



Product Sheet

## L8 (ATCC® CRL-1769™)

Please read this FIRST



### 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

A 4:1 mixture of Dulbecco's modified Eagle's medium and Medium 199, 89%; chicken embryo extract, 1%; horse serum, 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: L8 (ATCC® CRL-1769™)

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*, rat

**Disease:** Carcinogen

**Cell Type:** myoblast myoblast

**Age:** newborn

**Morphology:** fibroblast

**Growth Properties:** adherent

### 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°C, preferably in liquid nitrogen vapor, until ready for use.

### Handling Procedure for Frozen Cells

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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.

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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.

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 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 xg 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 cm<sup>2</sup> or a 75 cm<sup>2</sup> 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% CO<sub>2</sub> in air atmosphere is recommended if using the medium described on this product.

### Subculturing Procedure

**Subcultivation Ratio:** An inoculation density of 4000 viable cells per sq. cm. of flask or dish surface area is recommended

**Medium Renewal:** Twice per week

Remove medium, rinse the cell sheet once with fresh trypsin (0.25%), EDTA (0.03%) solution, remove trypsin and allow the flask to sit at room temperature (or at 37°C) until the cells detach (about 10 minutes).

Add fresh medium, aspirate and dispense into new flasks at a density of 2000 cells/sq. cm.



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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

This line was originally isolated by D. Yaffe in 1969 from primary rat skeletal muscle cultures; unlike the L6 cell line (ATCC CRL-1458) no carcinogen was used to establish the L8 line; upon becoming confluent, L8 will fuse to form cross striated multinucleated muscle fibers; it is recommended that early passages be preserved in liquid nitrogen and that the line be recloned periodically and reselected for progeny that have the ability to fuse; it is important that the cells be subcultured when the flask is about 60% confluent. The myoblastic population will be depleted if the cultures are allowed to become confluent since most of the cells will fuse into nondividing syncytia. To avoid this, one must subculture before the cultures become confluent and should reclone periodically and select myoblastic clones.



### 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

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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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