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14-906ACL: GFP/Ba/F3 Stable Cell Line

Application : Functional Assay,FACS

Description

GFP-Ba/F3 Stable Cell Line is a stably transfected Ba/F3 cell line which expresses enhanced green fluorescent protein (eGFP).

Sequence data: Amino acid sequence of eGFP

MVSKGEELFTGVVPILVELDGDVNGHKFSVSGEGEGDATYGKLTLKFIC
TTGKLPVPWPTLVTTLTYGVQCFSRYPDHMKQHDFFKSAMPEGYVQE
RTIFFKDDGNYKTRAEVKFEGDTLVNRIELKGIDFKEDGNILGHKLEYNY
NSHNVYIMADKQKNGIKVNFKIRHNIEDGSVQLADHYQQNTPIGDGP
VLLPDNHYLSTQSALSKDPNEKRDHMVLLEFVTAAGITLGMDELYK

Product Info

Amount :	1 vial
Content :	Each vial contains 2 ~ 3 x 10^6 cells in 1 ml of 90% FBS + 10% DMSO
Storage condition :	Immediately upon receipt, store in liquid nitrogen.

Application Note

Application:.

- Screen for GFP through Flow Cytometry.
- Screen for GFP through Fluorescence Microscopy.

Culture conditions:

Cells should be grown at 37°C with 5% CO_2 using RPMI medium supplemented with 10% heat-inactivated FBS, 1 mM sodium pyruvate, 10 mM HEPES and 1% Pen/Strep plus 5 ng/ml mIL-3 (Note: mIL-3 is essential for Ba/F3 cell maintenance) plus 3 µg/ml of Puromycin.

It is recommended to quickly thaw the frozen cells upon receipt or from liquid nitrogen in a 37^{O}C water-bath, transfer to a tube containing 10 ml of growth medium without Puromycin, spin down cells, resuspend cells in pre-warmed growth medium without Puromycin, transfer resuspended cells to T25 flask and culture in 37^{O}C-CO_2 incubator.

Monitor the cell viability by counting cells daily for 1-3 days until cells completely recover viability as cells are doubling daily. Once cells are over 90% confluent, harvest cells by centrifugation and passage cells. At first, switch to growth medium containing puromycin. Cells should be split before they reach complete confluence.

To passage the cells, transfer cells to a tube, spin down cells, resuspend cells and seed appropriate aliquots of cells suspension into new culture vessels. Subcultivation ration = 1:10 to 1:20 weekly. To achieve satisfactory results, cells should not be passaged over 16 times.

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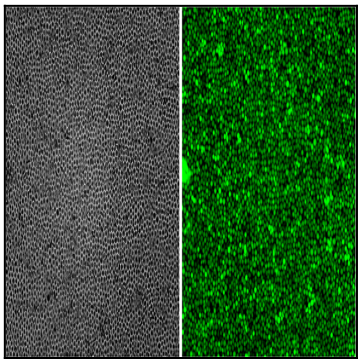

Fig-1: Analysis of the GFP-Ba/F3 stable cell line through fluorescence microscopy. Bright-field image (Left); Fluorescence image (Right).

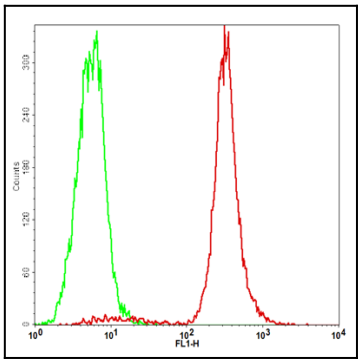

Fig-2: Detection of GFP in the GFP-Ba/F3 stable cell line through flow cytometry . Parental Ba/F3 cells (Green); GFP-Ba/F3 cells (Red).