

## A MOUSE BLEEDING TECHNIC YIELDING CONSISTENT VOLUME WITH MINIMAL HEMOLYSIS<sup>1,2,3,4</sup>

LANDRUM YOUNG AND TOM R. CHAMBERS

**SUMMARY** • A technic was described which provides a method for obtaining consistently sufficient amounts of whole blood for hematological assay with minimal hemolysis. The mouse was exsanguinated, while under anesthesia, by incising the brachial vessels and collecting the blood in a glass pipette. A consistent volume of 1.0–1.5 ml whole blood yielding 0.4–0.6 ml non-hemolyzed serum can be collected from an adult mouse by a trained technician.

As part of preparations for the Apollo XI quarantine of lunar soil, various bleeding technics for the laboratory mouse, tail vein (1, 2) periorbital venous sinus (2–4), brachial vessels, and heart (4, 5) were evaluated to determine which method yielded the most consistent volume of blood with minimal hemolysis.

Until lunar material was certified as being biologically safe, all work performed was accomplished within biological containment cabinets. (6). It was necessary for any animal manipulation or equipment operation to be accomplished while the technician worked through neoprene gloves. The gloves significantly reduced the technician's dexterity and sense of touch, which necessitated a technically simple method for collecting blood. The method also needed to be rapid, as many exsanguinations and necropsies were to be performed consecutively.

### METHOD

Before exsanguinating the mouse, the tips of 5/4" Pasteur pipettes<sup>5</sup> are broken off and the broken ends of the barrels fire polished. This preliminary procedure increases bore size, thus allowing a more rapid capillary flow to take place and permits collection of the entire sample to proceed before blood clotting. The mouse is anesthetized by inhalation of Metofane<sup>®</sup>.<sup>6</sup> It is important that the animal not be killed or anesthetized too deeply, as this will reduce the blood flow in the brachial vessels. The fore and hind legs are secured to a cork necropsy board. A portion of the skin in the axilla region lateral to the sternum is lifted with forceps and excised with scissors (Fig 1A). This procedure exposes the pectoralis major and latissimus dorsi muscles. (Fig 1B).

The muscles are gently separated from the skin by using the tips of the scissors. This will expose the brachial vessels and also form a pocket between skin and musculature (Fig 1C). A single cut with the scissors will sever the vessels passing into the forelimb. The blood can be collected in the previously-prepared Pasteur pipette by holding it in a horizontal position (Fig 1D). This should enhance capillary flow. It is important that the pipette be filled by capillary flow only. Prob-

<sup>1</sup> From Northrop Services Inc, PO Box 34416, Lunar Receiving Laboratory, NASA Manned Spacecraft Center, Houston, TEX 77034.

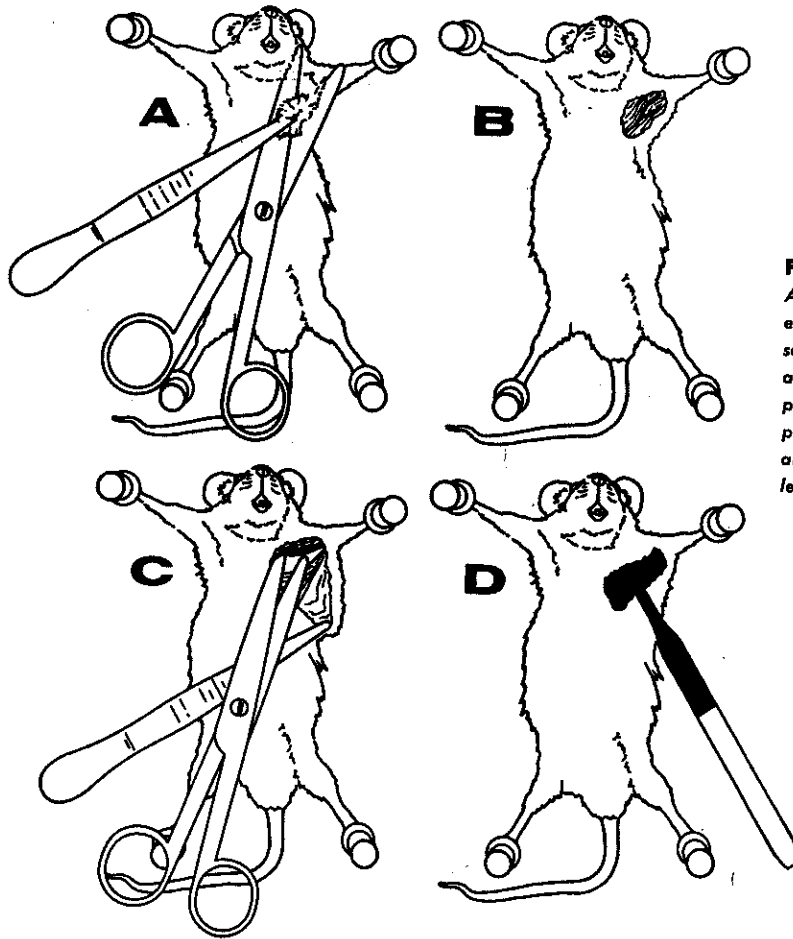
<sup>2</sup> This work was performed at the NASA/MSC Lunar Receiving Laboratory, Houston, TEX.

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<sup>5</sup> Fisher brand<sup>®</sup>. Fisher Scientific Co, Pittsburgh, PA.

<sup>6</sup> Pitman-Moore, Inc, Washington Crossing, NJ.



**Fig 1. Mouse bleeding technic**  
 A) skin in axillary region is lifted with forceps, excised with scissors; B) pectoralis major and latissimus dorsi muscles exposed; C) brachial vessels exposed; note pocket between skin and musculature; D) blood collected in Pasteur pipette.

ing or gouging the axilla area with the tip of the pipette or using suction bulbs to draw the sample up the barrel only increases the degree of hemolysis.

If the mouse has not been killed by exsanguination, it is killed by cervical fracture.

During the actual collection process, samples for hematologic assay can also be directly taken from the site with hematocrit tubes, diluting pipettes, capillary tubes for slides, etc.

#### RESULTS AND DISCUSSION

Collecting whole blood from the brachial vessels proved to be more successful than the other methods previously mentioned because

of the technical simplicity and resulting yields. Whole blood volume averaged 1.0–1.5 ml and serum quality was good. This allowed for all hematologic and serologic assays to be performed easily and accurately. Now that quarantine is no longer required and the use of biological containment cabinets is not needed, this method of exsanguination has proved even more efficient.

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