

Certificate of Analysis

Strain C57BL/6 Mouse Neural Stem Cells

With GFP

Cryopreservation Date: 2009-04-02

Passage Number: 2

Catalog No. MUBNF-01101

Lot Number: 090402A01

Viability

Cells are assayed for viability post-thaw using vital staining assay with trypan blue.

Specification: Cells should exhibit $\geq 70\%$ viability.

Sterility

Bacterial and Fungal Contamination: Samples are inoculated and cultured in blood agar plate, thioglycolate broth, tryptocase soy broth and sabouraud dextrose agar.

Specification: No growth must be observed.

Mycoplasma: Samples are tested for mycoplasma contamination using a PCR-based assay and direct culture.

Specification: Results must be negative.

Endotoxin: Samples are tested for endotoxin contamination with LAL test.

Specification: Results must show a concentration of $\leq 25\text{EU/ml}$.

Purity

Cells are assayed for purity using immunohistochemistry analysis of cell surface antigen expression after cryopreservation. Cells are immunostained with Nestin.

Specification: Cells must show $\geq 75\%$ positivity for expression of cell surface antigens Nestin. Cells must show $\leq 10\%$ positivity for expression of cell surface antigens GFAP, Galc and beta-III tubulin.

Proliferation Ability

The cells are characterized by their ability to proliferate in culture with neurosphere morphology for ≥ 5 passages, and $\leq 5\%$ cells exhibit spontaneous differentiation in each passage.

GFP Expression

Expression of constitutive GFP is assayed by visual inspection of GFP fluorescence signal.

Specification: The results must indicate $\geq 80\%$ of cells are visually inspected for GFP fluorescence signal during extensive subcultivation.

Differentiation Ability

The cells are assayed after cryopreservation for their ability to differentiate into Neuron, Astrocyte and Oligodendrocyte, about 50% cells are stained with GFAP, 10% cells are stained with beta-III tubulin, and 2% cells are stained with Galc that detects each cell type.

Results:

All specifications have been met.



Jane Chen
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