CTLL-2

TIB-214™

Description

CTLL-2 is a clone of cytotoxic T cells derived from a C57BL/6 mouse. The cells are dependent upon Interleukin-2 (IL-2) for growth and can be used to assay for IL-2.
Organism: *Mus musculus*, mouse
Cell Type: cytotoxic T lymphocyte
Morphology: lymphoblast
Growth properties: Suspension

Storage Conditions

Product format: Frozen
Storage conditions: Vapor phase of liquid nitrogen

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.

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.

Certificate of Analysis

For batch-specific test results, refer to the applicable certificate of analysis that can be found at www.atcc.org.

Growth Conditions

Temperature: 37°C
Atmosphere: 95% Air, 5% CO_2

Handling Procedures

Unpacking and storage instructions:

1. Check all containers for leakage or breakage.
2. Remove the frozen cells from the dry ice packaging and immediately place the cells at a temperature below -130°C, preferably in liquid nitrogen vapor, until ready for use.

Complete medium: The base medium for this cell line is ATCC-formulated RPMI-1640 Medium, (ATCC 30-2001). To make the complete growth medium, add the following

components to the base medium: additional 2 mM L-glutamine; additional 1mM sodium pyruvate; adjust to a final concentration of 10% fetal bovine serum and 10% T-STIM with Con A. T-STIM is available from Becton Dickinson.

Handling Procedure:

To insure 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°C. Storage at -70°C will result in loss of viability.

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. It is recommended that the cryoprotective agent be removed immediately. Centrifuge the cell suspension at approximately 125 x g for 5 to 10 minutes. Discard the supernatant and resuspend the cell pellet in an appropriate amount of fresh growth medium. Adjust the cell density of the suspension to 1 X 10^5 viable cells/mL.
4. Transfer cells to an appropriate size vessel. 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).
5. Incubate the culture at 37°C in a suitable incubator. A 5% CO_2 in air atmosphere is recommended if using the medium described on this product sheet.

Subculturing procedure:

Subculture actively growing suspension cultures before they have reached 2 X 10^5 cells/ml or the IL-2 will rapidly deplete and the cells will quickly lose viability Use inoculation densities of 1 to 2 X 10^4 viable cells/mL. Corning® T-75 flasks (catalog #431464) are recommended for subculturing this product.

Medium Renewal: Twice per week

Some Important Considerations in Handling CTLL-2, TIB-214

Frozen Cells: Viability immediately after thawing will be 70-80%. Expect viability to be very poor from day 1 to day 4 after culture initiation. Culture will appear to be completely dead. On the third to fifth day following initiation viable cell clusters will begin to appear in suspension. Usually cells will be ready to subculture on the 7th to the 10th day after the ampule is thawed. However, it may take from 2-3 weeks before vigorous growth is observed. It is best to leave the initial culture undisturbed until cells enter their growth phase.

Overgrowth: In the event cell density becomes too great and viability decreases to where culture appears totally dead, the culture may still be rescued. Inoculate a flask at a density of 1 X 10^4 viable cells/mL.

Reagents for cryopreservation: Complete growth medium supplemented with 10% (v/v) FBS and 7.5% (v/v) DMSO (ATCC 4-X)

Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: CTLL-2 (ATCC TIB-214)

References

References and other information relating to this material are available at www.atcc.org.

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