Standard Operating Procedure

Intravenous self-administration paradigm
CSNA IVSA v2.0

Area: G3B	JAX-CSNA

Controls:	
Superseded Document	CSNA IVSA v1.0
Reason for Revision Major or Minor changes	New Dose Response Curve Schedule Removed doses and altered extinction phase to criterion based in lieu of timed duration.
Effective Date	March 1, 2018

Signatures:	
Author	I indicate that I have authored or updated this SOP according to applicable business requirements and our company procedure: Preparing and Updating Standard Operating Procedures. Name: Tyler A. Roy and Troy Wilcox Signature: _____ Date: ___7/1/19
Approver	I indicate that I have reviewed this SOP, and find it meets all applicable business requirements and that it reflects the procedure described. I approve it for use. Name: __Price Dickson and Leona Gagnon Signature: _____ Date: _____1/1/19_____

1. PURPOSE

This SOP addresses the routine procedures used for conducting the Intravenous Cocaine Self-administration procedure in mice including methods for analysis of data, and quality monitoring procedures.

2. SCOPE

The SOP applies to laboratories within the JAX CSNA Behavioral Phenotyping Core

3. RESPONSIBILITIES

3.1. **Laboratory Staff**

3.1.1. Remain up to date in training with this SOP

3.1.2. Comply with this SOP

3.2. **Principal Investigator/Core Manager of JAX-CSNA**

3.2.1. Ensures that all personnel involved running this SOP are trained to comply with this SOP

4. GLOSSARY/DEFINITIONS

Item	Definition
Active lever	This is the lever that is programed to reward the mice with an intravenous infusion of cocaineAn active lever press resulted in a cocaine infusion and the illumination of both stimulus lights for five seconds. This was followed by a twenty-second time-out during which the house light was off and lever presses were recorded but had no consequences.Throughout the entire session, inactive lever presses were recorded but had no consequences. The left lever is always assigned to the active lever.
Inactive lever	The inactive lever has no programmed consequences, pressing this lever does not effect the stimulus lights of the house light. The right lever is always assigned to the inactive lever.
% Active Press	This is the percentage of active lever presses relative to cumulative leverpresses of the active and inactive levers.

Acquisition	The acquisition phase is the first phase of testing. During this phase responses on the active lever under a fixed-ratio-1 (FR-1) schedule of reinforcement are reinforced with an infusion of 1.0 mg/kg cocaine. The criterion for achieving acquisition and completing this phase is ≥ 10 infusions for 5 consecutive test sessions and two consecutive test days meeting stabilization criteria (see below).
Stabilization of Response	Stabilization is defined as two consecutive sessions during which the number of infusions received) does not vary by more than 20% between the 2 consecutive sessions
Generation of the Dose-Response Curve (DRC)	This is the second phase of testing. During this phase of testing subjects are assessed for self-administration responses across an 4-point cocaine dose-response curve. Doses were presented in the following order: 0.32, 0.10, 0.032, 1.0 mg/kg/infusion. Subjects are tested on consecutive days on the same dose until stabilization criteria are met. Once the subject meets stabilization criterion at each dose they are moved to the next subsequent dose on the following session.
Extinction of Cocaine IVSA	Following the DRC, mice are evaluated for extinction defined as a reduction in lever presses on the active lever that do not result in a cocaine infusion reward. During extinction sessions, subjects are connected to saline filled infusion tubing however the infusion pump is not activated. During extinction sessions, the house light is continuously illuminated, however stimulus lights are never illuminated and lever presses have no programmed consequences. Mice are tested for a minimum of 3 days and a maximum of 9 days depending on whether the extinction criteria are met. The number of active lever presses on first day of the extinction session is the reference baseline for establishing extinction. The criteria for extinction is defined as follows: a) When active lever presses are reduced to <50% of the established baselinefrom the initial extinction session; and b) , the variance of the final two days are within 20% of the established baseline from the initial extinction session; c) if active leverpresses are <10 on or after day 3. Subjects that do not reach extinction criteria within 9 days are advanced to the reinstatement stage.
Exctinction Baseline (EB)	The extinction baseline is the number of active presses on the first day of extinction, where mice are connected to the inactive pump via infusion tubing. See above for additional details..
Reinstatement of Cocaine IVSA	Following extinction, responding on the active and inactive levers was examined for 2 daily two-hour sessions. During reinstatement sessions, drug paired stimuli including infusion pump were presented as in the acquisition and DRC but without infusion into the mouse. Specifically, mice were connected to

	infusion tubing which was connected to a sterile-saline filled syringe, but the syringe was not in the pump, thus mice do not receive drug infusions. Initiation of lever pressing is due to conditioned stimuli.

5. MATERIALS

5.1 Instrumentation

5.1.1.Operant Conditioning Chambers: (wide, modular, ENV-307W-CT; Med Associates Inc., Saint Albins, Vermont) fitted with 2 retractable levers on the front wall flanking the right and left sides of the center panel food hopper. Directly over each lever (~ 2-3 inches above) are red stimulus lights. A modified Plexiglas floor, fabricated at the Jackson Laboratory, was fitted to cover the metal floor grids.

5.1.2.Counterbalance: (Med Associates Inc., PHM-124MW, Saint Albins, Vermont.)

5.1.4.Tether: (Med Associates Inc., PHM-KVAH62T/MED, Saint Albins, Vermont.)

5.1.5. Swivel: (Med Associates Inc., PHM-115IP-25, Saint Albins, Vermont.)

5.1.6. PE Tubing: (Med Associates Inc., PHM-122-18, Saint Albins, Vermont.)

5.1.7. Catheter: (Instech, C20PU-MJV,Plymouth Meeting, PA)Instech's mouse VAB™ permits quick, aseptic connection and disconnection of a catheterized mouse and an infusion tether.

5.1.8. 120V Single Speed Syringe Pump: (Med Associates Inc., PHM-100, Saint Albins, Vermont.) This pump is located in the back left corner of the sound attenuating chamber next to the operant conditioning box.

5.1.9. Expanded PVC Sound Attenuating Cubicle (Med Associates Inc., ENV-022V, Saint Albins, Vermont.)

5.1.10. VAB62CAP Protective aluminum cap: (Instech'., PHM-VAB95CAP,.) Protects the port.

5.1.11. Scale: A scale for weighing animals with 0.1 mg resolution

5.1.12. Restrainer: (TV-150 STD & TV-150 SM, Braintree Scientific, Inc., Braintree, MA)

5.2. Consumables

5.2.1 Cocaine HCl: Supplied by National Institute on Drug Abuse Drug Supply Program (NDSP) Division of Therapeutics and Medical Consequences. Stored in powder form until it is formulated into 0.9% Saline (100 mg/mL clear solution) in various concentrations. Additional details are described in the CSNA IVSA Drug Preparation SOP.

5.2.1.1. On the day before use, pre-formulated, frozen solutions are brought to room temperature in preparation for the following days use.

5.2.1.1.1. Pre-formulated frozen solutions should not be heated (e.g by bath sonication or under warm water) as this may result in degradation of the cocaine.Once a solution is at room temperature, it should be labeled with an expiration date 10 days from the date it is brought to room temperature and used within 10 days.

5.2.2. 70% ethanol (ETOH) in water solution: used to sanitize the chambers between subjects. Between groups of animals (~ 2x per month) chambers are cleaned with standard Virkon wipes and then cleaned with 70% ETOH solution.

5.2.3. Paper towels

5.2.4. Wavicide disinfectant solution: (Performance Health, Item # 081545508, Akron, OH.) is used to flush the PE tubing at the conclusion of each test day.

5.2.5. Heparin Sodium Inj. USP 10,000 unites/ 10 mL: (NDC Code 63739-931-28 — Heparin Sodium) stored at room temperature and diluted to a 1:10 concentration in saline and administered as a bolus dose of 20ul per subject.

5.2.6. Bayer Baytril® (enrofloxacin) Injectable Solution 2.27%: Baytril Injection Solution is a broad-spectrum antibiotic designed for the management of bacterial pathogens, with activity against both Gram-negative and Gram-positive bacteria, including those causing dermal, urinary, and respiratory tract infections. Stock solution 22.7mg/mL is cut in half with sterile saline to 11.35mg/mL.

5.2.7. Brevital (methohexital): Supplied by National Institute on Drug Abuse Drug Stored in powder form until it is formulated into 0.9% Saline (100 mg/mL clear solution) Brevital is an ultrashort acting barbiturate and is used to confirm catheter patency once subjects have completed IVSA testing and prior to euthanasia [see Table 1 below for dosing].

5.2.8. 1CC syringe with 25G x 1 in. needle (VENDOR and PRODUCT #) These are used to administer Heparin, Brevital, and Baytril.

5.2.9. 60mL Syringes: (BD, Luer-Lok Tip, REF 309653)

5.2.10. Virkon™ S Wipes: (LANXESS, MF #57811373, Broad Spectrum Virucidal Disinfectant) Tablets are mixed with water to make a 1% solution. Which is applied and left for 10 minutes of contact time.

5.2.11. Spor-Klenz: (Steris, SPOR-KLENZ CONCENTRATE) Spor-Klenz working solution (1 part Spor-Klenz Steris Life Science Concentrate to 32 parts water) Mouse forceps are soaked in this sterilant and rinsed in water between every mouse.

Table 1. Drugs for IVSA study

Drug	Dose	Volume to administer	when to administer	purpose
Heparin lock [dilute with saline to reach 100 units per milliliter]	100 units per mL of saline	20 µL bolus injection regardless of mouse weight	Daily before entering chamber and after coming out of chamber. When multiple solutions are infused, heparin lock should always be infused last so that it locks the catheter.	confirmation that catheter is not blocked (before placing subject into test chamber) and to lock the catheter with the intention of preventing it from becoming blocked (after subject completes the day's testing and is removed from the chamber).

Baytril (enrofloxacin) [dilute with saline to 1/2]	22.7 mg per kilogram	2 µL per gram of mouse test weight (e.g., 25 g mouse gets 50 µL)	Daily at the conclusion of testing, immediately upon removal from the test chamber and prior to administration of the heparin lock	Baytril is an antibiotic. It is used daily throughout the study to prevent bacterial infection.
Brevital (methohexital)	5 mg per kilogram	100 µL	Once only at the completion of the study (after reinstatement) before the subject is euthanized.	Brevital is an ultrashort acting barbiturate and is used to confirm catheter patency.

6. PROCEDURE

6.1. **Subjects**

6.1.1. Species. Mice

6.1.1.1. Subjects undergo jugular vein catheterization prior to testing (see Jugular Vein Catheterization and Maintenance Procedure)

6.1.1.2. Subjects are provided a minimum 10 day post-operative recovery period in their home cage. Approval to initiate testing in individual subjects post-operatively is determined by veterinarian

6.1.2. Sex. Males or females

6.1.3. Age. The test is validated for mice ≥ 12 weeks of age.

6.1.4. Housing. Subjects are individually housed for this test.

6.1.5. Transport to Procedure Room. Subjects are transported in their home cages from the housing room to the procedure room on a wheeled transport rack. The procedure room is located ~ 5 feet from the housing room on the same floor.

6.1.6. Body Weight. If required such as for dosing, body weights are recorded to the nearest 0.1 miligram. Body weights for dosing are recorded on the first day of testing, and then again once per week on Tuesday prior to testing.

6.1.7. Subject Identification. Mice are individually housed and identified by their JCMS number, group number, and box number printed on their cage card. Mice are also ear notched at six weeks of age.

6.1.8. Acclimation. Prior to the start of testing, upon transport to the procedure room, subjects are weighed (once a week), and briefly handled to be assessed for any welfare concerns that may result in exclusion from testing (e.g., wounds or looking sickly).

6.2. **Environment**

6.2.1. Procedure Room. The dimensions of the procedure room are 21ft x 10 ft. Thirty-two chambers, each placed on separate shelves located next to each other along the entire left side of the room and half of the right side of the room. A 5-ft. laboratory workbench with two computer systems and monitors, a -20°C freezer, and a large rack to hold all mice during testing subjects are positioned on the right side of the room.

6.2.2. Temperature. The temperature range in the testing room is 70± 3 F.

6.2.3. Humidity. The humidity range in the procedure room is 50 ± 20%.

6.2.4. 6.1.4. Lighting. Room lighting in the testing room is overhead flouresence lights with a dimmer switch illuminated to a setting that produces a light level in the testing room of ~ 500 lux. This is verified monthly.

6.2.5. Noise. The background noise levels in the procedure room are 55-70dB. No additional or ancillary noise (white noise) is provided. Audible timers are not used during this test.

6.2.6. Time of day. The test is conducted during the light phase of the circadian cycle; beginning at least 30 min after the lights on and concluding at least 30 min prior to lights off.

6.3. **Test Compound**

6.3.1. Refer to the CSNA IVSA Drug Preparation SOP

6.3.2. 6.3.1.1. One day prior to the test, solutions are thawed overnight at room temperature.

6.4. **Experiment Prep Work**

6.4.1. Prepare MedPC macro Excel file. Using the Excel file titled “VBA script to create MedPC macro” on both computers for both control units, to create the macro used by MedPC to run the chambers.

6.4.2. Confirm MedPC macro Excel file is accurate.

6.4.3. Mouse ID, chamber, dose, sex, strain, active lever assignment and weight.

6.4.4. Confirm that all variable values on the Excel file (e.g. mouse ID, dose, sex, strain, and active lever) match the values on the paper tracking sheet. This is a separate step from preparing the Excel file and must be

performed each time a macro is created.. If an error is identified, edit and re-create the macro. Save the Excel file after creating the macro.

6.4.5. Load cocaine syringes. Flush out Wavicide from the PE tubing by connecting the cocaine syringe to the tubing .Once connected, firmly seat the syringe in the pump and turn the knob one full turn to ensure that cocaine has completely filled the tubing. Before connecting ensure that there are no air bubbles in the syringe.

6.4.6. Confirm correct cocaine dose is prepared for each chamber. After all cocaine syringes have been loaded in the chambers, confirm that the dose written on the tracking sheet matches the dose that has been loaded into the syringe pump. This is a separate step from loading the syringes and must be performed.

6.4.7. Test operant chamber hardware. Run the testing program on MedPC to ensure that hardware is working. In each chamber, press the right lever followed by the left lever. Both levers should retract. Then break the infrared beam in the pellet receptacle. Following this, both stimulus lights, the house light, and the syringe pump should turn on briefly. Verify that all of this occurs for each box. Repair any hardware issues before testing mice. Replace bulbs from houselights or stimulus lights if necessary..

6.4.8. Wipe down Plexiglas floors of chambers with 70% ethanol solution. Do this immediately before testing the hardware in the chambers for group 1 (i.e. morning group) and group 2 (i.e. afternoon group).

6.5. **Macro Software**

6.5.1. Prepare MedPC macro Excel file. Using the Excel file titled “VBA script to create MedPC macro”, create the macro used by MedPC to run the chambers.

6.5.2. Confirm mouse IDs. Critically review the Excel file against the paper tracking sheet every time a macro is created to ensure that the correct mouse ID, dose, sex, strain, and active lever are used in the macro. systematically review the information for each chamber on the Excel file and confirm that all variable values match the values on the paper tracking sheet. This is a separate step from preparing the Excel file and must be performed each time a macro is created. It is possible that any of these variables could have been inadvertently changed (mouse ID, dose, sex, strain, active lever). Ensure all columns are checked and confirmed as accurate. If an error is identified, adjust the macro with the correct information and create a new macro. Save the Excel file after creating the macro.

6.5.3. Open MED-PC IV program from desktop computer in the procedure room and navigate to “Open Macro”.

6.5.4. Navigate to Desktop -> Macro -> IVSA_day. Confirm the date and time is correct and matches the updated VBA script. Open the file and, confirm all chambers are ready and accurate. Once confirmed, start the macro.

6.6.

6.6.1. Test mouse on acquisition dose (1.0) until the subject completes 5 sessions with >= 10 infusions. If mouse also meets stabilization criteria (see below) on sessions 4 and 5 move to next dose or maintain testing on the currentacquisition dose until stabilization criteria is met. Once mouse has stabilized on the acquisition dose move to next dose and continue testing until mouse stabilizes OR until testing has exceeded 5 sessions on that dose. Continue to do this through all doses. Once the mouse has stabilized on the last cocaine dose, test on extinction for 3-9 sessions (see calculation of extinction criteria below). Then test on reinstatement for 2 sessions. The mouse has completed the experiment at this point.

6.7. **Criteria for acquisition of IVSA (only for acquisition dose)**

6.7.1. 5 sessions with ≥ 10 infusions. These sessions do not have to be consecutive.

6.7.2. Stabilization on the acquisition dose as defined below.

6.8. **Criteria for stabilization of IVSA (all doses)**

6.8.1. Number of infusions doesn’t vary by more than 20% for the last 2 consecutive sessions

6.9. **How to calculate stabilization criterion #1 above**

6.9.1. Multiply the highest number of infusions over last 2 days within each subject by .80.

6.9.1.1. Follow standard rounding rules (i.e., 20.4 is rounded to 20; 20.5 is rounded to 21).

6.9.2. If the number of infusions for the day with the lower number of infusions is ≥ to the day with the higher number of infusion multiplied by .8 then stabilization criterion #1 above is met.

6.10. **How to calculate extinction criterion #1 above**

6.10.1. Method 1; Extinction has a minimum of 3 days and a miximum of 9 days. Day 1 on extinction is exctinction baseline (EB)

6.10.2. To calculate the extinction criteria multiply EB by 0.2.

6.10.3. If the number of most recent 2 session active lever presses are within EB*(0.2) and the last sessions active lever presses is ≤ half of the EB the mouse has reached extinction and moves on to reinstatement.

6.10.3.1. Example: Active presses on each day of extinction (200, 180, 150, 140, 95, 70) The mouse moves on after day 6 because 70 is less than half of EB (200), additionally 95 and 70 are within 20% of 200(40).

6.10.4. Method 2 for reaching extinction criteria; if a mouse presses 10 times or fewer on or after day 3 of extinction, then the mouse has reached extinction and moves on.

6.10.4.1. Example: (16, 30, 25, 20, 25,11, 9) This mouse move on on day 7 because it pressed less than 10.

6.11. **Exception to stabilization, meeting critera, and exclusion**

6.11.1. After acquisition on all doses of cocaine, if a mouse has not stabilized (criterea shown above) on a dose after five days those days are highlighted blue and the mouse is forced on to the next dose.

6.11.2. If a mouse comes detached or runs out of solution the data for that mouse for that day is excluded. These days do not count towards the five day rule. Mice can stabilize around an excluded day. During extinction and reinstatement day is not excluded since they do not receive infusions during those procedures. A note is made in the comments.

6.11.3. Mice are excluded from the study if they do not acquire within 18 days of starting. A mouse may be excluded earlier if it is impossible for them to meet the acquisition criteria within the 18 days.

6.11.4. Mice are also excluded if the port becomes damaged or clogged in anyway that hinders their ability to receive doses. (A mouse may continue testing if they are on extinction when they are no longer patient)

6.11.5. All excluded mice are euthanized according to IACUC guidelines.

6.12. **Testing**

6.12.1. Start IVSA Program in MedPC. Before loading mice in the chambers, start the IVSA program in MedPC. For each chamber, the program will wait for you to press the right lever twice before the two-hour testing session begins. This will allow you to load a mouse in a chamber and then immediately press the right lever twice to start the testing session for that mouse. The stimulus lights and house light will flash to confirm that the session has started. This strategy will ensure that the program starts for each mouse as soon as he is loaded in the chamber. It will also

shorten the time between groups because the mouse in chamber 1 will be ~45 minutes into the two-hour program by the time the mouse in chamber 32 is loaded.

6.12.2. Load mice into restrainer by guiding their tail through the slit on the top of the restrainer to the back so the mouse is completely in and place the stopper over the opening so they can not get out. Flush each catheter with 20 µL of heparin lock solution (100 units per milliliter). The purpose of this is to verify that solution will flow through the catheter. If the catheter is completely blocked, and can not be cleared out with the heparin, the mouse should not be tested. Provided that the catheter is not blocked, bring the restrainer to the chamber, connect mouse to the tether, place mouse in the chamber, hit the right lever(inactive lever twice, and close the chamber door.

6.12.3. Adjust counterbalance arm. After loading the mouse and before closing the door to the sound attenuating cubicle, adjust the knob on the counterbalance arm to ensure that the mouse can easily move to the levers without resistance.

6.12.4. Double check MedPC software after all mice have been loaded. Confirm that the session has started in all 32 chambers by looking at the MedPC program on the computer screen.

When program is finished remove mice from chambers. As soon as the mouse in chamber 1 has completed the session, remove the mouse from the chamber. Before removing a mouse, verify the mouse has completed the session by looking at MedPC on the computer screen. After removing mouse from chamber, flush with Baytril (2 µL per gram of mouse weight). Then flush with heparin lock solution (20 µL regardless of weight). Then return subject to the homecage. Repeat this step for each subsequent mouse and chamber.

6.12.5. Remove tubing from cocaine syringes

6.12.6. Fill all tubing in the chambers with Wavicide to sterilize overnight.

6.12.7. Clean chambers –in addition to the normal cleaning at the end of the day the chambers are completely taken apart and cleaned with ethanol to make sure no urine is left under the floor in the seams of the equipment. Before naïve mice are entered into the protocol this process is done with Virkon and cleaned off with ethanol after 10 minutes.

6.12.8. Analyze data. Using MedPC to Excel, analyze all data. A detailed discussion of this is provided in the section below entitled “Analyzing IVSA Data”.

6.12.9. Close MedPC.

6.12.10. Close all instances of Excel. Make sure to save changes to the macro Creator Excel file.

6.13. **Data Analysis and QC**

6.13.1. IVSA data should be analyzed every evening using MedPC to Excel.

6.13.2. First click on the IVSA Data shortcut icon found on the desktop

6.13.3. Using MedPC to Excel, filter data into the “currently testing” tab in the Excel file “IVSA data”

6.13.4. Once open click on the next available cell in column A (Subject) and then open MED-PC to Excel by clicking on the Icon on the desktop.

6.13.5. In the top section labelled Profile select the file path Desktop>Misc and select file IVSA- all vars.MRP

6.13.6. In MED_PC click transfer in second section of profile follow file path Desktop>CU1 data and select file with the date from this day of testing

6.13.7. Program will enter the data into the excel file

6.13.8. Save the excel file, close it and reopen on second computer with the second control unit. Repeat same steps for analyzing data labeling this control unit CU1

6.13.9. Then remove syringes for next day of testing from the freezer.

6.14. **Study Completion Criteria**

6.14.1. Subjects have completed the study at the conclusion of the reinstatement stage, or when he has definitively lost catheter patency (i.e. completely blocked catheter, absolutely no reaction to methohexital), or if he dies. When any of these happen, do the following:

6.14.2. Delete the subject ID from the paper tracking sheet and from the Excel file that generates the MedPC macros

6.14.3. Copy and paste the subject’s data from the “currently testing” tab in the IVSA data Excel file and paste it into the “finished” tab in the same file. This ensures that only mice that are currently being tested are in the “currently testing” tab. Otherwise that tab would continue to grow indefinitely and it would be difficult to work with when trying to determine which mice have met criteria.

7. Data Upload

7.1. Data uploads are verified and performed as described in the CSNA Data QC and Upload SOP.

8. Variables

Variable Name	Description
Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 01	Total number of active lever presses that occurred on acquisition session 01
Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 02	Total number of active lever presses that occurred on acquisition session 02
Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 03	Total number of active lever presses that occurred on acquisition session 03
Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 04	Total number of active lever presses that occurred on acquisition session 04
Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 05	Total number of active lever presses that occurred on acquisition session 05
Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 06	Total number of active lever presses that occurred on acquisition session 06
Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 07	Total number of active lever presses that occurred on acquisition session 07
Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 08	Total number of active lever presses that occurred on acquisition session 08
Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 09	Total number of active lever presses that occurred on acquisition session 09
Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 10	Total number of active lever presses that occurred on acquisition session 10
Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 11	Total number of active lever presses that occurred on acquisition session 11
Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 12	Total number of active lever presses that occurred on acquisition session 12
Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 13	Total number of active lever presses that occurred on acquisition session 13
Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 14	Total number of active lever presses that occurred on acquisition session 14
Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 15	Total number of active lever presses that occurred on acquisition session 15
Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 16	Total number of active lever presses that occurred on acquisition session 16
Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 17	Total number of active lever presses that occurred on acquisition session 17
Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 18	Total number of active lever presses that occurred on acquisition session 18
Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 19	Total number of active lever presses that occurred on acquisition session 19
Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 20	Total number of active lever presses that occurred on acquisition session 20

Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 21	Total number of active lever presses that occurred on acquisition session 21
Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 22	Total number of active lever presses that occurred on acquisition session 22
Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 23	Total number of active lever presses that occurred on acquisition session 23
Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 24	Total number of active lever presses that occurred on acquisition session 24
Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 25	Total number of active lever presses that occurred on acquisition session 25
Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 26	Total number of active lever presses that occurred on acquisition session 26
Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 27	Total number of active lever presses that occurred on acquisition session 27
Acquisition at 1.0 mg/kg-ActiveLeverPresses_Total-Session 28	Total number of active lever presses that occurred on acquisition session 28
Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 01	Total number of inactive lever presses that occurred on acquisition session 01
Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 02	Total number of inactive lever presses that occurred on acquisition session 02
Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 03	Total number of inactive lever presses that occurred on acquisition session 03
Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 04	Total number of inactive lever presses that occurred on acquisition session 04
Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 05	Total number of inactive lever presses that occurred on acquisition session 05
Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 06	Total number of inactive lever presses that occurred on acquisition session 06
Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 07	Total number of inactive lever presses that occurred on acquisition session 07
Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 08	Total number of inactive lever presses that occurred on acquisition session 08
Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 09	Total number of inactive lever presses that occurred on acquisition session 09
Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 10	Total number of inactive lever presses that occurred on acquisition session 10
Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 11	Total number of inactive lever presses that occurred on acquisition session 11
Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 12	Total number of inactive lever presses that occurred on acquisition session 12
Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 13	Total number of inactive lever presses that occurred on acquisition session 13
Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 14	Total number of inactive lever presses that occurred on acquisition session 14

Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 15	Total number of inactive lever presses that occurred on acquisition session 15
Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 16	Total number of inactive lever presses that occurred on acquisition session 16
Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 17	Total number of inactive lever presses that occurred on acquisition session 17
Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 18	Total number of inactive lever presses that occurred on acquisition session 18
Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 19	Total number of inactive lever presses that occurred on acquisition session 19
Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 20	Total number of inactive lever presses that occurred on acquisition session 20
Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 21	Total number of inactive lever presses that occurred on acquisition session 21
Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 22	Total number of inactive lever presses that occurred on acquisition session 22
Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 23	Total number of inactive lever presses that occurred on acquisition session 23
Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 24	Total number of inactive lever presses that occurred on acquisition session 24
Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 25	Total number of inactive lever presses that occurred on acquisition session 25
Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 26	Total number of inactive lever presses that occurred on acquisition session 26
Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 27	Total number of inactive lever presses that occurred on acquisition session 27
Acquisition at 1.0 mg/kg-InactiveLeverPresses_Total-Session 28	Total number of inactive lever presses that occurred on acquisition session 28
Acquisition at 1.0 mg/kg-Infusions_Total-Session 01	Total number of infusions that occurred on acquisition session 01
Acquisition at 1.0 mg/kg-Infusions_Total-Session 02	Total number of infusions that occurred on acquisition session 02
Acquisition at 1.0 mg/kg-Infusions_Total-Session 03	Total number of infusions that occurred on acquisition session 03
Acquisition at 1.0 mg/kg-Infusions_Total-Session 04	Total number of infusions that occurred on acquisition session 04
Acquisition at 1.0 mg/kg-Infusions_Total-Session 05	Total number of infusions that occurred on acquisition session 05
Acquisition at 1.0 mg/kg-Infusions_Total-Session 06	Total number of infusions that occurred on acquisition session 06
Acquisition at 1.0 mg/kg-Infusions_Total-Session 07	Total number of infusions that occurred on acquisition session 07
Acquisition at 1.0 mg/kg-Infusions_Total-Session 08	Total number of infusions that occurred on acquisition session 08

Acquisition at 1.0 mg/kg-Infusions_Total-Session 09	Total number of infusions that occurred on acquisition session 09
Acquisition at 1.0 mg/kg-Infusions_Total-Session 10	Total number of infusions that occurred on acquisition session 10
Acquisition at 1.0 mg/kg-Infusions_Total-Session 11	Total number of infusions that occurred on acquisition session 11
Acquisition at 1.0 mg/kg-Infusions_Total-Session 12	Total number of infusions that occurred on acquisition session 12
Acquisition at 1.0 mg/kg-Infusions_Total-Session 13	Total number of infusions that occurred on acquisition session 13
Acquisition at 1.0 mg/kg-Infusions_Total-Session 14	Total number of infusions that occurred on acquisition session 14
Acquisition at 1.0 mg/kg-Infusions_Total-Session 15	Total number of infusions that occurred on acquisition session 15
Acquisition at 1.0 mg/kg-Infusions_Total-Session 16	Total number of infusions that occurred on acquisition session 16
Acquisition at 1.0 mg/kg-Infusions_Total-Session 17	Total number of infusions that occurred on acquisition session 17
Acquisition at 1.0 mg/kg-Infusions_Total-Session 18	Total number of infusions that occurred on acquisition session 18
Acquisition at 1.0 mg/kg-Infusions_Total-Session 19	Total number of infusions that occurred on acquisition session 19
Acquisition at 1.0 mg/kg-Infusions_Total-Session 20	Total number of infusions that occurred on acquisition session 20
Acquisition at 1.0 mg/kg-Infusions_Total-Session 21	Total number of infusions that occurred on acquisition session 21
Acquisition at 1.0 mg/kg-Infusions_Total-Session 22	Total number of infusions that occurred on acquisition session 22
Acquisition at 1.0 mg/kg-Infusions_Total-Session 23	Total number of infusions that occurred on acquisition session 23
Acquisition at 1.0 mg/kg-Infusions_Total-Session 24	Total number of infusions that occurred on acquisition session 24
Acquisition at 1.0 mg/kg-Infusions_Total-Session 25	Total number of infusions that occurred on acquisition session 25
Acquisition at 1.0 mg/kg-Infusions_Total-Session 26	Total number of infusions that occurred on acquisition session 26
Acquisition at 1.0 mg/kg-Infusions_Total-Session 27	Total number of infusions that occurred on acquisition session 27
Acquisition at 1.0 mg/kg-Infusions_Total-Session 28	Total number of infusions that occurred on acquisition session 28
Dose response FR1-ActiveLeverPresses_Total-0.032 mg/kg	Mean active lever presses that occurred on the final two sessions of this dose: 0.032 mg/kg
Dose response FR1-ActiveLeverPresses_Total-0.056 mg/kg	Mean active lever presses that occurred on the final two sessions of this dose: 0.056 mg/kg

Dose response FR1-ActiveLeverPresses_Total-0.1 mg/kg	Mean active lever presses that occurred on the final two sessions of this dose: 0.1 mg/kg
Dose response FR1-ActiveLeverPresses_Total-0.18 mg/kg	Mean active lever presses that occurred on the final two sessions of this dose: 0.18 mg/kg
Dose response FR1-ActiveLeverPresses_Total-0.32 mg/kg	Mean active lever presses that occurred on the final two sessions of this dose: 0.32 mg/kg
Dose response FR1-ActiveLeverPresses_Total-0.56 mg/kg	Mean active lever presses that occurred on the final two sessions of this dose: 0.56 mg/kg
Dose response FR1-ActiveLeverPresses_Total-1.0 mg/kg	Mean active lever presses that occurred on the final two sessions of this dose: 1.0 mg/kg
Dose response FR1-ActiveLeverPresses_Total-1.8 mg/kg	Mean active lever presses that occurred on the final two sessions of this dose: 1.8 mg/kg
Dose response FR1-InactiveLeverPresses_Total-0.032 mg/kg	Mean inactive lever presses that occurred on the final two sessions of this dose: 0.032 mg/kg
Dose response FR1-InactiveLeverPresses_Total-0.056 mg/kg	Mean inactive lever presses that occurred on the final two sessions of this dose: 0.056 mg/kg
Dose response FR1-InactiveLeverPresses_Total-0.1 mg/kg	Mean inactive lever presses that occurred on the final two sessions of this dose: 0.1 mg/kg
Dose response FR1-InactiveLeverPresses_Total-0.18 mg/kg	Mean inactive lever presses that occurred on the final two sessions of this dose: 0.18 mg/kg
Dose response FR1-InactiveLeverPresses_Total-0.32 mg/kg	Mean inactive lever presses that occurred on the final two sessions of this dose: 0.32 mg/kg
Dose response FR1-InactiveLeverPresses_Total-0.56 mg/kg	Mean inactive lever presses that occurred on the final two sessions of this dose: 0.56 mg/kg
Dose response FR1-InactiveLeverPresses_Total-1.0 mg/kg	Mean inactive lever presses that occurred on the final two sessions of this dose: 1.0 mg/kg
Dose response FR1-InactiveLeverPresses_Total-1.8 mg/kg	Mean inactive lever presses that occurred on the final two sessions of this dose: 1.8 mg/kg
Dose response FR1-Infusions_Total-0.032 mg/kg	Mean infusions that occurred on the final two sessions of this dose: 0.032 mg/kg
Dose response FR1-Infusions_Total-0.056 mg/kg	Mean infusions that occurred on the final two sessions of this dose: 0.056 mg/kg
Dose response FR1-Infusions_Total-0.1 mg/kg	Mean infusions that occurred on the final two sessions of this dose: 0.1 mg/kg
Dose response FR1-Infusions_Total-0.18 mg/kg	Mean infusions that occurred on the final two sessions of this dose: 0.18 mg/kg
Dose response FR1-Infusions_Total-0.32 mg/kg	Mean infusions that occurred on the final two sessions of this dose: 0.32 mg/kg
Dose response FR1-Infusions_Total-0.56 mg/kg	Mean infusions that occurred on the final two sessions of this dose: 0.56 mg/kg
Dose response FR1-Infusions_Total-1.0 mg/kg	Mean infusions that occurred on the final two sessions of this dose: 1.0 mg/kg

Dose response FR1-Infusions_Total-1.8 mg/kg	Mean infusions that occurred on the final two sessions of this dose: 1.8 mg/kg
Extinction-ActiveLeverPresses_Total-Session 01	Total number of active lever presses that occurred on extinction session 01
Extinction-ActiveLeverPresses_Total-Session 02	Total number of active lever presses that occurred on extinction session 02
Extinction-ActiveLeverPresses_Total-Session 03	Total number of active lever presses that occurred on extinction session 03
Extinction-ActiveLeverPresses_Total-Session 04	Total number of active lever presses that occurred on extinction session 04
Extinction-ActiveLeverPresses_Total-Session 05	Total number of active lever presses that occurred on extinction session 05
Extinction-ActiveLeverPresses_Total-Session 06	Total number of active lever presses that occurred on extinction session 06
Extinction-ActiveLeverPresses_Total-Session 07	Total number of active lever presses that occurred on extinction session 07
Extinction-InactiveLeverPresses_Total-Session 01	Total number of inactive lever presses that occurred on extinction session 01
Extinction-InactiveLeverPresses_Total-Session 02	Total number of inactive lever presses that occurred on extinction session 02
Extinction-InactiveLeverPresses_Total-Session 03	Total number of inactive lever presses that occurred on extinction session 03
Extinction-InactiveLeverPresses_Total-Session 04	Total number of inactive lever presses that occurred on extinction session 04
Extinction-InactiveLeverPresses_Total-Session 05	Total number of inactive lever presses that occurred on extinction session 05
Extinction-InactiveLeverPresses_Total-Session 06	Total number of inactive lever presses that occurred on extinction session 06
Extinction-InactiveLeverPresses_Total-Session 07	Total number of inactive lever presses that occurred on extinction session 07
Reinstatement-ActiveLeverPresses_Total-Session 01	Total number of active lever presses that occurred on reinstatement session 01
Reinstatement-ActiveLeverPresses_Total-Session 02	Total number of active lever presses that occurred on reinstatement session 02
Reinstatement-InactiveLeverPresses_Total-Session 01	Total number of inactive lever presses that occurred on reinstatement session 01
Reinstatement-InactiveLeverPresses_Total-Session 02	Total number of inactive lever presses that occurred on reinstatement session 02