



# hTERT Immortalized RPTEC Growth Kit

ACS-4007™

## Description

hTERT Immortalized RPTEC Growth Kit can be added to DMEM: F12 (ATCC 30-2006) to produce a complete medium for hTERT Immortalized RPTEC cells. The kit consists of individually packaged supplements; each supplement must be added in appropriate volumes as per Product Sheet directions to 500 ml basal medium, DMEM: F12.

**Shipping information:** Frozen supplement kit for 500 mL base medium

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## Storage Conditions

**Product format:** Frozen

**Storage conditions:** -20°C or colder

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## 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.

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## Biosafety Information

ATCC determined that a biosafety level is not applicable to this material based on our risk assessment as guided by the current edition of *Biosafety in Microbiological and Biomedical Laboratories (BMBL)*, U.S. Department of Health and Human Services. It is your responsibility to complete your own risk assessment and understand any potential hazards associated with the material per your organization's policies and procedures and any other applicable regulations as enforced by your local or national

agencies.

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## Certificate of Analysis

For batch-specific test results, refer to the applicable certificate of analysis that can be found at [www.atcc.org](http://www.atcc.org).

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## Handling Procedures

### Unpacking and Storage Instructions

1. Check all containers for leakage or breakage.
2. Store the growth kit(s) at either -20°C in a freezer that is not self-defrosting or at -70°C for long term storage. If thawed upon arrival, the Growth Kit can be stored at 2°C to 8°C and added to the basal medium within 72 hours of receipt.

### Preparation of Complete Growth Media

1. Obtain one hTERT immortalized RPTEC Growth Kit from the freezer; make sure that the caps of all components are tight.
2. Thaw the components of the growth kit and mix by gentle vortex or inversion just prior to adding them to the basal medium. **Note: Only use 8.0 mL of Supplement B to obtain the optimal final concentration.**
3. Obtain one bottle of DMEM/F12 Medium (500 mL) from cold storage.
4. Decontaminate the external surfaces of all growth kit component vials and the basal medium bottle by spraying them with 70% ethanol.
5. Using aseptic technique and working in a laminar flow hood or biosafety cabinet, transfer the indicated volume of each growth kit component, as indicated in Table 1, to the bottle of basal medium using a separate sterile pipette for each transfer.

**Table 1.** hTERT immortalized RPTEC Growth Kit

Component	Volume	Final Concentration
RPTEC Supplement A	5.0 ml	1% RPTEC Supplement A
RPTEC Supplement B	8.0 ml	1.6% RPTEC Supplement B
G418 (not supplied)	Varies based on source	0.1 mg/ml

The final concentration for each growth kit component in the complete hTERT immortalized RPTEC growth medium is as follows:

25 ng/mL Hydrocortisone

3.5 µg/mL Ascorbic Acid

8.65 ng/mL Sodium selenite

5.0 µg/mL Transferrin

5.0 µg/mL insulin

5 pM Triiodo-L-thyronine

25 ng/mL Prostaglandin E1

10 ng/mL rhEGF

1.2 mg/mL Sodium Bicarbonate

**Required but not supplied:** G418 solution **MUST** be added to the above medium to a final concentration of 0.1 mg/mL G418 to maintain the selective pressure for immortalization.

Fetal Bovine Serum is not required for proliferation but may be added if it is desired to transfect the cells. The recommended volume of each **optional** component to be added to the complete growth media is summarized in Table 2.

**Table 2.** Addition of Serum (Optional)

Component	Volume	Final Concentration
Fetal Bovine Serum,	1 ml	0.2% FBS

6. Tightly cap the bottle of complete growth medium and swirl the contents gently to assure a homogeneous solution. Do not shake forcefully to avoid foaming. Label and date the bottle.
7. Complete growth media should be stored in the dark at 2°C to 8°C (do not freeze). When stored under these conditions, complete growth media is stable for four weeks.

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## Quality Control Specifications

**Bacterial and fungal testing:** Not detected

**Functional tests:** Rate of proliferation and morphology comparable to control. Cells cultured in complete medium exhibit dome structures in 7-14 days

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## Material Citation

If use of this material results in a scientific publication, please cite the material in the following manner: hTERT Immortalized RPTEC Growth Kit (ATCC ACS-4007)

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## References

References and other information relating to this material are available at [www.atcc.org](http://www.atcc.org).

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## Contact Information

ATCC

10801 University Boulevard

Manassas, VA 20110-2209

USA

US telephone: 800-638-6597

Worldwide telephone: +1-703-365-2700

Email: [tech@atcc.org](mailto:tech@atcc.org) or contact your local distributor