

# Sf9-ET

# **CRL-3357**<sup>™</sup>

# Description

Sf9-ET is a cell line with rounded morphology that was isolated in 1993 from the ovary of a fall armyworm (Spodoptera frugiperda). The Sf-9 cell line has been modified to express eGFP upon infection with baculovirus. It was deposited by the Frederick National Laboratory for Cancer Research.

**Organism:** Spodoptera frugiperda, fall armyworm

**Tissue:** Ovary **Age:** pupa

**Gender:** Female

Morphology: rounded

**Growth properties:** Adherent and/or suspension

Volume: 1.0 mL

# **Storage Conditions**

**Product format:** Frozen

**Storage conditions:** Vapor phase of liquid nitrogen

#### Intended Use

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

### BSL<sub>1</sub>

ATCC determines the biosafety level of a material based on our risk assessment as guided by the current edition of *Biosafety in Microbiological and Biomedical Laboratories* 



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(BMBL), U.S. Department of Health and Human Services. It is your responsibility to understand the hazards associated with the material per your organization's policies and procedures as well as any other applicable regulations as enforced by your local or national agencies.

ATCC highly recommends that appropriate personal protective equipment is always used when handling vials. For cultures that require storage in liquid nitrogen, it is important to note that some vials may 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 vial exploding or blowing off its cap with dangerous force creating flying debris. Unless necessary, ATCC recommends that these cultures be stored in the vapor phase of liquid nitrogen rather than submersed in liquid nitrogen.

# Certificate of Analysis

For batch-specific test results, refer to the applicable certificate of analysis that can be found at www.atcc.org.

### **Growth Conditions**

Temperature: 27°C Atmosphere: 100% Air

# Handling Procedures

### **Unpacking and 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.

### **Complete medium:**

Complete medium: Complete medium for this cell line is SF-X Insect Medium (Hyclone Cat# SH30278.02) + 50 ug/mL G418.

Due to the limited stability, the required G418 should be added to an aliquot of the SF-X Insect Medium only prior to seeding or performing fluid changes. Media supplemented with G418 is only stable for IO days.

 Use 1 uL of 50 mg/mL G418 (Gibco catalog # 10131-035) per mL of complete basal medium

**Note:** Do not use G-418 with other antibiotics/antifungals (ie. Pen/Strep). These agents are competitive inhibitors of G418.

### **Handling Procedure:**

To ensure 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 27°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 centrifuge at approximately 150 to 400 x g for 8 to 10 minutes (280 x g for 10 minutes).
- 4. Resuspend cell pellet with the recommended complete medium (see the lot specific information provided on the Certificate of Analysis for the culture recommended dilution ratio) and dispense into an appropriate size culture flask (recommended on the Certificate of Analysis). The recommended seeding density for CRL-3357 is  $5.0 \times 10^4$  to  $1.0 \times 10^5$  viable cells/mL. 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 27°C in a suitable incubator. A 100% air atmosphere is recommended if using the medium described on this product sheet.

### **Subculturing procedure:**

Volumes used in this protocol are for 75 cm<sup>2</sup> flask; proportionally reduce or increase amount of dissociation medium for culture vessels of other sizes. Non-vented flasks (Falcon 353135 or Corning<sup>®</sup> 431081) with plug caps are recommended for culturing this product.

- 1. Remove and discard culture medium.
- 2. Briefly rinse the cell layer with DPBS (ATCC catalog # 30-2200) to remove all traces of serum that contains trypsin inhibitor.
- 3. To detach cells, bang and or pipet fresh culture media over cell monolayer. No rinsing reagent, dissociation agent or neutralizing agent are recommended. Cells that are difficult to detach may be placed at 27°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. Cultures can be established between  $4.0 \times 10^4$  and  $8.0 \times 10^4$  viable cells/cm<sup>2</sup>.
- 6. Incubate cultures at 27°C.

**Interval:** Maintain cultures at a cell concentration between  $4.0 \times 10^4$  and  $2.8 \times 10^5$  cell/cm<sup>2</sup>.

Subcultivation Ratio: A subcultivation ratio of 1:8 to 1:10 is recommended

Medium Renewal: 2 to 3 times per week

Reagents for cryopreservation: Complete growth medium supplemented with 10%

(v/v) FBS and 10% (v/v) DMSO (ATCC 4-X)

#### Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: Sf9-ET (ATCC CRL-3357)

### Sf9-ET CRL-3357

#### References

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

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