

iPSC-Derived Human Astrocytes Kit (African-American, Male Line)

Applied StemCell has developed an efficient integration-free, small molecule-based method to differentiate high-quality astrocytes from human iPSCs. The differentiated astrocytes recapitulate the phenotype and functional parameters of primary and in vivo astrocytes.

We provide astrocytes differentiated from an integration-free, control human iPSC line (ASE-9211), reprogrammed from the fibroblasts of an African-American male donor. These high-purity cells express high levels of astrocyte biomarkers, GFAP and s100beta (Figure 2).

To harness the full potential of our astrocytes, we also provide optimized Astrocyte Basal Culture Media (ASE-9743MM) and Astrocyte Culture Media Supplement A (ASE-9743MM-A) that support the robust maintenance and functionality of the astrocytes in culture.

These iPSC-differentiated astrocytes can be used as control lines to compare phenotype and functionality of patient-derived, genome edited iPSC-derived astrocytes for drug screening applications. These astrocytes can also be used in neurotoxicity assays.

Parental Tissue Control human iPSC (ASE-9211); p15
 Age: Neonate
 Gender: Male
 Ethnicity: African-American
 Tissue Source: Dermal Fibroblasts
 Reprogramming Method: Episomal
 Culture Conditions: Feeder-free

Catalog #	Product	Size
ASE-9743	iPSC-derived Astrocytes; African-American, Male Line	$\geq 1 \times 10^6$ cells/ vial

Kit Components

Catalog #	Product	Size	Storage
ASE-9743-C	iPSC-derived Astrocytes; African-American, Male Line	$\geq 1 \times 10^6$ cells/ vial	Liq. N ₂
ASE-9743MM	Astrocyte Basal Culture Media	100 mL	-20°C
ASE-9743MM-A	Astrocyte Culture Media Supplement A (100x)	50 μ L	-20°C

Intended Use

For research or further manufacturing purposes. Not for injection or diagnostic procedures.

Quality Assurance

Each lot of iPSC-derived human cortical neurons has been tested for growth, viability and purity ($\geq 80\%$) following recovery from cryopreservation. In addition, each lot has been tested for expression of a cortical neuron marker and for the absence of mycoplasma and pathogens.

Storage Instructions and Stability

Store the components of the kit at the appropriate storage conditions as indicated in the media and materials table, immediately upon arrival. Shelf-life of the product is contingent upon proper storage conditions.

Precautions

PLEASE READ BEFORE HANDLING ANY FROZEN VIALS. Please wear appropriate Personal Protection Equipment (lab coat, thermal gloves, safety goggles and a face shield) when handling frozen vials. Please be aware that the following scenario can occur: Liquid nitrogen can leak into the vials when the vials are submerged in liquid nitrogen. Upon thawing, the liquid nitrogen returns to the gas phase, resulting in a dangerous build-up of pressure within the vial. This can result in the vial exploding and expelling not only the vial contents but also the vial cap and plastic fragments of the vial.

Directions for Use

1. Coating Cell Culture Vessels with Coating Matrix

- 1.1. Coat the plates with 80 $\mu\text{g}/\text{mL}$ Matrigel®.
Note: Please follow manufacturer's instructions in coating plates using Matrigel®.
- 1.2. Incubate at room temperature for at least 1 hour before use.

2. Preparation of Astrocyte Culture Media

- 2.1. Thaw the Astrocyte Culture Media at room temperature before thawing the cryopreserved astrocyte.
- 2.2. The Basal Culture Media should be aliquoted and stored at -20°C if it will not be used immediately.
Note: The media can be stored at 4°C for up to 2 weeks or at -20°C for up to 12 months.

3. Thawing and Culturing Cryopreserved Astrocytes

- 3.1. To thaw the cryopreserved Astrocytes, remove one vial from the storage unit.
- 3.2. Immerse the vial in the bath (up to 2/3 of the vial height) and thaw the cells rapidly until only a small piece of ice is still visible (approximately one minute).
Note: Do not shake the vial during thawing.
- 3.3. Bring the vial to the biological cabinet immediately and spray the outside of the vial thoroughly with 70% ethanol and wipe it with an autoclaved paper towel.
- 3.4. Remove the cells from the vial using a p1000 micropipette (or serological pipette) and transfer it slowly, drop-wise while swirling into a 15 mL conical tube containing 5 mL of pre-warmed Astrocyte Basal Culture Medium. Wash the vial with 1 mL medium from the 15 mL conical tube and transfer it back to the tube.
Note: Do not mix cells up and down and avoid generation of bubbles.

- 3.5. Centrifuge cells at 250 x g for 5 minutes at room temperature.
- 3.6. Aspirate the medium very carefully using a vacuum (or pipette if preferred), leaving only a drop of liquid in the tube.
Note: Take extra care not to remove or disturb the cell pellet during aspiration of medium.
- 3.7. Using a p1000 micropipette, add 5 mL of the pre-warmed astrocyte complete culture medium (5 mL Astrocyte Basal Culture Media + 50 μ L Astrocyte Culture Media Supplement A (100X)) into the tube and gently re-suspend cells by pipetting up and down 2-3 times.
- 3.8. Remove a 10 μ L aliquot of the cell suspension and mix it with 10 μ L of Trypan blue solution.
- 3.9. Count the cells.
- 3.10. Aspirate the coating matrix from the pre-warmed cell culture vessel.
- 3.11. Seed the Astrocytes at a density ranging from 100,000-150,000 live cells/cm² in Cortical astrocyte complete C\culture medium.
- 3.12. Distribute the cells evenly.
- 3.13. Place cell culture vessels in the incubator (37°C/ 5% CO₂/ humidity control) overnight.
- 3.14. Completely change the media on the second day using the Astrocyte Basal Culture Medium.

Characterization of the ASE-9743 Astrocytes

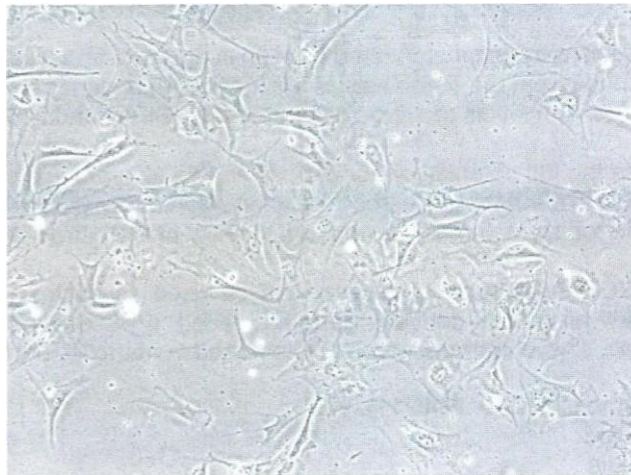


Fig 1. Bright Field Image of iPSC-derived Astrocytes from Applied StemCell's (ASC's) Control Line, ASE-9211.

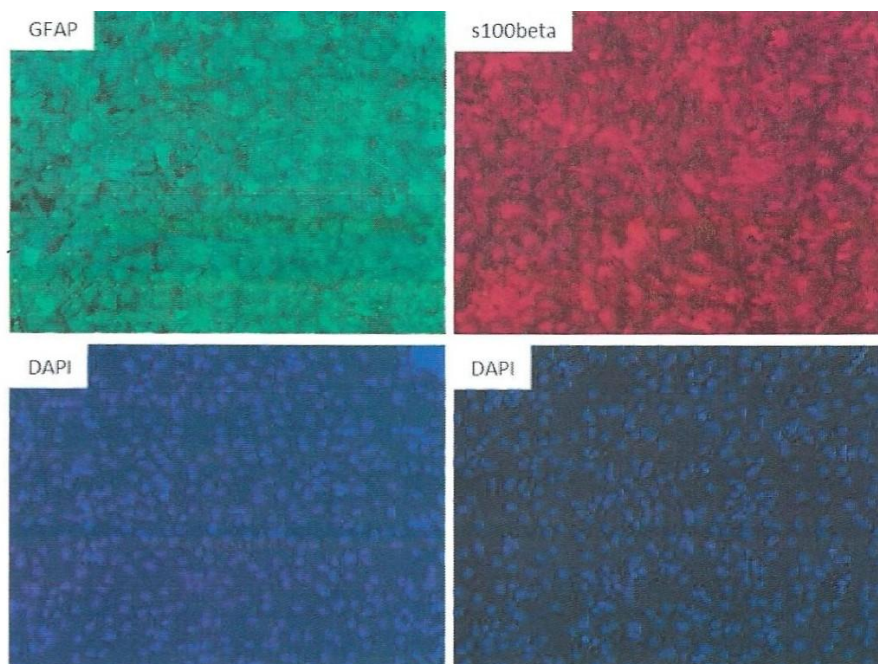


Fig 2. Antibody Staining of the Astrocytes differentiated from the NIST Control Line, ASE-9211. The astrocytes differentiated from ASC's iPSC line were stained with the astrocyte markers GFAP and s100beta. Left Three Images: Top Image: GFAP (Green), Middle Image: DAPI (Blue), Bottom Image: GFAP (Green) & DAPI (Blue); Right Three Images: Top Image: s100beta (Red), Middle Image: DAPI (Blue), Bottom Image: s100beta (Red) & DAPI (Blue).

Additional Reagents Required

The reagents below are recommended for use with cortical neurons. If you use reagents other than those recommended, we suggest that you do a batch test to validate quality of the cells and culture protocol.

- Matrigel®, Corning, Cat# 354230
- Antibodies:
 - GFAP R&D System, Cat# AF2594
 - S100B R&D Systems, Cat# AF1820
- Secondary antibodies: corresponding secondary antibodies were purchased from ThermoFisher

Technical Support

CONTACT US

For more information or assistance, contact Customer Service at:

- Email: info@appliedstemcell.com
- Direct line: (408) 773-8007

WEBSITE RESOURCES

Visit appliedstemcell.com for technical resources and information including:

- Safety Data Sheets (SDS)
- Certificates of Analysis (when available)
- FAQs
- Product literature
- Complete contact information

Applied StemCell, Inc.

521 Cottonwood Dr, Suite # 111, Milpitas, California, 95035, USA • Telephone: +1 866.497.4180 • appliedstemcell.com

© 2026 Applied StemCell, Inc. All rights reserved. TARGATT™ and ActiCells™ are trademarks of Applied StemCell, Inc. All other trademarks are the property of their respective owners. PN: IFU-ASE-9741 Rev. 1

Effective Date: 15 September 2026