It is important to note that some vials leak when submersed in liquid nitrogen.

Incubate the culture at 37°C in a suitable incubator. A 5% CO₂ gas phase may result in the vessel exploding or blowing off its cap with dangerous force creating flying debris.

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. 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. Resuspend cell pellet with the recommended complete medium (see the specific batch information for the culture recommended dilution ratio), and dispense into a 25 cm² or a 75 cm² culture flask. 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).
5. Incubate the culture at 37°C in a suitable incubator. A 5% CO₂ in air atmosphere is recommended if using the medium described on this product.

Cultures can be maintained by addition or replacement of medium. Start new cultures at 2 X 10⁶ viable cells/mL and subculture at 1 X 10⁷ cells/mL.

Medium Renewal: Every 2 to 3 days
Complete culture medium described above supplemented with 5% (v/v) DMSO. Cell culture tested DMSO is available as ATCC Catalog No. 4-X.

This clone, picked from soft agar, was selected for its ability to undergo eosinophilic differentiation when treated with butyric acid. Initially, 80% of the treated cells will differentiate as eosinophils. However, with continued passage the line will gradually exhibit a modest degree of reversion such that a higher percentage of the cells will exhibit neutrophilic differentiation. The cells express IL-5 receptors and eosinophil granule proteins, and exhibit chemotaxis to eosinophil specific eosinophilotactins.

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

Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the current publication of the Biosafety in Microbiological and Biomedical Laboratories from the U.S. Department of Health and Human Services Centers for Disease Control and Prevention and National Institutes for Health.

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Additional information on this culture is available on the ATCC web site at www.atcc.org.

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