

SOP No: AHT 50

SUBJECT: MONOCLONAL (POLYCLONAL) ANTIBODY
PRODUCTION (Ascites in Mice).

POLICY The *in vivo* production of antibodies by this method must be justified to the AEC. This technique is strongly discouraged by the NHMRC.
The volume of pristine/Freunds complete should not exceed 0.2ml.
A priming agent should not be used in individual mice more than once.
No more than 2 survival taps should be attempted. A final tap can be performed at termination.
Mice should be euthanased within 4 weeks of priming.

PRECAUTIONS: Gloves, eye protection, long-sleeve gown and closed in shoes.

EQUIPMENT: 18 – 22g needles
70% Alcohol
100µl and 5ml syringes
collection tubes
Pristane

PROCEDURE:

1. Prime adult female mice (> 6 weeks) by i/p injection of 0.2ml of pristane .
2. After 7 – 14 days, inject 5×10^5 to 5×10^6 hybridoma cells i/p in a volume of 0.1 – 0.5ml.
3. Tap the ascitic fluid when the abdomen becomes distended, using an 22g or smaller needle. (anaesthetizing the mouse will improve the ease of collection and should be used for all recovery collections).
4. Replace lost fluid with 2 - 3ml of warm saline.

RECOMMENDATIONS:

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REFERENCES

1. Monoclonal Antibody Production (1999) Institute for Laboratory Animal Research.
2. NHMRC (2001). Guidelines on Monoclonal Antibody Production.
<http://.health.gov.au/nhmrc/research/awc/monosyn.htm>.