Standard Operating Procedure: Automated Reach-to-Grasp Task (AutoRG)

1. Purpose

- 1.1. This SOP describes the procedures necessary to properly utilize the hardware and software of the AutoRG system to assess forelimb motor function in rats.
- 1.2. This SOP utilizes the default, 7-stage training scheme provided in the standard code
- 1.3. This SOP assumes that the following processes are already completed:
 - 1.3.1. The apparatus is completely assembled and primed (see: AutoRG Assembly SOP)
 - 1.3.2. The software has been downloaded/integrated with a Windows computer (see: AutoRG SoftwareInitialization SOP)
 - 1.3.3. The system has been calibrated and the calibration file is accessible (see: AutoRG_Calibration_SOP)

2. Equipment and Terminology

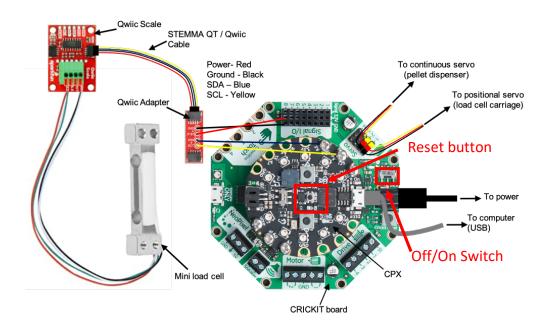
- 2.1.Cage Refers to acrylic cage in which the rat is placed for the duration of the session
- 2.2. Stage Refers collectively to the 4 stage blocks that are pre-assembled into one single stage, of which the cage is placed on top.
- 2.3. Handle Base Refers to the base to which the linear motion rail is affixed
- 2.4. CPX Refers to the Circuit Playground Express
- 2.5. CRICKIT Refers to CRICKIT board that is attached to the CPX
- 2.6. Pellet Dispenser Refers collectively to the Pellet Dispense Base and Food Silo that compose the pellet dispense apparatus
- 2.7. Pellet Dispense Base Refers to base of Pellet Dispenser that has the 4 legs and includes an insertion point for the Pellet Servo
- 2.8. Pellet Servo Actuator Refers to circular piece of Pellet Dispenser that connects to Pellet Servo and rotates within the Pellet Dispenser to allow pellets to be delivered
- 2.9. Food Silo Refers to top of Pellet Dispenser that holds the reservoir of pellets
- 2.10. Pellet Dispense Tower Refers to the tower block on which the pellet dispenser sits

- 2.11. Pellet Receptacle Refers to the receptacle that receives the pellet tubing from the pellet dispenser and re-directs the pellet into the slit of the cage
- 2.12. Pellet Tubing Refers to the rubber tubing that connects the pellet dispenser to the pellet receptacle (Saint-Gobain Tygon S3 E-3603 Food Tubing)
- 2.13. Pellet Servo Refers to the continuous servo that actuates the pellet dispenser
- 2.14. Calibration Knob Refers to knob that is used to calibrate the continuous servo
- 2.15. Load Cell Refers to mini load attached to handle
- 2.16. Load Cell Carriage Refers to bottom half of load cell casing that connects load cell to linear motion rail
- 2.17. Load Cell Handle Adapter Refers to top half of load cell casing that connects load cell to handle
- 2.18. Proximal Servo Arm Adapter Refers to servo arm adapter that connects directly to positional servo arm
- 2.19. Distal Servo Arm Adapter Refers to servo arm adapter that connects the proximal servo arm to the high cube intermediate joint
- 2.20. High Cube Intermediate Joint Refers to joint connecting the distal servo arm adapter to the load cell rail adapter
- 2.21. Handle Refers to 1.5mm AF hex key that is affixed to the load cell apparatus
- 2.22. Load Cell Apparatus Refers to load cell, load cell casing components, and rail attachment component
- 2.23. Rail Refers to the 100mm-long linear motion rail on which the load cell apparatus sits
- 2.24. Positional Servo Refers to the positional servo that actuates the movement of the load cell apparatus to modulate the handle distance
- 2.25. Positional Servo Casing Refers to SLA-printed casing that encloses the Positional Servo and sits at the back of the Handle Base
- 2.26. Qwiic Scale Refers to red analog-to-digital converter that connects the load cell to the Qwiic Adapter.

- 2.27. Qwiic Adapter Refers to the component that receives a plug from the Qwiic Scale and projects wires to the CRICKIT and CPX boards.
- 2.28. CPX Holder Refers to component where CPX rests; snaps into back of Pellet Dispense Tower.
- 2.29. Power Cord Refers to the cord connecting the CRICKIT board to power
- 2.30. USB Cord Refers to the cord connecting the CPX (micro-USB) to the computer (USB)
- 2.31. GUI1 Refers to the primary MatLab graphic user interface through which the AutoRG assay is run
- 2.32. GUI1 Code Refers to the code underlying GUI1
- 2.33. Calibration GUI Refers to the MatLab graphic user interface that creates the calibration file
- 2.34. Calibration GUI Code Refers to the code underlying the calibration GUI
- 2.35. Trigger Threshold: Minimum force required to trigger a successful trial and a pellet dispense

3. Assay Procedure

- 3.1. Plug in USB from CPX into the computer, ensure power cord is plugged into outlet and CRICKIT
- 3.2. Switch CPX on. Click reset button in center of CPX.



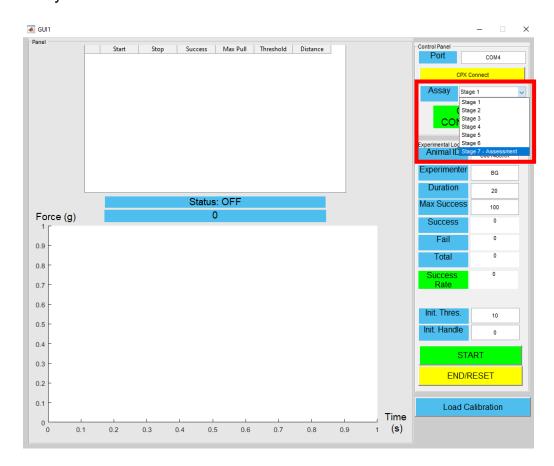
- 3.3.Open GUI1.m on MatLab
- 3.4. Click "Run" to open GUI1



- 3.5. In the textbox beside "Port", enter the port designation for your USB serial connection (CPX) (e.g. COM5). Click the yellow "CPX Connect" button below.
 - 3.5.1. To check the port designation for your CPX, go to Device Manager > Click on "Ports" > See port designation for "USB Serial Connection"
 - 3.5.2. Entering the COM designation directly in the GUI1 code saves a default COM designation so that you do not have to type in the designation each time (see: AutoRG_SoftwareInitialization_SOP). Note that the COM designation will change if a different CPX is used.
 - 3.5.3. Check that a green "COM[] CONNECTED" box appears (where [] is the number of your COM designation)



3.6. Select the Appropriate stage for the current rat in the dropdown box beside "Assay"



3.7. Enter any identifying number/code for the current rat in the textbox beside "Animal ID"



3.8. Enter any identifying information (initials, name) for the experimenter in the textbox beside "Experimenter"



- 3.9. To change the default session duration: Enter desired session duration in minutes in the textbox beside "Duration"
 - 3.9.1. Default: 20 minutes
 - 3.9.2. If user desires to change session duration permanently, it is recommended to make this change directly in the GUI1 code. See AutoRG_SoftwareCustomization for more information



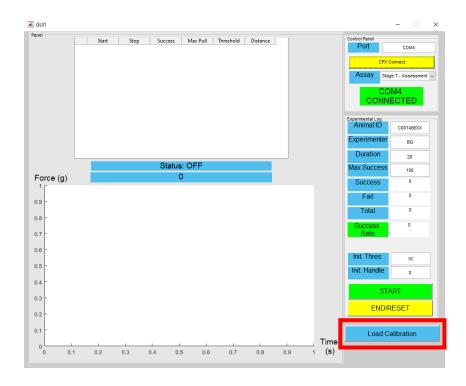
- 3.10. To change the default maximum number of successful trials for a given session: Enter desired maximum number of successful trials in the textbox beside "Max Success"
 - 3.10.1. Default: 100 successes
 - 3.10.2. If user desires to change maximum successes permanently, it is recommended to make this change directly in the GUI1 code. See AutoRG SoftwareCustomization for more information



- 3.11. All sessions begin with a "warmup" trial where the handle begins at 0.30 in inside the cage and the trigger threshold begins at 10g for one trial, regardless of the stage. To change the default:
 - 3.11.1. Trigger Threshold: Enter desired warmup threshold in grams in the textbox beside "Init. Thres."
 - 3.11.2. Handle Distance: Enter the desired warmup handle distance in degrees in the textbox beside "Init. Handle" (Note that 0 degrees is approximately 0.30 in inside the cage)



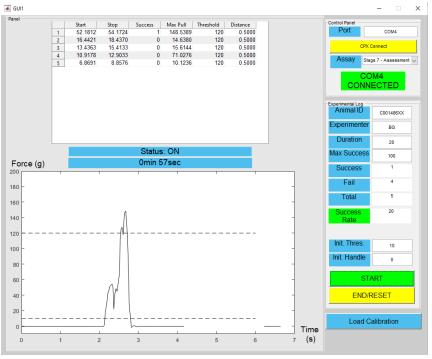
3.12. Click the blue "Load Calibration" button and select the most recent calibration file from the calibration GUI.



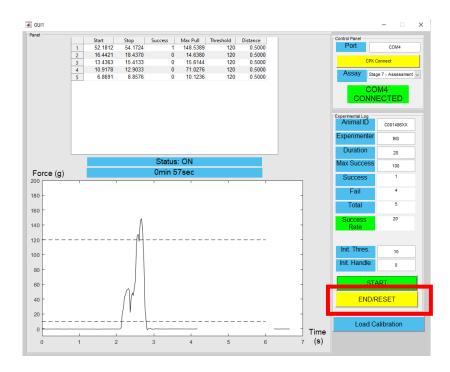
- 3.13. Once GUI1 has been prepared, place subject rat inside the cage
- 3.14. Click the green "START" button to begin the session.



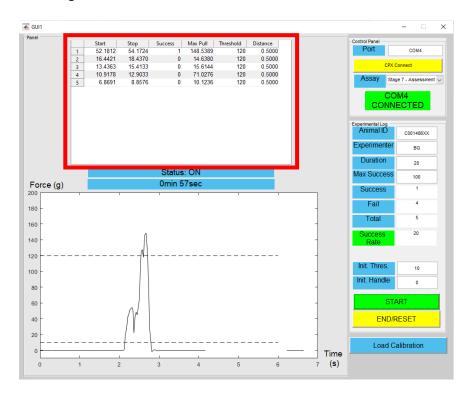




3.15. If session needs to end early, click the yellow "END/RESET" button to end the session. Session data will save automatically to the pre-specified location (see AutoRG SoftwareInitialization SOP)



3.16. Monitor session to ensure force readings are displayed on the GUI1 figure and trials are being recorded in the GUI1 data table



- 3.17. At the end of the session, session data will automatically save to the prespecified location. Remove the rat from the cage. Click yellow "END/RESET" button to clear GUI1
- 3.18. Port designation should not be affected by reset. Re-click yellow "CPX Connect" button
- 3.19. Repeat 3.4 3.9 as needed. Port designation, Animal ID, and Experimenter ID should not reset. Enter next animal ID's, and select the stage for this next rat. Place rat in cage, and click green "START" button to begin next session.
- 3.20. Repeat 3.13 3.17 for each session
- 3.21. After all sessions are complete: Clean cage with veterinary approved cleaner. Remove and rinse pellet receptacle to avoid pellet dust buildup. Place somewhere to dry. Switch CPX off. Unplug USB from computer.