# Trapping and blood-sampling small mammals in semi-arid environments

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ABSTRACT

Brief guide to trapping and blood-sampling of small terrestrial mammals, including materials, procedures and recommendations.

EXTERNAL LINK

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PROTOCOL STATUS

**Working**

MATERIALS TEXT

*Trapping*

a. Thick gloves (leather, rubber or textile plus nitrile gloves)
b. Wire-mesh (e.g. Rodentrap®), Sherman™, Tomahawk™, or Havahart™ live-traps.
c. Baits: rolled oat or another adequate attractant for the target species according to the researcher's experience or references.
d. Marking material: masking tape, flags, flagging tape, markers, etc.
e. Bedding material: hydrophilic cotton.
f. Plastic or paper shelter material.
g. GPS or mobile phone equipped with geo-positioning software.
h. Other personal protection equipment

*Blood-sampling*

a. Tent or isolated room nearby (field laboratory).
b. Anesthesia equipment: anesthetics agents (isoflurane, ketamine, xylazine), portable anesthesia machine or induction chamber, maintenance chamber.
c. Weighing implements: portable balance or spring scale.
d. Blood-sampling implements: shaving implements (electric shaver, scissors), 70% ethanol, needles (21 G, 23 G, 25 G), syringes (1 ml, 3 ml), nuclease-free microcentrifuge tubes or cryovials (0.6 ml, 1.6 ml, 2 ml), anticoagulants and blood preservatives (e.g. 6M Guanidine-HCl 0.2M EDTA).
e. Optional: Warming pad.

*Marking*

a. Uniquely coded mouse ear tags.
b. Tag applicator

## Trapping

1 If the target or sympatric species are suspected to transmit infectious diseases, wear disposable gloves underneath and over the thick gloves, a whole-body disposable suit (e.g. Tyvek™), filtered mask, or even a respirator through the whole process.

2 Visually inspect the areas where is intended to trap micrommamals, searching for direct or indirect signs of animals' presence (urine marks, droppings, hair, burrows, etc.).

3 Manipulate the traps with thick gloves. Mark each trap with a unique code. Set it with the chosen bait and bedding material, paying attention to put the device in a horizontal surface, checking its stability (it should not move when the animal is entering it). The traps have to be located in places protected of extreme weather (under bushes, trees or cactuses) and/or use shelter material to protect the captured animals. The trap setting strategy (transect, web design, directed sampling, etc.) will depend on the aim of the study.

4 Mark the site with a flag or flagging tape, writing the code of the respective trap. Georeference the point where the trap is located.

5 The traps should remain activated (i.e. open and baited) during the maximum activity period of the target species. In the case of nocturnal mammals, the traps can be activated since sunset until the next morning. In the case of diurnal mammals, the traps can be opened immediately before the start of their foraging activity until the time when the animals rest. In the latter, the traps should be monitored ideally every 2-3 hours to avoid capture mortality due to thermal stress.

6 Closed traps should be inspected carefully to check if there is an animal captured. Sometimes non-target species are captured (birds, reptiles), and they should be gently released as soon as possible.

7 If the small mammals have *trap-happy* behavior, the bait of traps without captured animals should be removed (released to the surroundings) and the traps should ideally be folded and/or placed in a less accessible place (e.g. over shrubs), to avoid inadvertent captures in the time between activation of the grid. On the contrary, if they are reluctant to new objects, the trap should be closed with some bait inside, in the same capture point.

8 Transport the captured small mammals to an equipped laboratory inside the trap and wrapped in newspaper or thick plastic. It is recommended to place the traps in a container, such as a travel bag, to restrain them during transport and to protect them from extreme environmental conditions, but allowing air to enter the container.

## Blood-sampling

9 Always wear protective clothing and googles, regular or filtered mask and nitrile or latex gloves through this step; in case of suspected reservoirs of airborne infectious diseases, wear a respirator.

10 Anesthesia: We use isoflurane for induction, and for manipulation isoflurane can be used* or an injectable agent, such as ketamine/xylazine 40-85/5-21 mg/Kg (henceforth, KX). The animal is induced inside the trap, and after immobilization it is manipulated, allowing it to be weighed and later injected or directly measured and blood sampled. Blood pressure is low when anesthetized with isoflurane, and better with KX, which facilitates blood sampling. On the other hand, the recovery time for isoflurane is very short (a minute after cessation of administration); KX takes longer, according to dosage (half an hour to an hour, depending on species). Warming (heating) pads are useful, since anesthetized small mammals tend to hypothermia, and warmer animals have better peripherical blood flow; however, its temperature should be checked regularly to avoid overheating.

11 • See https://www.umt.edu/research/LAR/sops/SOPopendropisoflurane.php for field recommendation of isoflurane use and dosage.

12 Blood withdrawal: In our protocol, the blood sample consisted in a minimum of 0.2 ml of peripheral venous blood. As the maximum blood volume to extract corresponds to 1% of body weight [1], only individuals that weighed more than 20 gr are sampled. The shaving of areas with hair cover facilitates blood withdrawal. Ethanol can be used to disinfect after all hair of the venipuncture area has been clipped.

13 1] Joslin JO. Blood collection techniques in exotic small mammals. J Exot Pet Med. 2009;18(2):117-139.

14 Anatomical sites and material:
- For species of the genus *Octodon* sp., *Rattus* sp., and *Abrocoma bennetti*: Venipuncture of the saphenous vein, using a 19 or 21G needles.
- For *Phyllotis darwini* and *Mus musculus*: Venipuncture of the submandibular zone (in the union of the orbital and submandibular vein), with a 21 G needle. When anesthetized with KX, in *P. darwini* also by venipuncture of the saphenous vein, using 23 or 21G needles. *M. musculus* blood can also be obtained by jugular extraction with a 1 ml 25 G syringe. This procedure must be done very slowly.
- For *Thylamys elegans*, *Oligoryzomys longicaudatus* and *Abrothrix* sp.: Blood extraction from the jugular vein, with a 1 ml 25 G syringe. This procedure must be done very slowly. For *Abrothrix* sp., sometimes when anesthetized with KX, venipuncture of the saphenous vein can be achieved, using 23 or 21G needles.
- For lagomorphs (*Oryctolagus cuniculus*, *Lepus europaeus*): Blood extraction from the ear vein, with a 1 ml 25 G syringe, or from the saphenous vein, with a 3 ml 21 or 23 G syringe.

15 We usually replace the volume extracted with physiological serum (saline) subcutaneously. Currently, we prefer to administer 0.2 ml before blood withdrawal (with a 1 ml 25 G syringe). If more blood is obtained, the difference in volume should be replaced with saline afterwards.

16 Blood samples are placed in cryovials or microcentrifuge tubes and immediately mixed in a 1:1 proportion with 6 M Guanidine-HCl 0.2 M EDTA, for DNA preservation at room temperature [2] and subsequent molecular techniques.
For serological techniques, blood should not be mixed with the mentioned solution; it can be left to coagulate in the tube, and serum can be retrieved posteriorly after clotting; it is easily separated with a microcentrifuge. Serum has to be frozen if not used immediately. The clot can also be frozen and later used for molecular techniques.

17 2] Avila HA, Sigman DS, Cohen LM, Millikan RC, Simpson L. Polymerase chain reaction amplification of Trypanosoma cruzi kinetoplast minicircle DNA isolated from whole blood lysates: diagnosis of chronic Chagas' disease. Mol Biochem Parasitol. 1991;48(2):211-221.

## Mark and release

18 We mark each individual in both ears with a metallic ear tag uniquely coded, to prevent resampling. Other marks can be used (hair clipping, one ear tag per individual, identification chip, etc.).

19 Once small mammals are completely recovered from anesthesia, they should be transported as explained in Step 8 and released at their exact point of capture.