cyagen | OriCell

User Manual

OriCell™ A20 Mouse B-Cell Lymphoma Cell Line

Catalog No. M5-1601

We help you discover life

Introduction

The A20 mouse B-cell lymphoma cell line is derived from a Balb/c B-cell lymphoma found in aged Balb/cAnN mice with a spontaneously occurring reticulum cell tumor. It is diploid, with a chromosome number ranging from 33 to 38 and a modal number of 37.

OriCell™ A20 Mouse B-Cell Lymphoma Cell Line is widely used in immunology, oncology, and cell biology research. They are commonly employed in studies of mouse B-cell lymphoma, including investigations into the effects of transforming growth factor-beta (TGF-β) or ganoderic acid on these cells in lymphoma models.

Note: **This product is intended for research use only and is not for diagnostic, therapeutic, clinical, household, or any other applications.**

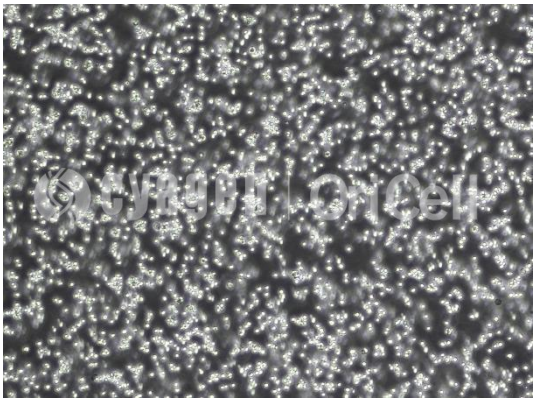
When citing our products in academic publications, please use the following format: "OriCell™ [Product Name] + [Catalog Number], from Cyagen Biosciences."

Product Information

Product Name	OriCell™ A20 Mouse B-Cell Lymphoma Cell Line
Alternative Name	A-20
Catalog Number	M5-1601
Amount of Cells	1×10^6 cells/vial
Tissue Origin	Mouse B Lymphoma
Cell Characteristics	Suspension Growth; Lymphoblastic-like
Culture Conditions	95% air; 5% CO_2; 37 °C
Culture Medium	RPMI-1640 + 10% FBS + 0.05 mM β-mer
Doubling Time	48 ~ 72 h
Biosafety Level	1
Storage at	Liquid Nitrogen (-196 °C)
Precautions	——

Note: This product is manufactured under strict aseptic conditions. You may choose to add antibiotics during subsequent culturing based on your specific needs.

The Morphology of OriCell™ A20 Mouse B-Cell Lymphoma Cell Line



Tumorigenicity Validation Data

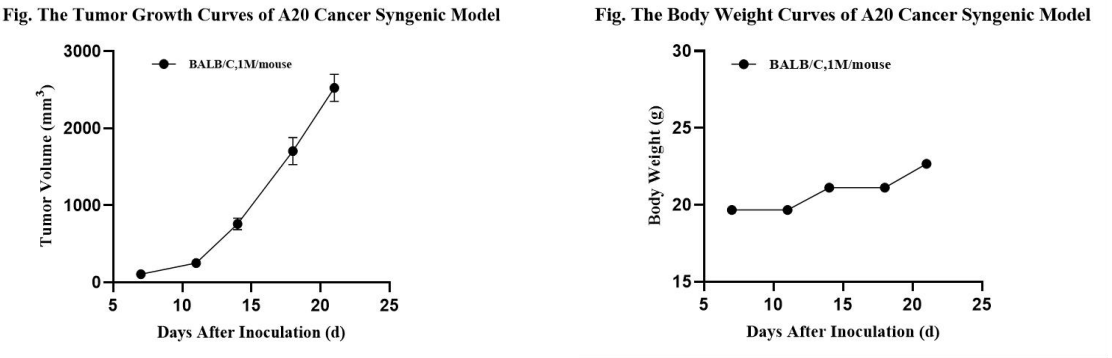


Figure 1. Tumor volume growth curve and body weight changes in mice bearing subcutaneous A20 mouse B cell lymphoma xenografts (n = 9).

A20 cells were injected subcutaneously into 7-week-old BALB/c mice, and tumor volumes were measured at different time points. Each mouse received 1×10^6 cells, and data are presented as Mean± SEM. The results indicate that A20 cells readily form tumors in BALB/c mice. Tumor volumes are expected to reach 100–200 mm^3 by day 6 post-injection and 2,000 mm^3 at the experimental endpoint on day 21, with an anticipated drug treatment window of approximately 15 days.

QC

- Pass the detection of bacteria, fungi, mycoplasma, and endotoxins.
- Verified by cell recovery viability testing.
- Verified by STR analysis.

Please refer to "COA" for details.

General Handling Principles

1. Maintain strict aseptic technique. Ensure complete sterility throughout all procedures, particularly within the laminar flow hood and incubator.
2. Follow standardized protocols. Adhere strictly to the product manual. Implement rigorous control over experimental variables and include appropriate parallel controls.
3. Use high-quality consumables and reagents. This product requires culture vessels suitable for suspension growth, and the reuse of these vessels is not recommended. The reagents used must be validated for reliability, cell compatibility, and batch-to-batch consistency.

Note: **The cryopreservation medium of this product contains DMSO, which may pose potential risks. Please handle it with care.**

* The abbreviations used are defined as follows:

Abbreviation	Name	Cat. No.
FBS	Fetal Bovine Serum	See official website
BCS	Bovine Calf Serum	SBCST-01001
Glu	Glutamine	SGLU-10201
SP	Sodium Pyruvate	SCSP-10301
Dex	Dexamethasone	SDEX-10401
NBCS	Newborn Calf Serum	NCSST-01001
HS	Horse Serum	SCHST-01001
NEAA	Non Essential Amino Acid	NEAA-10201
β-mer	β-mercaptoethanol	BMER-10301
P/S	Penicillin- Streptomycin	ATPS-10001
ITS	Insulin, Transferrin, Selenite	ITSS-10201

Thawing and Culturing of Cells

Materials Required

- OriCell™ A20 Mouse B-Cell Lymphoma Cell Line (Cat. No.: M5-1601)
- OriCell™ Complete Medium For A20 Cell Line (Cat. No.: CMM5-1601)

Steps

Note: **If thawing is planned within 24 hours of receipt, store the cells in an ultra-low temperature freezer at -80 °C. For long-term storage (>24 hours) , keep them in liquid nitrogen. Before thawing, transfer the cells from liquid nitrogen to -80 °C and hold them there for 10 minutes. This will allow any residual liquid nitrogen to evaporate and prevent vial explosion.**

1. Pre-warm the water bath to 37 °C.
2. Warm the complete medium to 37 °C.
3. Add at least 5 mL of pre-warmed complete medium to a 15 mL centrifuge tube for subsequent use.
4. Remove the cryovial containing cells from the −80 °C freezer, immerse it in the 37 °C water bath, and gently and quickly swirl to thaw the cryopreservation medium.

   Note:

   (1) Gently swirl the cryovial during thawing to ensure rapid and uniform thawing.

   (2) Avoid submerging the cap in water to prevent contamination.

   (3) Stop thawing in the water bath when only a single ice crystal (approximately 2 mm in diameter) remains, then continue gently swirling the vial until it is completely thawed.

5. Wipe the outer surface of the cryovial with 75% ethanol.
6. In a biosafety cabinet, open the cryovial and transfer the cell suspension to the prepared centrifuge tube using a Pasteur pipette.

7. Rinse the cryovial once with 1 mL of complete medium to collect residual cells and minimize loss.
8. Centrifuge the cell suspension at 140 × g for 5 minutes.

   Note: **Please calculate the corresponding rotational speed using the formula: RCF = 1.118 × 10^{-5} × r × RPM^2 (where RCF is the relative centrifugal force, r is the rotor radius in cm, and RPM is the rotational speed).**

9. Carefully remove the supernatant after centrifugation. Add 2 mL of complete medium, gently resuspend the cell pellet by pipetting up and down to mix thoroughly.
10. Seed the cells into a T25 flask or culture vessel with an equivalent growth surface area. Add sufficient complete medium so that the total volume in a T25 flask is no less than 5 mL.
11. Gently swirl the flask to evenly distribute the cells, then incubate in a CO_2 incubator at 37 °C with 5% CO_2 and saturated humidity.
12. On the day after recovery, observe cell status and either replace the medium with fresh complete medium or passage the cells as needed.
13. Refresh the complete medium every 3 days until the cells reach an appropriate density, at which point passage is required.

Medium Replacement

Materials Required

- OriCell™ Complete Medium For A20 Cell Line (Cat. No.: CMM5-1601)

Steps

Note: **Avoid repeatedly warming the entire medium. Aliquot it into appropriate sterile containers and prewarm only the amount needed for the day.**

1. Observe the cells under a microscope. If cells have adhered to the surface, avoid tapping the culture vessel to prevent detachment.

2. Transfer the cell suspension to a centrifuge tube using a Pasteur pipette.

3. Centrifuge at 140 × g for 4 minutes to pellet the cells. Remove the supernatant.

4. Resuspend the cell pellet in 1 mL of complete medium. Gently mix to obtain a uniform suspension.

5. Transfer the resuspended cells to a new culture vessel.

6. Add an appropriate volume of culture medium to cover the cells and incubate at 37 °C in a humidified incubator with 5% CO_2.

7. Subsequently, change the medium or passage the cells according to medium condition and cell growth. Generally, the medium should be changed every two days.

Passaging Timing

Under standard conditions, the OriCell™ A20 Mouse B-Cell Lymphoma Cell Line is typically subcultured every 2–3 days.

Passaging of Cells

Materials Required

- OriCell™ Phosphate-Buffered Saline Solution (1X) (Cat. No.: PBS-10001)
- OriCell™ Complete Medium For A20 Cell Line (Cat. No.: CMM5-1601)

Steps

1. Prewarm the complete medium to 37 °C.
2. Transfer the medium from the culture vessel to a centrifuge tube using a Pasteur pipette.

Wash the culture vessel once with PBS (approximately 3 mL for a T25 flask or 6 mL for a T75 flask) to collect residual cells.

3. Centrifuge all collected cell suspensions at 140 × g for 5 minutes.
4. Carefully remove the supernatant after centrifugation. Add 2 mL of complete medium and gently resuspend the cell pellet by pipetting up and down to thoroughly mix.
5. Seed the cells into a suitable culture vessel at a density of (2 – 3) × 10^4 live cells/cm^2.

   Note: **We recommend manual cell counting when conditions permit and counting efficiency is high, in order to obtain an accurate cell concentration to guide seeding. If precise counting is not feasible, subculturing at an appropriate ratio is a reliable alternative. Typically, A20 cells are passaged at a ratio of 1:3 to 1:4, with cells reaching passage confluence within 72 hours. Please adjust the subculture ratio according to the actual condition of the cells.**

6. Gently agitate the vessel to ensure uniform cell distribution and place it in an incubator at 37 °C, 5% CO_2, and saturated humidity.
7. On the day after passaging, observe the cell condition. If a significant number of floating cells are present, replace the culture medium.
8. Refresh the complete medium every 3 days until the cells reach an appropriate density, at which point passage or cryopreservation is required.

Cryopreservation of Cells

Materials Required

- OriCell™ NCR Protein-Free Cryopreservation Medium For General Use (Cat. No.: NCPF-10001)
- OriCell™ NCR Cryopreservation Medium For General Use (Cat. No.: NCRC-10001)

Steps

1. Cells should be cryopreserved once they reach an appropriate density suitable for passaging.
2. For cell digestion, please refer to passaging steps 1-3 above.
3. Carefully remove the supernatant after centrifugation and gently resuspend the cells in an appropriate volume of cryopreservation medium.
4. Aliquot the cells into cryovials according to the desired cell number or proportion.

   Note: **If accurate cell counting is not feasible, we recommend aliquoting cells proportionally for freezing. Prolonged exposure to non-culture conditions will significantly compromise cell viability. Maintain the cells at 4 °C during counting to minimize metabolic activity and preserve cell integrity.**

5. When using any of the recommended NCR cryopreservation media above, cryovials can be directly placed individually into a -80 °C freezer.

   Note: **Avoid opening the freezer door during the first 4 hours of freezing, as temperature fluctuations can adversely affect cell viability.**

6. After approximately 8 hours, transfer the cryovials to liquid nitrogen for long-term storage.

   Note: **Do not store the cryovials at -80 °C for more than 48 hours.**