Resuspend cell pellet with the recommended complete medium (see the specific batch information for more details).

Remove the vial from the water bath as soon as the contents are thawed, and decontaminate by dipping or spraying with 70% ethanol. All of the operations from this point on should be carried out under strict aseptic conditions.

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.

Resuspend cell pellet with the recommended complete medium (see the specific batch information for more details). 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).

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 sheet.

Handling Procedure for Flask Cultures

The flask was seeded with cells (see specific batch information), grown, and completely filled with medium at ATCC to prevent loss of cells during shipping.

1. Upon receipt visually examine the culture for macroscopic evidence of any microbial contamination. Using an inverted microscope (preferably equipped with phase-contrast optics), carefully check for any evidence of microbial contamination.
2. Incubate the flask in an upright position for several hours at 37°C. After the temperature has equilibrated, aseptically remove the entire contents of the flask and centrifuge at 125 x g for 5 to 10 minutes. Remove shipping medium and save for reuse. Resuspend the cell pellet in 10 mL of this medium.
3. From this cell suspension remove a sample for a cell count and viability. Adjust the cell density of the suspension to 1 x 10⁶ viable cells/mL in the shipping medium.
4. Incubate the culture, horizontally, at 37°C in a 5% CO₂ in air atmosphere. Maintain the cell density of the culture as suggested under the subculture procedure.

Subculturing Procedure

Cultures can be maintained by addition or replacement of fresh medium. Subculture every two days at 2.5 x
**Intended Use**

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

**Complete Growth Medium**

The base medium for this cell line is ATCC-formulated RPMI-1640 Medium, Catalog No. 30-2001. To make the complete growth medium, add the following components to the base medium:

- 0.05 mM 2-mercaptoethanol
- 62 ng/ml human recombinant macrophage colony stimulating factor (M-CSF)
- Fetal bovine serum to a final concentration of 10%.

**Citation of Strain**

If use of this culture results in a scientific publication, it should be cited in that manuscript in the following manner: M-NFS-60 (ATCC® CRL-1838™)

**Cryopreservation Medium**

Complete culture medium described above supplemented with 5% (v/v) DMSO. Cell culture tested DMSO is available as ATCC Catalog No. 4-X.

**Comments**

The cells are responsive to both interleukin 3 (interleukin-3, IL-3) and macrophage colony stimulating factor (M-CSF).

The cells contain a truncated c-myb proto-oncogene caused by integration of a retrovirus.

**References**

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

**Biosafety Level:** 1

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.

**ATCC Warranty**

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Please see the enclosed Material Transfer Agreement (MTA) for further details regarding the use of this product. The MTA is also available on our Web site at [www.atcc.org](http://www.atcc.org).

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

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