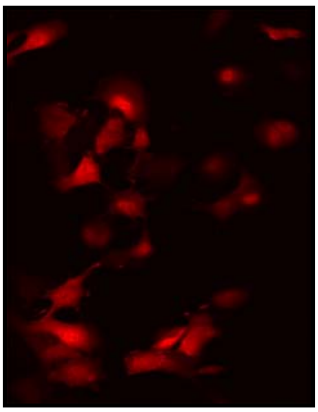
REF: P20321

VAMPIRO™ CELL LINES

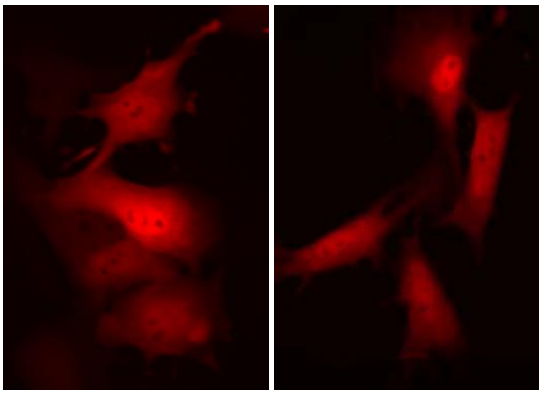
RED FLUORESCENT B16-F10 CELLS



Product Name:	VAMPIRO™ – B16-F10 Cell line
Catalog Number:	P20321
Cell Line:	B16-F10 Mouse melanoma
Fluorescent Protein:	turboFP602
Resistance:	Puromycin
Format:	>3x10^6 cells in Cryopreserved vials
Storage:	Liquid Nitrogen

This cell line has been produced with the technology developed within FP7 PASCA EU project, and is 100% certified truly monoclonal.

A novel red fluorescent B16-F10 cell line has been developed through stable transfection with TurboFP602 protein. This cell line expresses red fluorescent protein as a free cytoplasmatic protein.



Turbo FP602-B16-F10 Cell line is stably-transfected and it is ready to use in cell-based assay applications. This stably transfected cell line provides consistent levels of expression, which helps to simplify the interpretation of the results. This cell line is intended to be used as an "in vitro" model for research studies.

About B16-F10

B16F$_{10}$ cell line is a high metastatic variant of the murine B16 mouse melanoma, which was originated in the syngenic C57BL/6 (H-2b) mouse strain. This cell line is a mixture of adherent spindle-shaped and epithelial-like cells. B16F10 cell line is a well-established model for metastasis and it is applicable for the study of experimental cancer therapies.

Melanoma tumors are known to express different tumor-associated antigens, which usually induce weak immune responses of short duration. Expression of both tumor-associated antigens p53 and TRP2 by melanoma cells raises the possibility of simultaneously targeting more than one antigen in a therapeutic vaccine. Melanomas express.

V.A.T. Nº: ESB95481909
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About turboFP602 protein

TurboFP602 protein is a red shifted variant of the red fluorescent protein TurboRFP from sea anemone *Entacmaea quadricolor* [Merzlyak et al., 2007].

TurboFP602 possesses true-red fluorescence (with excitation/emission maxima at 574/602 nm, respectively), optimal for detection via most popular filter sets, and is easily distinguished from background signals. TurboFP602 exhibits fast maturation and high pH stability.

Quality Control

All cells are performance assayed and test negative for mycoplasma, bacteria, yeast and fungi. Cell viability, morphology and proliferative capacity are measured after recovery from cryopreservation. Innoprot guarantees stable expression for many generations and provides support for cell culture and visualization.

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CELL CULTURE INSTRUCTIONS

A. Complete Growth medium

·RPMI 1640
·10% FBS
·5 μg/ml Puromycin

B. Set up culture after receiving

1. Decontaminate the external surfaces of medium and medium supplements with 70% ethanol.

2. Prepare coated flask (T-75 flask is recommended). Add 9 ml of RPMI 1640 and then add 1ml of FBS (without selection antibiotic). Leave the flask in incubator minimum one hour at 37ºC incubator.

3. Place the vial in a 37ºC water bath, hold and rotate the vial gently until the contents are completely thawed. Remove the vial from the water bath immediately, wipe it dry, and rinse the vial with 70% ethanol. Remove the cap, being careful not to touch the interior threads with fingers.

4. Dispense the contents of the vial using 1 ml eppendorf pipette and gently resuspend the contents of the vial in T75 flask containing pre-warmed complete growth media.

5. Place the flask in the incubator.

6. For best results, do not disturb the culture for 16 hours after the culture has been initiated. Change the growth medium (including the selection antibiotic) the next day to remove the DMSO and unattached cells, then every other day thereafter.

C. Maintenance of Culture:

1. Change the medium to fresh supplemented medium the next morning after establishing a culture from cryopreserved cells. For subsequent subcultures, change medium 48 hours after establishing the subculture.

2. Once the culture reaches 50% confluence, change medium every day until the culture is approximately 0% confluent.

3. Subculture the cells when they are over 90% confluent.

4. Incubate cells with 1 ml of trypsin/EDTA solution (in the case of T-75 flask) until 80% of cells are rounded up (monitored with microscope). Add 1ml of trypsin neutralization solution to the digestion immediately and gently rock the culture vessel.

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