


Institutional Animal Care & Use Committee		
Title: Use of Tribromoethanol (Avertin)		
Policy#: IACP 027	Date in Effect: 05/10/19	
Rev #: 00	Rev Date:	
In Effect <input checked="" type="checkbox"/> Rescinded <input type="checkbox"/>	Date Rescinded:	

A) BACKGROUND INFORMATION:

- 1) 2,2,2-Tribromoethanol (TBE) in Mice: TBE is an anesthetic agent used in mice. It was once manufactured specifically for use as an anesthetic under the trade name Avertin®, but this product is no longer available. Investigators who wish to use TBE as an anesthetic must make their own solutions.
- 2) TBE is appropriate for short-term procedures in mice, especially surgical procedures. It is best used in situations where it will be given only on a single occasion.
- 3) Justification:
 - a) TBE is considered a non-pharmaceutical grade compound. As such it requires justification and adherence to IACP 015, *Use of Non-Pharmaceutical Grade Compounds and/or Expired Medical Materials*, http://research.utsa.edu/wp-content/uploads/2014/12/IACP_Policy_015.pdf.
 - (1) The exception to IACP 015 is, whereas IACP 015 requires mixed drugs to be discarded after 30 days, TBE, must be discarded 14 days after mixing.

B) PROCEDURES

- 1) Advantages and Disadvantages
 - a) Advantages:
 - (1) TBE induces anesthesia rapidly and provides good surgical analgesia for approximately one hour.
 - (2) Since it is given by injection, one is spared the occupational health risks and technical difficulties associated with volatile anesthetics.
 - (3) If used appropriately, TBE has a good margin of safety.
 - b) Disadvantages:
 - (1) TBE is an irritant, especially at high doses, high concentrations, or with repeated use. Intra-abdominal adhesions may sometimes result after IP injections.
 - (2) TBE degrades in the presence of heat or light to produce toxic byproducts. Degraded solutions can be both nephrotoxic and hepatotoxic. Administration of degraded TBE solutions has been associated with death, often 24 hours after surgery.

- (3) TBE can cause intestinal ileus (stopping of the gut motility and subsequent death of the animal) several weeks after injection. This is more common with TBE stored in the presence of light or heat, stored at higher than recommended doses or when given at higher than recommended concentrations.
 - (4) The effects of TBE are also somewhat unpredictable in mice younger than 16 days, or in animals with altered carbohydrate metabolism, such as various mouse strains used for diabetes or obesity models (db/db mice or ob/ob mice).
- 2) Instructions for Compounding and Use
- a) Chemicals – Two chemicals are necessary to imitate Avertin:
 - (1) 2,2,2 Tribromoethanol and amylene hydrate (tertiary amyl alcohol).
 - (2) Both can be obtainable from Sigma-Aldrich. There may be other sources as well.
 - b) Compounding Ingredients:
 - (1) 2.5 gm 2,2,2 Tribromoethanol.
 - (2) 5 ml 2-methyl-2-butanol (amylene hydrate, tertiary amyl alcohol).
 - (3) 200 ml distilled water - neutral pH.
 - c) Compounding Instructions:
 - (1) Dissolve 2.5 grams TBE in 5 ml of amylene hydrate. This requires heating to approximately 40° Celsius and stirring vigorously.
 - (2) Add distilled water, stirring continuously, up to a final volume of 200 ml.
 - (3) Filter sterilize the solution through a 0.22-micron Millipore filter.
 - (4) Aliquot the final solution into appropriate containers - empty, sterile, red-cap blood collection tubes make a good receptacle, as do brown injection bottles with appropriate caps. It's often easiest to filter the material through a luer-fitted Millipore filter directly into a sterile, red-cap blood collection tube.
 - (5) Refrigerate the aliquots and protect from light. The material degrades rapidly in the presence of heat or light. Even refrigerated and wrapped in foil, the material is stable for only about two weeks. If the material degrades, it becomes toxic.
 - (6) TBE degrades to dibromoacetaldehyde and hydrobromic acid. If the pH of the solution is less than 5, it should be presumed to have degraded. Test the solution by adding one drop of Congo Red to 5 ml of solution. If a purple color results, the solution has degraded and should be discarded. (Note: this method is only useful if the original pH of the solution is greater than 5 - hence the recommendation for neutral distilled water).

- (7) As prepared above, the solution contains 12.5 mg TBE/ml. Do not attempt to make a more concentrated solution - the material is irritating at higher concentrations.
- d) Dosage – Use:
- (1) Mix by stirring or swirling prior to administration.
 - (2) The material is given by IP injection at a dose of 250 mg/Kg. This amounts to 0.5 ml of the above solution to a 25 gm mouse.
 - (3) Induction requires only 1-2 minutes and the righting reflex returns in approximately 40-90 minutes.
- e) Cautions:
- (1) Do not administer non-sterile solutions, outdated solutions, more concentrated solutions, or higher doses than recommended above. Store the solution under refrigeration and in the dark. Containers should be wrapped in aluminum foil.
 - (2) Although some authors report that refrigerated solutions may be kept for months, most authors recommend preparing a new solution every 2 weeks. The IACUC requires discarding refrigerated TBE after 14 days (after mixing).

REFERENCES

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