If the cells are still attached, incubate the culture at 37°C in air atmosphere until they are ready to be subcultured.

2. If the cells are still attached, aseptically remove all but 5 to 10 mL of the shipping medium. The shipping medium can be saved for reuse. Incubate the cells at 37°C in a 5% CO₂ in air atmosphere until they are ready to be subcultured.

3. If the cells are not attached, aseptically remove the entire contents of the flask and centrifuge at 125 x g for 5 to 10 minutes. Remove shipping medium and save. Resuspend the pelleted cells in 10 mL of this medium and add to 25 cm² flask. Incubate at 37°C in a 5% CO₂ in air atmosphere until cells adhere.
Product Sheet
bEnd.3 [BEND3] (ATCC® CRL-2299™)

Please read this FIRST

Storage Temp.
liquid nitrogen
vapor
temp... cientific publication, it
should be cited in that manuscript in the following
manner: bEnd.3 [BEND3] (ATCC®
CRL-2299™)

Citation of Strain

If use of this culture results in a scientific publication, it
should be cited in that manuscript in the following
manner: bEnd.3 [BEND3] (ATCC® CRL-2299™)

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
Dulbecco's Modified Eagle's Medium, Catalog No. 30-
2002. To make the complete growth medium, add the
following components to the base medium: fetal bovine
serum to a final concentration of 10%.

Cryopreservation Medium

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

Subculturing Procedure

Remove and discard culture medium. Briefly rinse the cell layer with 0.25% (w/v) Trypsin-0.03% (w/v) EDTA
solution to remove all traces of serum which contains trypsin inhibitor. Add 1.0 to 2.0 mL of Trypsin-EDTA
solution to flask and observe cells under an inverted microscope until cell layer just begins to detach. Add 6.0
to 8.0 mL of complete growth medium and aspirate cells by gently pipetting. Add appropriate aliquots of the
cell suspension to new culture vessels. Corning® T-75 flasks (catalog #430641) are recommended for
sub culturing this product.

Subcultivation Ratio: A subcultivation ratio of 1:6 to 1:10 is recommended

Medium Renewal: Every 2 to 3 days

Cytokine and Lipopolysaccharide (LPS).

The expression of Peyer's Patch high endothelial receptor for lymphocytes, the mucosal vascular addressin
(MAdCAM-1) and E-selectin can be induced on bEnd.3 cells by cytokines and lipopolysaccharide (LPS).

MAdCAM-1 is expressed on the surface of unstimulated bEnd.3 cells at early passages but not at passages
greater than 30.

Intracellular adhesion molecule 1 (ICAM-1) is constitutively expressed on the cells, and expression is
increased by treatment with LPS, IL-1 and TNF alpha.

Vascular cell adhesion molecule 1 (VCAM-1) is constitutively expressed on the cells at early passages but
not at passages over 30.

P-selectin can be induced on bEnd.3 cells by Tumor Necrosis Factor alpha (TNF alpha) at both early and late
passages but expression is greater at passages over 30.

References

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.

ATCC Warranty

ATCC® products are warranted for 30 days from the date of shipment, and this warranty is valid only if the
product is stored and handled according to the information included on this product information sheet. If the
ATCC® product is a living cell or microorganism, ATCC lists the formula that has been found to be
effective for this product. 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 product. If an alternative medium formulation is used, the ATCC warranty for viability is no
longer valid.

Disclaimers

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

While ATCC uses reasonable efforts to include accurate and up-to-date information on this product sheet,
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patents are provided for informational purposes only. ATCC does not warrant that such information has been
confirmed to be accurate.

This product is sent with the condition that you are responsible for its safe storage, handling, and use. ATCC
is not liable for any damages or injuries arising from receipt and/or use of this product. While reasonable effort
Product Sheet

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