

# TeSR™-AOF

**cGMP, animal origin-free, stabilized, and feeder-free maintenance medium for human ES and iPS cells**

Catalog #100-0401

1 Kit



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## Product Description

TeSR™-AOF is an animal origin-free (AOF), stabilized, serum-free cell culture medium for the feeder-free maintenance and expansion of human embryonic stem (ES) and induced pluripotent stem (iPS) cells. It is based on the TeSR™ formulation (Ludwig et al., 2006a, 2006b), the most widely published feeder-free cell culture medium for human ES and iPS cells.

To enhance cell quality attributes, particularly during restricted feeds, critical medium components have been stabilized, including fibroblast growth factor 2 (FGF2; also known as basic FGF [bFGF]). As a result, TeSR™-AOF allows for compatibility with both daily and restricted feeding schedules while maintaining cell quality and equivalent performance.

TeSR™-AOF is compatible with a variety of culture matrices, including Corning® Matrigel® hESC-Qualified Matrix (Corning Catalog #354277), Vitronectin XF™ (Catalog #07180), and CellAdhere™ Laminin-521 (Catalog #77003).

No materials of animal or human origin are used in the manufacture of this medium or its components, to at least the secondary level of manufacturing. TeSR™-AOF is animal component-free to the secondary level. Each lot of TeSR™-AOF 20X Supplement that is used to prepare complete TeSR™-AOF medium is performance-tested in a culture assay using human pluripotent stem cells.

TeSR™-AOF is manufactured under relevant cGMPs, ensuring the highest quality and consistency for reproducible results. For additional quality information, refer to [www.stemcell.com/compliance](http://www.stemcell.com/compliance).

## Product Information

The following components are sold as a complete kit (Catalog #100-0401) and are not available for individual sale.

COMPONENT NAME	COMPONENT #	SIZE	STORAGE	SHELF LIFE
TeSR™-AOF Basal Medium	100-0402	475 mL	Store at 2 - 8°C.	Stable until expiry date on label.
TeSR™-AOF 20X Supplement	100-0403	25 mL	Store at -20°C.	Stable until expiry date on label.

## Preparation of Complete TeSR™-AOF Medium

Use sterile technique to prepare complete TeSR™-AOF medium (Basal Medium + 20X Supplement). The following example is for preparing 500 mL of complete medium. If preparing other volumes, adjust accordingly.

NOTE: Thaw supplement or complete medium at room temperature (15 - 25°C) or overnight at 2 - 8°C. **Do not thaw in a 37°C water bath.**

1. Thaw TeSR™-AOF 20X Supplement at room temperature (15 - 25°C) or overnight at 2 - 8°C. **Warm to room temperature.** Mix thoroughly.

NOTE: Once thawed, use supplement immediately or aliquot and store at -20°C for up to 3 months. Do not exceed the shelf life of the supplement. After thawing the aliquoted supplement, use immediately. Do not re-freeze.

2. Add 25 mL of TeSR™-AOF 20X Supplement to 475 mL of TeSR™-AOF Basal Medium. Mix thoroughly.

NOTE: If not used immediately, store complete TeSR™-AOF medium at 2 - 8°C for up to 2 weeks. Alternatively, aliquot and store at -20°C for up to 3 months. Do not exceed the shelf life of the individual components. After thawing the aliquoted complete medium, use immediately or store at 2 - 8°C for up to 2 weeks.

If prepared aseptically, complete TeSR™-AOF medium is ready for use. If desired, the medium can be filtered using a 0.2 - 0.22 µm low protein binding polyethersulfone (PES) filter unit (e.g. Fisher 09-741-04 [0.2 µm, 250 mL]; Fisher SCGP00525 [0.22 µm, 50 mL]).

## Directions for Use

For complete instructions on how to maintain human ES and iPS cells in TeSR™-AOF, refer to the Technical Manual: Maintenance of Human Pluripotent Stem Cells in TeSR™-AOF (Document #10000008160), available at [www.stemcell.com](http://www.stemcell.com) or contact us to request a copy.

### Feeding Schedule

TeSR™-AOF accommodates a more flexible feeding schedule without affecting culture quality. To determine a convenient schedule that suits your lab's routine, refer to the table below. Any combination of feeding intervals can be used throughout a passage when following these guidelines.

NOTE: Daily feeding is recommended when confluence is greater than 50% before passage.

FEEDING INTERVAL		
DAILY FEEDING	SKIP ONE DAY	SKIP TWO CONSECUTIVE DAYS
Standard feed volume (e.g. 2 mL per well of a 6-well plate)	Standard feed volume (e.g. 2 mL per well of a 6-well plate)	Double feed volume (e.g. 4 mL per well of a 6-well plate)

### Notes for TeSR™-E8™ Users

Cultures grown in TeSR™-AOF are very similar to cultures grown in TeSR™-E8™. An enhanced growth rate may be observed with TeSR™-AOF, resulting in higher confluence cultures sooner after passaging. Therefore, experienced TeSR™-E8™ users may note one or more of the following slight adjustments to the passaging parameters established for TeSR™-E8™ cultures, as outlined in the Technical Manual (Document #10000008160):

- Increased split ratio to maintain similar confluency
- Decreased passaging interval for more rapidly growing cultures

## Assessment of hPSCs

The following antibodies can be used to characterize hPSCs by flow cytometry or immunocytochemistry:

- Anti-Human SSEA-4 Antibody, Clone MC-813-70 (Catalog #60062)
- Anti-Human TRA-1-60 Antibody, Clone TRA-1-60R (Catalog #60064)
- Anti-Human OCT4 (OCT3) Antibody, Clone 3A2A20 (Catalog #60093)

For complete flow cytometry protocols and antibodies that can be used, refer to the Technical Manual: Maintenance of Human Pluripotent Stem Cells in TeSR™-AOF (Document #10000008160), available at [www.stemcell.com](http://www.stemcell.com) or contact us to request a copy.

## Related Products

For related products, including specialized cell culture and storage media, matrices, antibodies, cytokines, and small molecules, visit [www.stemcell.com/hPSCworkflow](http://www.stemcell.com/hPSCworkflow) or contact us at [techsupport@stemcell.com](mailto:techsupport@stemcell.com).

## References

Ludwig TE et al. (2006) Derivation of human embryonic stem cells in defined conditions. *Nat Biotechnol* 24(2): 185–7.

Ludwig TE et al. (2006) Feeder-independent culture of human embryonic stem cells. *Nat Methods* 3(8): 637–46.



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