



OKT 3 | 300499

**Culture Medium** RPMI 1640 (2.0 mM Glucose, 2.0 mM NaHCO<sub>3</sub> (820700a))

**Supplements** 10% FBS

**Subculturing** 1:5

**Freeze medium** RPMI 1640 (2.0 mM Glucose, 2.0 mM NaHCO<sub>3</sub>) + 10% DMSO + 10% FBS

- Thawing and Culturing Cells**
1. Thaw cells in a 37°C water bath.
  2. Centrifuge cells at 300 x g for 3 minutes.
  3. Resuspend cells in 10 ml of culture medium.
  4. Seed cells into a 75 cm<sup>2</sup> flask.
  5. Incubate cells for 15-18 hours.
  6. Seed cells into a 300 x 3 cm<sup>2</sup> flask.
  7. Incubate cells for 10-12 hours.
  8. Harvest cells.

**Incubation Atmosphere** 37°C, 5% CO<sub>2</sub>

**Flask Coating** Cell culture medium

**Freezing Procedure** 1. Seed cells into a 300 x 3 cm<sup>2</sup> flask. 2. Harvest cells.

**Shipping Conditions** 4°C

