

**Data Sheet**

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<b>BCRJ Code:</b>	0035
<b>Cell Line:</b>	AA 4.1
<b>Species:</b>	<i>Rattus norvegicus</i>
<b>Vulgar Name:</b>	Rat
<b>Tissue:</b>	Blood
<b>Cell Type:</b>	Hybridoma: B Lymphocyte
<b>Morphology:</b>	Lymphoblast
<b>Growth Properties:</b>	Suspension
<b>Biosafety:</b>	1
<b>Culture Medium:</b>	RPMI-1640 medium modified to contain 2 mM L-glutamine, 1 mM sodium pyruvate, 4500 mg/L glucose and fetal bovine serum to a final concentration of 10%.
<b>Subculturing:</b>	Cultures can be maintained by addition of fresh medium. Alternatively, cultures can be established by centrifugation with subsequent resuspension at 1 x 10 <sup>5</sup> viable cells/mL. Maintain cultures at a cell concentration between 1 x 10 <sup>5</sup> and 1 x 10 <sup>6</sup> cells/mL. NOTE: Do not allow the cell concentration to exceed 1 x 10 <sup>6</sup> cells/mL.
<b>Subculturing Medium Renewal:</b>	Every 2 to 3 days
<b>Culture Conditions:</b>	Atmosphere: air, 95%; carbon dioxide (CO <sub>2</sub> ), 5% Temperature: 37°C
<b>Cryopreservation:</b>	95% FBS + 5% DMSO (Dimethyl sulfoxide)

**Thawing Frozen Cells:**

**SAFETY PRECAUTION:** It is strongly recommended to always wear protective gloves, clothing, and a full-face mask when handling frozen vials. Some vials may leak when submerged in liquid nitrogen, allowing nitrogen to slowly enter the vial. Upon thawing, the conversion of liquid nitrogen back to its gas phase may cause the vial to explode or eject its cap with significant force, creating flying debris.

1. Thaw the vial by gently agitating it in a 37°C water bath. To minimize contamination, keep the O-ring and cap out of the water. Thawing should be rapid (approximately 2 minutes).
2. Remove the vial from the water bath as soon as its contents are thawed and decontaminate it by dipping in or spraying with 70% ethanol. From this point, all operations must be performed under strict aseptic conditions.
3. For cells sensitive to DMSO, it is recommended to remove the cryoprotective agent immediately. Transfer the vial contents to a centrifuge tube containing 9.0 mL of complete culture medium and centrifuge at approximately  $125 \times g$  for 5 to 7 minutes.
4. Discard the supernatant and resuspend the cell pellet in the recommended complete medium (see specific batch information for the appropriate dilution ratio).
5. Incubate the culture under appropriate atmospheric and temperature conditions (see "Culture Conditions" for this cell line).

**NOTE:** It is important to avoid excessive alkalinity of the medium during cell recovery. To minimize this risk, it is recommended to place the culture vessel containing the growth medium in the incubator for at least 15 minutes before adding the vial contents. This allows the medium to stabilize at its normal pH (7.0 to 7.6).

**Depositors:**

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**Cellosaurus:**[CVCL\\_D664](#)