

Data Sheet

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.

Data Sheet

PAGE 2/3

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)

Thawing Frozen Cells:

SAFETY PRECAUTION: It is highly recommended that protective gloves and clothing always be used and a full face mask always be worn when handling frozen vials. It is important to note that some vials leak when submerged in liquid nitrogen and will slowly fill with liquid nitrogen. Upon thawing, the conversion of the liquid nitrogen back to its gas phase may result in the vessel exploding or blowing off its cap with dangerous force creating flying debris. 1. Thaw the vial by gentle agitation in a 37°C water bath. To reduce the possibility of 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 the contents are thawed, and decontaminate by dipping in or spraying with 70% ethanol. All of the operations from this point on should be carried out under strict aseptic conditions. 3. For cells that are sensitive to DMSO it is recommended that the cryoprotective agent be removed immediately. Transfer the vial contents to a centrifuge tube containing 9.0 mL complete culture medium and spin at approximately 125 x g for 5 to 7 minutes. 4. Discard the supernatant and Resuspend cell pellet with the recommended complete medium (see the specific batch information for the culture recommended dilution ratio). 5. Incubate the culture in an appropriate atmosphere and temperature (see "Culture Conditions" for this cell line). **NOTE:** It is important to avoid excessive alkalinity of the medium during recovery of the cells. It is suggested that, prior to the addition of the vial contents, the culture vessel containing the growth medium be placed into the incubator for at least 15 minutes to allow the medium to reach 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

Depositors:

Pedro Paulo Elsas, Universidade Federal do Rio de Janeiro.

ATCC:

CRL-2444



