



# LO-CD2A HYBRIDOMA

CRL-3535™

## Description

LO-CD2A is a rat cell line that produces LO-CD2a, a rat (IgG2b-Kappa) anti-CD2 monoclonal antibody. This cell line was originally deposited on July 28, 1993, at ATCC (original ATCC accession number HB 11423).

**Organism:** *Rattus norvegicus* (B cell); *Rattus norvegicus* (myeloma), rat (B cell); rat (myeloma)

**Morphology:** lymphoblast-like

**Growth properties:** Suspension

**Cells per vial:** Approximately 5.0 to 7.0 x 10<sup>6</sup>

**Volume:** 1.0 mL

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## Storage Conditions

**Product format:** Frozen

**Storage conditions:** Vapor phase of liquid nitrogen

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## Intended Use

This product is intended for laboratory research use only. It is not intended for any animal or human therapeutic use, any human or animal consumption, or any diagnostic use.

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## BSL 1

ATCC determines the biosafety level of a material based on our risk assessment as guided by the current edition of *Biosafety in Microbiological and Biomedical Laboratories*

(BMBL), U.S. Department of Health and Human Services. It is your responsibility to understand the hazards associated with the material per your organization's policies and procedures as well as any other applicable regulations as enforced by your local or national agencies.

ATCC highly recommends that appropriate personal protective equipment is always used when handling vials. For cultures that require storage in liquid nitrogen, it is important to note that some vials may leak when submersed 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 vial exploding or blowing off its cap with dangerous force creating flying debris. Unless necessary, ATCC recommends that these cultures be stored in the vapor phase of liquid nitrogen rather than submersed in liquid nitrogen.

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## Certificate of Analysis

For batch-specific test results, refer to the applicable certificate of analysis that can be found at [www.atcc.org](http://www.atcc.org).

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## Growth Conditions

**Temperature:** 37°C

**Atmosphere:** 95% Air, 5% CO<sub>2</sub>

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## Handling Procedures

**Complete medium:** The base media for this cell line is RPMI-1640 (ATCC 30-2001). To make the complete media add 2 mM L-glutamine (ATCC 30-2214) to a 1% final

concentration and Fetal Bovine Serum (FBS; ATCC 30-2020) to a 10% final concentration.

### Handling Procedure:

To ensure the highest level of viability, thaw the vial and initiate the culture as soon as possible upon receipt. If upon arrival, continued storage of the frozen culture is necessary, it should be stored in liquid nitrogen vapor phase and not at  $-70^{\circ}\text{C}$ . Storage at  $-70^{\circ}\text{C}$  will result in loss of viability.

1. Thaw the vial by gentle agitation in a  $37^{\circ}\text{C}$  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. Transfer the vial contents to a centrifuge tube containing 9.0 mL complete culture medium. and spin at approximately  $125 \times g$  for 5 to 7 minutes.
4. Resuspend cell pellet with the recommended complete medium (see the specific batch information for the culture recommended dilution ratio). 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 complete 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). pH (7.0 to 7.6).
5. Incubate the culture at  $37^{\circ}\text{C}$  in a suitable incubator. A 5%  $\text{CO}_2$  in air atmosphere is recommended if using the medium described on this product sheet.

### Subculturing procedure:

Cultures can be maintained by addition or replacement of fresh medium. Start cultures at  $5 \times 10^5$  cells/mL and maintain between  $3 \times 10^5$  and  $3 \times 10^6$  cells/mL.

**Medium Renewal:** Add fresh medium every 2 to 3 days (depending on cell density).

Culture Notes:

Do not use stacked vessels and/or roller bottles

Start-Up Seeding Densities:

$1.0 \times 10^6$  viable cells/mL for 3-day growth period.

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2.0 x 10E6 viable cells/mL for 2-day growth period.

Maintain cell density from 1.0 x 10E5 to 1.0 x 10E6 viable cells/mL

Addition and/or replacement of culture medium

## Expansion Comments:

1. CRL-3535 cells are extremely sensitive to overgrowth and media exhaustion. Perform a fluid add every 2 to 3 days.
2. Recommended feeding schedule is Monday, Wednesday, and Friday.
3. Cells are subject to spin down only if media is extraordinary acidic since viability will drop after centrifugation.
4. Cells form small aggregates. These cluster may be broken up by light pipetting.

**Reagents for cryopreservation:** 84% RPMI-1640 + 10% FBS + 1% (2 mM) L-glutamine + 5% DMSO

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## Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: LO-CD2A HYBRIDOMA (ATCC CRL-3535)

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## References

References and other information relating to this material are available at [www.atcc.org](http://www.atcc.org).

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The product is provided 'AS IS' and the viability of ATCC® products is warranted for 30 days from the date of shipment, provided that the customer has stored and handled the product according to the information included on the product information sheet, website, and Certificate of Analysis. For living cultures, ATCC lists the media formulation and reagents that have been found to be effective for the product. While other unspecified media and reagents may also produce satisfactory results, a change in the ATCC and/or depositor-recommended protocols may affect the

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Product Sheet

Please see the material transfer agreement (MTA) for further details regarding the use of this product. The MTA is available at [www.atcc.org](http://www.atcc.org).

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