

Data Sheet

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BCRJ Code:	0010
Cell Line:	2A5
Species:	Mus musculus (B cell); Mus musculus (myeloma), mouse (B cell); mouse (myeloma)
Vulgar Name:	Mouse
Cell Type:	Hybridoma: B Lymphocyte
Morphology:	Lymphoblast
Growth Properties:	Suspension
Derivation:	The cell line was established by Jose A. Lebron and Pamela J. Bjorkman in 1997. Spleen cells were fused with HL-1 Friendly myeloma cells (a derivative of P3X63Ag8.653 that is deficient in both hypoxanthine phosphoribosyl transferase [HPRT] and adenine phosphoribosyl transferase [APRT]).
Applications:	This antibody can be used for ELISA applications and to immunoprecipitate soluble HFE; it is not reactive in Western Blot assays.
Products:	immunoglobulin; monoclonal antibody; against HFE
Biosafety:	1
Additional Info:	The Hybridoma secretes a monoclonal antibody to murine IL10.
Culture Medium:	HL-1 medium supplemented with 4 mM L-glutamine, 1 mM sodium pyruvate and 1% fetal bovine serum. HL-1 medium can be obtained from Lonza (catalog number 77201).
Subculturing:	Cultures can be maintained by addition of fresh medium. Alternatively, cultures can be established by centrifugation with subsequent resuspension at 1 x 10 ⁵ viable cells/mL. Maintain cultures at a cell concentration between 1 x 10 ⁵ and 1 x 10 ⁶ cells/mL. Attached cells may be subcultured by tapping the sides of the flask until cells are dispersed. NOTE: Do not allow the cell concentration to exceed 1 x 10 ⁶ cells/mL.

**Subculturing Medium
Renewal:**

Every 2 to 3 days

Culture Conditions:

Atmosphere: air, 95%; carbon dioxide (CO₂), 5% Temperature: 37°C

Cryopreservation:

95% FBS + 5% DMSO (Dimethyl sulfoxide)

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 × 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).

Thawing Frozen Cells:

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).

References:

Lebron JA, et al. Tolerization of adult mice to immunodominant proteins before monoclonal antibody production. *J. Immunol. Methods* 222: 59-63, 1999. PubMed: 10022372
Lebron JA, et al. Crystal structure of the hemochromatosis protein HFE and characterization of its interaction with transferrin receptor. *Cell* 93: 111-123, 1998. PubMed: 9546397

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Cellosaurus:

[CVCL D143](#)