

ATCC medium: 1011 Diphasic Blood Agar Medium

Complete Medium

Blood Agar base (ATCC Medium 449).....	700 ml
Rabbit Blood, defibrinated.....	300 ml
Locke's solution (added as an overlay; see below).....	450 ml

Cool ATCC Medium 449 (see below) until it may be held comfortably in the hand. Aseptically add 30% sterile, defibrinated rabbit blood; dispense 5 ml per 16 X 125 mm screw-capped test tube, slant and cool; add 3 ml sterile Locke's Solution overlay (see below).

ATCC Medium 449

(composition per 750 ml)

Meat extract (dry, granulated).....	25.0 g
DI Water.....	250.0 ml

Infuse beef in water by bringing to a rapid boil for 2-3 minutes, stirring constantly. Filter through Whatman #2V filter paper if required and add:

Neopeptone (BD 211681).....	10.0 g
NaCl.....	2.5 g
Agar.....	10.0 g
DI Water to.....	500.0 ml

Adjust pH to 7.2-7.4. Autoclave at 121°C.

Locke's Solution:

NaCl.....	8.0 g
KCl.....	0.2 g
CaCl ₂	0.2 g
KH ₂ PO ₄	0.3 g
Glucose.....	2.5 g
DI Water.....	1000 ml