



## Product Sheet

# M18/2.a.12.7 (new clone of M18/2.a.8) (ATCC® TIB-218™)

### Please read this FIRST



Storage Temp.  
**liquid nitrogen  
vapor phase**

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Biosafety Level  
**1**

### 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:

- fetal bovine serum to a final concentration of 10%
- 0.05 mM 2-mercaptoethanol

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### Citation of Strain

If use of this culture results in a scientific publication, it should be cited in that manuscript in the following manner: M18/2.a.12.7 (new clone of M18/2.a.8) (ATCC® TIB-218™)

American Type Culture Collection  
PO Box 1549  
Manassas, VA 20108 USA  
[www.atcc.org](http://www.atcc.org)

800.638.6597 or 703.365.2700  
Fax: 703.365.2750  
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Or contact your local distributor

### Description

**Organism:** *Rattus norvegicus* (B cell); *Mus musculus* (myeloma), rat (B cell); mouse (myeloma)

**Isotype:** rat IgG2a kappa

**Tissue:** spleen

**Cell Type:** hybridoma: B lymphocyte

**Morphology:** lymphoblast

**Growth Properties:** suspension

### Batch-Specific Information

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

### SAFETY PRECAUTION

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 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 vessel exploding or blowing off its cap with dangerous force creating flying debris.

### Unpacking & 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  $-130^{\circ}\text{C}$ , preferably in liquid nitrogen vapor, until ready for use.

### Handling Procedure for Frozen Cells

#### Handling Procedure for Frozen Cells

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  $-70^{\circ}\text{C}$ . Storage at  $-70^{\circ}\text{C}$  will result in loss of viability.

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1. Thaw the vial by gentle agitation in a  $37^{\circ}\text{C}$  water bath. To reduce the possibility of contamination, keep the O-ring and cap out of the water. Thawing should be rapid (approximately two 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 growth medium and spin at approximately  $125 \times g$  for 5 to 7 minutes.
4. Resuspend cell pellet with the recommended complete growth medium (see the specific batch information for the culture recommended dilution ratio) and dispense into a  $25 \text{ cm}^2$  or a  $75 \text{ cm}^2$  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  $37^{\circ}\text{C}$  in a suitable incubator. A 5%  $\text{CO}_2$  in air atmosphere is recommended if using the medium described on this product.

### Subculturing Procedure

**Protocol:** Cultures can be maintained by the addition of fresh medium or replacement of medium.

Alternatively, cultures can be established by centrifugation with subsequent resuspension at  $1$  to  $2 \times 10^5$  viable cells/ml.

**Interval:** Maintain cell density between  $1 \times 10^5$  and  $1 \times 10^6$  viable cells/ml.


**Medium Renewal:** Every 2 to 3 days



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## Cryopreservation Medium

### Cryoprotectant Medium

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



### Comments

Animals were immunized with mouse cytotoxic T cell glycoproteins.

Spleen cells were fused with P3X63Ag8.653 myeloma cells.

The Mac-1 antigen is a macrophage differentiation antigen associated with type three complement receptor (CR3).

The LFA-1 antigen is associated with antigen specific T lymphocyte mediated cell killing.

The LFA-1 and Mac-1 antigens are composed of two chains or subunits, alpha and beta.

The beta chain of LFA-1 (Leukocyte Function Associated antigen) is immunologically identical to the beta chain of the Mac-1 antigen.

Tested and found negative for ectromelia virus (mousepox).

The line was recloned by TA Springer in 1988, and a new seed stock was created at the ATCC.



### 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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### Disclaimers

This product is intended for laboratory research purposes only. It is not intended for use in humans.

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Additional information on this culture is available on the ATCC web site at [www.atcc.org](http://www.atcc.org).

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