

Avertin Solution for Mouse Anesthesia

We obtain **2,2,2-tribromoethanol** (Sigma-Aldrich catalog # T48402-5GM) as a powder and **2-methyl-2-butanol, tertiary amyl alcohol 99+ %** (Sigma-Aldrich catalog # 240486-5ML) as a liquid. We store the tribromoethanol powder at -20°C in a sealed foil package, and the tertiary amyl alcohol in the dark at room temperature (organic storage cabinet).

To prepare 1.2% Avertin solution:

Dissolve 2.5g of tribromoethanol into 5ml of tertiary amyl alcohol by warming to 55°C with gentle swirling in a small covered glass flask.

Place a sterile glass beaker containing 200ml of sterile milliQ water and a sterile stir bar on a mixing plate, turn on to create water vortex, and slowly (drop-wise) add with a glass pipet the 5ml of dissolved tribromoethanol-tertiary amyl alcohol mixture into center of swirling water (you may see some crystallization at this point).

Cover beaker with foil lid, and vigorously stir until all traces of crystals disappear. Check pH by paper; should be approximately 7.0 to 7.3. Once fully in solution, pass the working solution through a Nalgene 0.22 micron filter (catalog #569-0020).

Working under a laminar flow hood and using sterile technique, aliquot 5mls of the Avertin working solution into small tubes, cap securely, and cover the entire tube with foil to block light. Store at 4°C for 3-4 months.

To use Avertin solution:

Warm a 1.2% Avertin working solution tube to room temperature prior to use. Discard this tube and any unused Avertin solution at the end of the day. **Remember to maintain sterility to avoid peritonitis.** To prevent decomposition, do not use Avertin if the working solution is older than 4 months, or if the pH drops below 7.0

Administer 0.23ml Avertin per 10 grams bodyweight i.p. using a sterile tuberculin syringe (26g needle). Dosage reference: *Lab Animal Science*, **43**:189-93, 1993.

Avertin (1.2% formulation)	
<u>Bodyweight (grams)</u>	<u>Dose (ml)</u>
23	0.53
24	0.55
25	0.58
26	0.6
27	0.62
28	0.64
29	0.67
30	0.69
31	0.71
32	0.74
33	0.76
34	0.78
35	0.81