

- SOP No:** AHP 76
- SUBJECT:** Serial plasma collection from jugular vein cannula (j.v.c.) in rats
- POLICY:** This procedure may only be performed by operators skilled in the technique.
Surgery must be performed under aseptic conditions
This SOP is to be read in conjunction with [SOP AHT52](#)
- PRECAUTIONS:** Surgical gloves, eye protection, long-sleeved gown, closed in shoes.
All instruments and materials (including catheter) must be sterilized before use.
- EQUIPMENT:** Rat with jugular vein cannula (j.v.c.) at least seven day post-implantation.
Polyvinyl chloride (PVC) tubing (ID= 0.8mm, OD= 1.2mm, available from Microtube (Extrusions Pty Ltd, NSW, Australia), length approx 5-10cm
Blunted and trimmed 23G needle (pin), length approx. 1cm. MUST be patent.
Heparinised sterile saline (20UI heparin/ml saline)
Haemostats x2 (sanitised in 70% ethanol)
Sterile saline (approx 10-50ml)
1ml syringes (as many as samples to be collected), pre-heparinised (by flushing and evacuating with 100UI/ml saline)
23G needle, blunted.
Microfuge tubes; pre-labelled and pre-heparinised (by flushing and evacuating with 100UI/ml saline)
Timer
Esky/cool-box containing ice
70% ethanol
Kidney dish
-80°C freezer
- PROCEDURE:**
1. Clean bench-top thoroughly with 70% ethanol
 2. Line the bench with bench-coat
 3. Attach the PVC line to a syringe filled with heparinised saline using the blunted 23G needle. Connect the pin to the other end.
 4. Push 0.5ml the hep. saline through the line. NOTE: no air bubbles may be present.
 5. Obtain the animal with an implanted j.v.c. ([SOP AHT52](#))
 6. Manually restrain animal during the procedure.
 7. Clean the external cannula and the surrounding area thoroughly with 70% ethanol.
 8. Clamp the indwelling cannula with the haemostats just below the stopper-pin.

9. Remove the stopper-pin from the indwelling cannula using the other haemostats, and place it in 70% ethanol in the kidney dish.
10. Connect the pin and line to the indwelling cannula and release the haemostats.
11. Gently push the hep. saline (0.5ml) into the cannula to make patent and remove clots at a rate of approx 1ml/min.
12. Wait 30sec.
13. Gently pull the plunger back (approx. rate of 1ml/min) and withdraw blood from jugular vein. Collect no more than 0.5ml.
14. Clamp the indwelling cannula just below the pin with haemostats.
15. Remove the line and replace the stopper-pin after dipping it in sterile saline.
16. Evacuate the blood from the line and syringe into a pre-labelled/heparinised microfuge tube and place on ice.
17. Start timer for next collection interval.
18. Discard syringe and flush line with fresh syringe/blunted needle filled with sterile saline.
19. Repeat 1-17 at next intended time point.

RECOMMENDATIONS

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REVISED:

REFERENCES

1. Putcha, L., Bruckner, J. V., Muralidhara, S. and Feldman, S.J. (1982) *Pharmacol Methods* 8, 145.
2. Phillips, W. A., Stafford, W. W., and Stuu, J., Jr., (1973). *Proc Soc Exp Biol Med* 143, 733.
3. Bardelmeijer H. A. *et al.*, (2003) *Lab Anim* 37, 181.