



Rotarod

Version: 1/Sept 2023

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Summary:

The Rotarod tests is used to assess motor coordination and balance in rodents. Mice need to keep their balance on a rotating rod. The test can be perform using continuous acceleration (e.g., from 4 to 40 rpm) condition, or using different speeds. The instrument measures the time (latency) subject falls off the rotating rod.

Reagents and Materials:

Reagent/Material	Vendor	Stock Number
Rotarod	Columbus Instruments	Rotamex
Insulation foam (covering pipes)		
Timer		
Lab coats/gloves/PPE		
70% ethanol/paper towels		
Cleaning solution	10% Nolvasan	

Protocol:

1. SET-UP

- a. Acclimate mouse in their home cages in the testing room for 30 min.
- b. Weigh mouse as body weight influences performance on the rotarod.
- c. Replace grey rubber foam prior to testing (and between cohorts).
- d. Test Rotarod by turning the apparatus ON. Go to “Main Menu”, “Diagnostics”, the display should read 00-00-00-00, indicating that the optical sensors are working. Verify motor by activating speed of Rotarod manually.

IMPORTANT NOTES:

- Testing should be done at approximately same time of day for all experimental mice.
- Test is better managed with 2 persons (a handler and a recorder).
- Rubber foam can be cleaned thoroughly using 70% alcohol before use.

2. PROCEDURE (total time is ~45-60 min)

This test consists of 3 trials (T1, T2, T3) separated by 15 min inter-trial intervals (ITI). The apparatus can run 4 mice at once. Depending on the protocol, the test can be repeated over 2 days. The test may require an acclimation trial (depending on mouse strains to be tested). The conditions (acceleration, speed, etc) can vary according to study design.

- a. Turn on the Rotarod. Set experimental conditions by going to “Select Task” and then “Experimental Setup” Set the following conditions using the up/down arrows to enter values and enter to access the nest condition (field):
 - Accel = 0.6 RPM
 - Start speed (S-SP) = 4.0 RPM
 - Acceleration interval (Acc-In) = 5 sec
 - End speed (E-SP) = 40RPM
- b. This programs the continuous accelerating mode from 4 to 40 rpm in 300 sec. The instrument should read “acceleration waiting” and run at 4 rpm constant speed until the “Run Experiment” is pressed; make sure to activate all occupied lanes by pressing on the corresponding channel (CH1 to CH4) buttons.
- c. Place mice on lanes (lanes are separated by spacer disks. If mice handling is difficult, leave empty spaces and run a smaller no. of mice. **NOTE: the mice should be walking “forward” to maintain balance.** Once all the mice are loaded, press the start button for the continuous acceleration trial 1.
- d. Record the latency at which each mouse falls off the rotarod (elapsed), the fall time and fall speed on the Rotamex instrument.
- e. When all mice have completed T1, replace in their home cage and for the 15 min ITI.
- f. Repeat T2 and T3 (with a 15 min ITI between each trial) for a cohort of mice.
- g. Wipe the apparatus with 70% ethanol, then wipe dry between each trial.
- h. At the end of the procedure, disinfect the instrument with Nolvasan.

IMPORTANT NOTES:

- A mouse can “cling” onto the rod, without walking during the rotation. If the mouse completes a full *passive rotation* stop the timer for that mouse by pressing down the lever and record the latency.
- Passive rotation is considered a failure in performance as is failing of the rotarod. Remove these mice and place back in their home cage. Make note of passive rotations, falls and jumps.
- Data to collect per mouse include latency to fall at T1, T2, T3; fall time & speed at T1, T2, T3, passive rotation (yes/no) at T1, T2, T3 & body weights.