**PRODUCT DESCRIPTION**


**INTENDED USE**

StemXVivo™ EMT Inducing Media Supplement is designed for the induction of EMT. It has been shown to induce EMT in the following human cell lines: MCF-7 human breast cancer cells, MCF-10A human breast epithelial cells, HT-29 human colon adenocarcinoma cells, A549 human lung carcinoma cells, and A431 human epithelial carcinoma cells.

**STABILITY & STORAGE**

Upon receipt, StemXVivo™ EMT Inducing Media Supplement should be stored at ≤ -20 °C in a manual defrost freezer. The EMT Inducing Media Supplement should be thawed at 2-8 °C before use. Thawed EMT Inducing Media Supplement can be stored at 2-8 °C for up to 2 weeks or aliquoted and stored at ≤ -20 °C for up to 3 months.

**LIMITATIONS**

- FOR LABORATORY RESEARCH USE ONLY. NOT FOR USE IN DIAGNOSTIC PROCEDURES.
- The safety and efficacy of this product in diagnostic or other clinical uses has not been established.
- This reagent should not be used beyond the expiration date indicated on the label.
- Results may vary with cells cultured by different methods.

**OTHER SUPPLIES REQUIRED**

- Epithelial cells of interest
- Cell culture medium
- Dissociation solution (*e.g.*, TrypLE™ Express; Invitrogen®) or equivalent
- 0.4% Trypan Blue solution
- Tissue culture flasks
- 15 mL centrifuge tubes
- Serological pipettes
- Pipettes and pipette tips
- 37°C, 5% CO₂ humidified incubator
- Centrifuge
- Hemocytometer
- Inverted Microscope
- Water bath
EMT INDUCTION PROCEDURE

Note: EMT induction is carried out in the culture media you are currently using to culture your cells of interest.

1. Warm culture media to 37 °C.
2. Gently detach the cells of interest from the culture dish using a dissociation solution (e.g. TrypLE™ Express, or equivalent). Resuspend the cells in warmed culture media.
3. Centrifuge the cell suspension at approximately 400 x g for 5 minutes. Aspirate the liquid.
4. Gently resuspend the cell pellet in warmed culture media and count viable cells using Trypan blue.
5. On tissue culture treated plates or flasks, plate cells at 0.9-1.0 x 10⁴ cells per cm² (e.g. 0.5 x 10⁶ cells in a 10 cm plate) in standard culture media (6 mL/10 cm plate) containing 1X StemXVivo™ EMT Inducing Media Supplement.
6. Incubate at 37 °C with 5% CO₂.
7. Three days after plating, remove the media from the plates and replace with fresh cell culture media containing 1X StemXVivo™ EMT Inducing Media Supplement.
8. Five days after plating, the cells are ready for analysis.

TYPICAL DATA

MCF-7 human breast cancer cells were cultured either without (Panels A-C) or with (Panels D-F) StemXVivo™ EMT Inducing Media Supplement for 5 days. On day 5 of induction, bright field images were taken to look for the mesenchymal morphology of cells (Panels A and D). Cells were then stained for the epithelial phenotype with goat anti-human E-Cadherin (Panels B and E; R&D Systems, Catalog # AF648) and the mesenchymal phenotype with sheep anti-human Fibronectin (Panels C and F; R&D Systems, Catalog # AF1918).