleaned with 10% ethanol between trials.

Data Analysis

- Scores for each test are summed to determine the overall Garcia test score (possible scores range from 3 to 18).

Appendix A: Scoring

Each score system may be broken down into half-point scores.

1. Spontaneous activity

(Activity is observed in test home cage for 5min.)

3 *Normal*: approaches all four walls of cage
2 *Slightly affected*: hesitates but still explores walls
1 *Moderately affected*: barely moves but eventually approaches wall
0 *Severely affected*: does not move

2. Symmetry in the movement of forepaws

(Outstretching and movement of forepaws are observed when animal is lifted by base of tail above ground.)

3 *Normal*: forepaws are symmetrical
2 *Slight*: limbs on impaired side do not extend as much as on normal side
1 *Moderate*: limbs on impaired side do not extend but are held close to body
0 *Severe*: limbs on impaired side do not move at all

3. Forepaw outstretching

(Forepaw outstretching and forepaw walking are observed when animal is lifted by base of tail and allowed to walk on forepaws across table. Forepaw use may also be observed when animal is dragged backwards by base of tail.)

3 *Normal*: forepaws are outstretched and animal is able to walk on forepaws in straight line
2 *Slight*: forepaws are outstretched and animal walks on forepaws but towards impaired side
1 *Moderate*: impaired forepaw does not outstretch well and forepaw walking is heavily biased toward impaired side
0 *Severe*: impaired forepaw does not outstretch or move during walking

4. Climbing

(Climbing is observed when animal is lifted by base of tail and placed on wire sheet resting on side of home cage.)

3 *Normal*: grips wire sheet and climbs up to home cage with ease
2 *Moderate*: climbs up to home cage but impaired paw misses wires while climbing
1 *Severe*: cannot grip wire sheet with impaired paw and cannot climb up to home cage

5. Body proprioception

(Reaction is observed when each side of animal is poked separately while crawling on floor. Animal should not see stick before poking.)

3 *Normal*: inspects both sides or is startled after poking on either side
2 *Moderate*: inspects impaired side after poking more slowly than unaffected side
1 *Severe*: does not respond after being poked on impaired side

6. Response to vibrissae touch

(Reaction is observed when whiskers are brushed on each side separately with a long, thin stick. Stick should approach from the rear and animal should be distracted with hand in front of head to ensure animal does not see stick while testing. Behavior should be scored during first few brushes, as too many brushes will accustom the animal to whiskers being touched.)

3 *Normal*: brushes side of head after whiskers are touched with stick, or head retreats from stick
2 *Moderate*: head retreats from stick more slowly after brushing whiskers on affected side as compared to unaffected side
1 *Severe*: does not respond after brushing whiskers on impaired side

Example Garcia Test Score Sheet

Study #:	**Garcia Test Group:**	Date:
Test #:		Initials:

Animal ID#	Forepaw Sym.	Forepaw Stretch	Spont. Activity	Climbing	Body Touch	Vibrissae Touch	Sum