



Product Sheet

Swiss SFME (Serum Free Mouse Embryo) (ATCC® CRL-9391™)

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

Dulbecco's modified Eagle's medium with 4.5 g/L glucose, 50%; Ham's F12 medium, 50%, Supplements: 0.01 mg/ml bovine insulin, 0.025 mg/ml human transferrin, 0.02 mg/ml human high density lipoprotein (HDL), 100 ng/ml mouse epidermal growth factor (EGF), 10 nM sodium selenite, 1 unit/ml human platelet derived growth factor (PDGF), and 15 mM HEPES. NOTE: HDL is available from Sigma Chemical Co. (catalog number L2014).

Citation of Strain

If use of this culture results in a scientific publication, it should be cited in that manuscript in the following manner: Swiss SFME (Serum Free Mouse Embryo) (ATCC® CRL-9391™)

American Type Culture Collection
PO Box 1549
Manassas, VA 20108 USA
www.atcc.org

800.638.6597 or 703.365.2700
Fax: 703.365.2750
Email: Tech@atcc.org

Or contact your local distributor

Description

Organism: *Mus musculus*, mouse

Strain: Swiss

Cell Type: Astrocyte

Age: embryo; 16 days

Morphology: fibroblast

Growth Properties: clusters in suspension; the cells will attach to flasks coated with 0.02 mg/ml human fibronectin

Cytogenetic Analysis: modal number = 40; range = 39 to 42

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

HANDLING PROCEDURE FOR FROZEN CELLS

- Initiate culture as soon as possible upon receipt.
- Thaw by rapid agitation in 37°C water bath. Thawing should be rapid (within 40-60 seconds). As soon as the ice is melted, remove the ampule from the water bath and immerse in 70% ethanol at room temperature. All of the operations from this point on should be carried out under strict aseptic conditions.
- The cells are supplied in two different types of glass ampules. One is a standard ampule, the neck of which must be scored with a sharp file that has been immersed in ethanol. A definitive sharp nick about 1/8" in length on one side is necessary. The second type is prescored and is identifiable by a gold band around the ampule neck, and should not be scored with a file.
- Break the neck of the ampule between several folds of a sterile towel.
- Transfer the cell suspension and dilute it with the recommended culture medium in a centrifuge tube (see specific batch information above for dilution ratio). Centrifuge the cells at 200 x g for 10 minutes, resuspend in 8 ml of medium and dispense into a 25 cm. sq. flask. Incubate at 37°C with 5% CO₂ in air atmosphere. Since it is important to avoid excessive alkalinity of the medium during recovery of the cells, it is suggested that the culture medium be placed into the culture flask, tube, etc. and the pH be adjusted, as necessary, prior to the addition of the ampule contents. Note that the bicarbonate content of the culture medium will determine whether an atmosphere containing CO₂ will be required.

FLUID RENEWAL

Twice weekly.

SUBCULTURE PROCEDURE

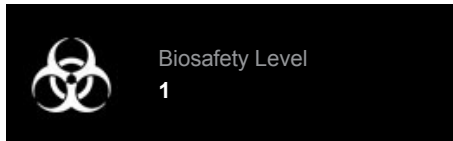
Attached cells may be removed with 0.25% trypsin - 0.02% EDTA. Once the cells have loosened, disperse the cells in an equal volume of medium plus soybean



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trypsin inhibitor (0.1%), centrifuge to collect cells, resuspend in fresh medium and dispense into new flasks. Floating cells can be transferred by pipetting.

Subcultivation ratio: 1:10 to 1:100.



Handling Procedure for Flask Cultures

HANDLING PROCEDURE FOR FLASK CULTURES (MIXED MONOLAYER and SUSPENSION)

The flask was seeded with cells (see specific batch information above for concentration), grown and completely filled with medium to prevent loss of cells in transit. Upon receipt incubate the flask in an upright position for several hours to return the flask contents to 37°C. After the temperature has equilibrated, aseptically remove the entire contents of the flask and centrifuge at 300 x g for 15 minutes. Draw off the excess supernatant medium, resuspend the cell pellet in 10-12 ml of the shipping medium and return the cells to the shipping flask. Incubate the culture in a flat position at 37°C in a 5% CO₂ in air atmosphere. Maintain the cell density of the culture as suggested under the subculture procedure described above.



Subculturing Procedure

Subcultivation Ratio: A subcultivation ratio of 1:10 to 1:100 is recommended

Medium Renewal: Twice per week

Remove medium, and collect suspended clusters by centrifugation.

To disperse clusters or to remove adherent cells, add fresh 0.25% Trypsin, 0.02% EDTA at room temperature until the cells disperse (2 to 3 minutes).

Add an equal volume of fresh serum free medium plus 0.1% soybean trypsin inhibitor and centrifuge at 1000 X g.

Resuspend the cell pellet in growth medium, aspirate and dispense into new (fibronectin coated, if adherence is desired) flasks.

Floating clusters may also be removed using a pipette and transferred to a flask containing fresh growth medium.



Comments

This line was derived from minced, trypsinized Swiss mouse embryos grown in serum free medium.

BALB/c SFME cells infrequently form colonies in soft agar and are not tumorigenic if injected into nude mice. Either serum or TGF-beta induce astrocyte differentiation accompanied by GFAP (glial fibrillary acidic protein) expression. Furthermore, the presence of serum causes cell growth arrest.

This process is reversible upon removal of the serum.

Swiss SFME cells do not grow in conventional media supplemented with fetal bovine serum or bovine calf serum.

When cultured in the serum free medium (see below), Swiss SFME cells reportedly can be propagated for extended periods without undergoing "crisis" or gross chromosomal aberration.



References

References and other information relating to this product are available online at 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

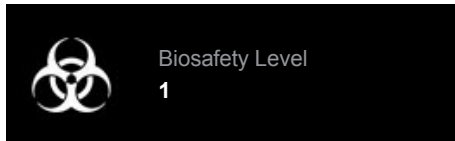
The viability of ATCC® products is warranted for 30 days from the date of shipment, and is valid only if the product is stored and cultured according to the information included on this product information sheet. ATCC lists the media formulation that has been found to be effective for this strain. While other, unspecified media may also produce satisfactory results, a change in media or the absence of an additive from the ATCC recommended media may affect recovery, growth and/or function of this strain. If an alternative medium formulation is used, the ATCC warranty for viability is no longer valid.



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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

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

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