Organism: Homo sapiens, human
Tissue: skin
Disease: melanoma
Cell Type: epithelial
Morphology: epithelial
Growth Properties: adherent

Refer to the Certificate of Analysis for batch-specific test results.

ATCC highly recommends that protective gloves and clothing always be used and a full face mask always be worn when handling frozen vials. It is important to note that some vials leak when submerged 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 vessel exploding or blowing off its cap with dangerous force creating flying debris.

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.

1. Remove and discard culture medium.
2. Briefly rinse the cell layer with 0.25% (w/v) Trypsin - 0.53 mM EDTA solution to remove all traces of serum which contains trypsin inhibitor.
3. Add 2.0 to 3.0 mL of Trypsin-EDTA solution to flask and observe cells under an inverted microscope until cell layer is dispersed (usually within 5 to 15 minutes).
Note: To avoid clumping do not agitate the cells by hitting or shaking the flask while waiting for the cells to detach. Cells that are difficult to detach may be placed at 37°C to facilitate dispersal.
4. Add 6.0 to 8.0 mL of complete growth medium and aspirate cells by gently pipetting.
5. Add appropriate aliquots of the cell suspension to new culture vessels.
6. Incubate cultures at 37°C.

Subcultivation Ratio: A subcultivation ratio of 1:6 to 1:10 is recommended
Medium Renewal: Two to three times weekly

The cells originally tested positive for mycoplasma. After treatment with ciprofloxacin, the cells have remained negative for mycoplasma for 60 days.

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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media or the absence of an additive from the ATCC recommended media may affect recovery, growth and/or function of this product. If an alternative medium formulation is used, the ATCC warranty for viability is no longer valid.

**Disclaimers**

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**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 Eagle's Minimum Essential Medium (EMEM; ATCC 30-2003). Add the following components to 500 mL base medium:

- 5.6 mL HEPES (1 M stock, Gibco catalog # 15630-080)
- 56 mL FBS (ATCC 30-2020)

**Citation of Strain**

If use of this culture results in a scientific publication, it should be cited in that manuscript in the following manner: HMCB [Human Melanoma Cell Bowes] (ATCC® CRL-9607™)