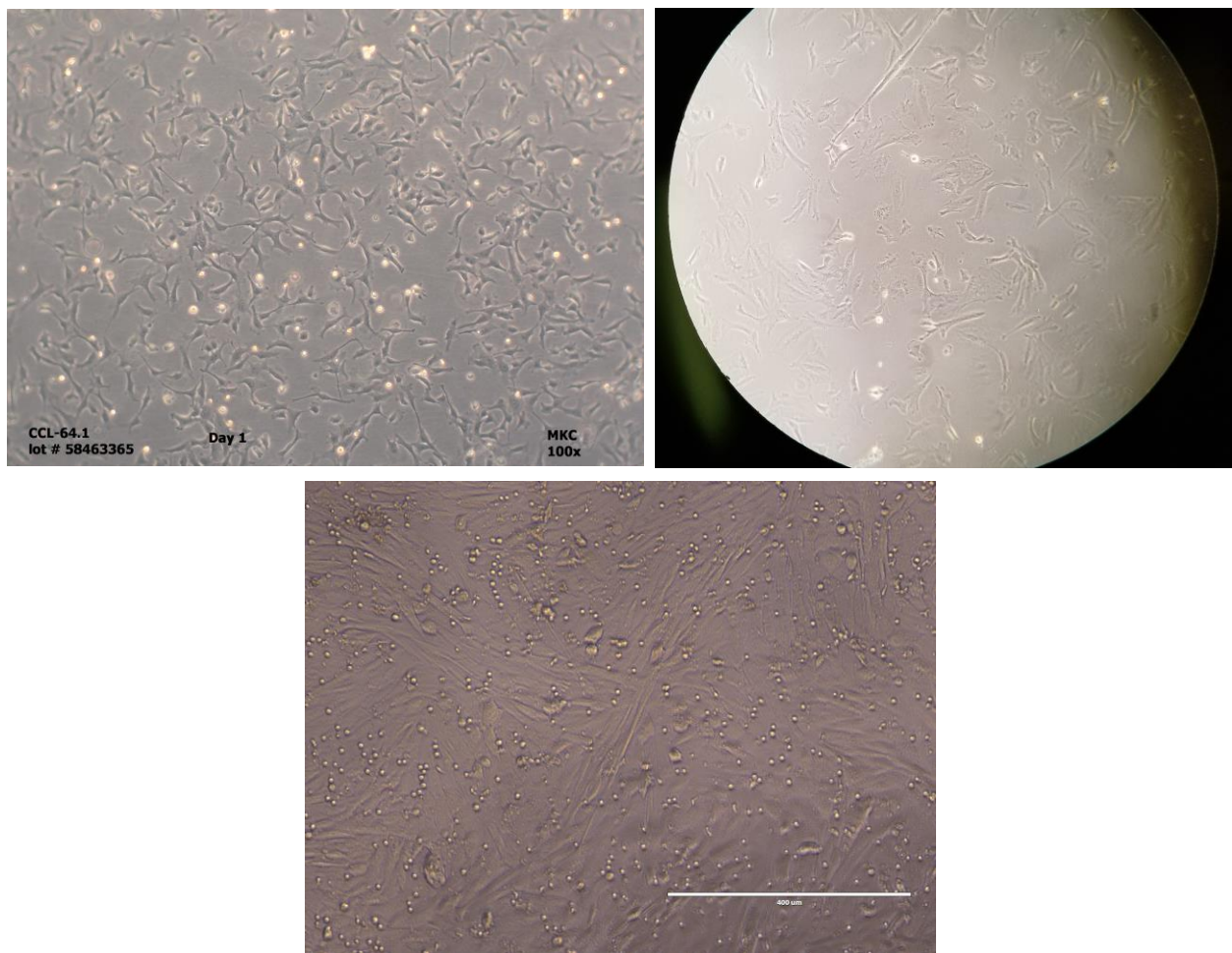

Name of Products	Human Neuron Cells
Catalogue Number	HNC001
Product Format	Frozen Vial
Cell Number	> 3x10 ⁵ cells/vial

GENERAL INFORMATION: HNCs are initiated by digestion of minced brain cortical tissue with collagenase. HNCs are separated from the mixture of cell populations and offered at passage 1 in a frozen vial. HNCs Growth Medium (contains 10% serum and growth supplements).



*Images: **Top Right:** Human neurons cultured alone. Data courtesy of Edu at Scripps. **Bottom:** Human neurons cultured with T-cells. Data courtesy of Edu at Scripps.*

PRODUCT USAGE Cells are offered for Research Use Only.

CHARACTERIZATION OF THE CELLS

1. Neurofilament protein Positive
2. Neuron specific enolase (NSE) Positive
3. Glial fibrillary acidic protein (GFAP) Negative
4. Myelin basis protein (MBP) Negative
5. HNCs are negative for HIV-1, HBV, HCV, and mycoplasma.

SHIPPING Frozen Vials on Dry Ice.

HANDLING OF ARRIVING CELLS When you receive the dry ice package with cells in frozen vials, transfer the frozen vials of cells into a -80C freezer for short period storage or a liquid nitrogen tank for long- term storage.

PROTOCOLS FOR THAWING:

1. Pre-coating of T25 flasks- Add 2ml of AlphaBiocoat into a T25 flask to cover the whole surface of the flask, Place the flasks in a 37C incubator for 30 minutes. After 30 minutes, remove your coated flask from the incubator, dispose the excessive coating solution by aspiration under a biosafety cabinet. Rinse the T25 flask twice with 1x PBS and the flask is ready to be used.
2. Thaw the frozen cell vial in a 37C water bath first, and then transfer the cells into the pre-coated T25 flask with 10ml of Neuro Growth media, cells usually become confluent with 10-12 days.

SUBCULTURING THE CELLS

Note: If you have any questions or need clarification regarding the protocol for culturing these cells, please reach out to Dr. Jensen Auguste at (978) 608-1766 with your questions before beginning.

3. Pre-coat 2 or 3 T25 flasks- Add 2ml of AlphaBiocoat into each T25 flask to cover the whole surface of the flask. Place the flasks in a 37C incubator for 30 minutes. After 30 minutes, remove your coated flask from the incubator, dispose the excessive coating solution by aspiration under a biosafety cabinet. Rinse the T25 flask twice with 1x PBS and the flask is ready to be used.
4. To passage the cells, rinse the cells in the T25 flask with 5ml 1x PBS (RT) twice; discard the 1x PBS, then add 2ml Trypsin/EDTA (RT) to the T25 flask.
5. Place the T25 flask with the cells in a 37C incubator for 1 min (most cells usually will detach from the surface within 1-2 mins; or monitor the cells under a microscope until most of cells become rounded up and are detach from the flask.
6. Add 5ml of Trypsin Neutralization Buffer or Neuro Growth media spin down the cells at 800g centrifugation for 5 mins. Once the five minute is completed, under a biosafety cabinet discard old medium.
7. Re-suspend the cell pellet with 10 or 15ml Neuro Growth media and transfer 5 ml each into 2 or 3 *pre-coated T25 flasks (for 1/2 to 1/3 subculture ratio).
8. Change medium every 2 or 3days and the cells usually become confluent within 10-12 days (when split at a 1/3 ratio). It is recommended that these cells have a minimum average population doubling capacity > 8 when cultured following our detailed protocol).

Note: Should any issues arise while using our cells, our team is here to help troubleshoot any issues. Our cells are backed by our one-time replacement or refund policy. Our recommended protocol including recommended products must be used to be eligible for replacement or refund. Cells that have been refrozen are no longer eligible for refund or replacement.

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