

## PRODUCT DESCRIPTION

The StemXVivo Serum-Free Tumorsphere Media is a semi-solid media formulated and optimized for tumorsphere formation. This product does not contain antibiotics.

## INTENDED USE

StemXVivo Serum-Free Tumorsphere Media is designed for the tumorsphere formation assay. It has been tested to support sphere formation of the following human tumor cell lines: MCF-7, HT-29, HeLa, MDA-MB-231, A172, LNCaP, and PC-3.

## STABILITY & STORAGE

Upon receipt, StemXVivo Serum-Free Tumorsphere Media should be stored at  $\leq -20\text{ }^{\circ}\text{C}$  in a manual defrost freezer. The media can be thawed at 2-8  $^{\circ}\text{C}$  or at room temperature. Thawed media can be aliquoted and stored at  $\leq -20\text{ }^{\circ}\text{C}$  in a manual defrost freezer for up to 3 months or used within two weeks when stored in the dark at 2-8  $^{\circ}\text{C}$ . Avoid repeated freeze-thaw cycles. Do not use beyond the expiration date.

## PRECAUTION

This product contains human transferrin. This transferrin was tested at the donor level using an FDA licensed method and found to be non-reactive for anti-HIV-1/2 and Hepatitis B surface antigen. As no testing can offer complete assurance of freedom from infectious agents, this product should be handled as if capable of transmitting infection.

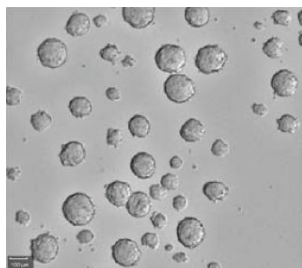
## LIMITATIONS

- 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.

## PROCEDURE

1. Warm the desired volume of StemXVivo Serum-Free Tumorsphere Media to 37  $^{\circ}\text{C}$ .
2. For optimal tumorsphere growth, adding the following reagents to the media is recommended:
  - 2 U/mL Heparin (Tocris, Catalog # 2812)
  - 0.5  $\mu\text{g}/\text{mL}$  Hydrocortisone (Tocris, Catalog # 4093)
3. Gently detach the cells of interest from the culture dish using a dissociation solution. Resuspend the cells in warmed culture media.
4. Centrifuge the cell suspension at approximately 400 x g for 5 minutes. Aspirate the liquid.
5. Gently resuspend the cell pellet in warmed StemXVivo Serum-Free Tumorsphere Media, and transfer the cell suspension to ultralow adhesion plates. Incubate cultures in a 5%  $\text{CO}_2$  incubator at 37  $^{\circ}\text{C}$ . Sphere formation should occur within 3-10 days.

**Note:** StemXVivo Serum-Free Tumorsphere Media contains methylcellulose. Pipette slowly to ensure the media does not stick to the inside of the pipette. Cell plating density depends on the experimental design of each individual investigator. Alternatively,  $3 \times 10^4$  cells in 2 mL per well in 6-well ultralow adhesion plates is recommended for MCF-7 cells.



**Figure 1: Morphology of MCF-7 tumorspheres formed in StemXVivo Serum-Free Tumorsphere Media.** Imaged using a CloneSelect™ Imager from Molecular Devices (New Milton) Limited.