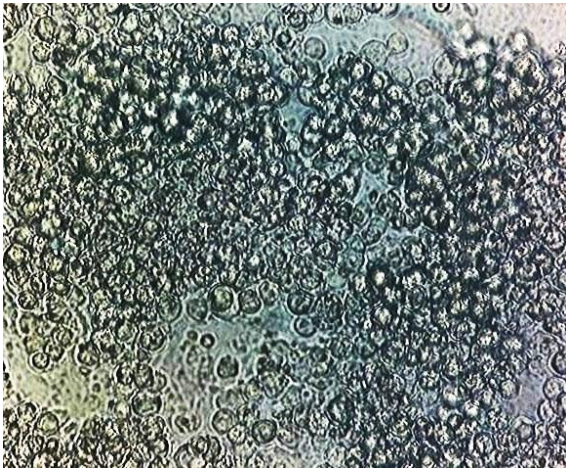


Human Colorectal Adenocarcinoma Enriched CancerStemCell™

Growing "solid tumor" without matrix gel or growth factors

BioMedicure Number:	CSC-221	Price:	\$899.99
Disease:	Colorectal adenocarcinoma, colon		
Designations:	SC-221		
Depositors:	BioMedicure		
Biosafety level:	1		
Shipped:	Frozen		
Organism:	<i>Homo sapiens</i> (human)		
Growth Properties:	Adherent		
Genome:	Unstable		
Tumorigenic <i>in vitro</i>:	Yes, without matrix gel or growth factors		



Tumorigenic <i>in vivo</i>:	Yes
Heterogeneous:	Yes
Randomly Mutating:	Yes
Resist to apoptosis inducers:	Yes

Why Is This Product Special?

1. Growing "solid tumor" *in vitro* without the aid of matrix gel.
2. Growing in any complete medium without special supplement such as growth factors.
3. Heterogeneous with antigens comparable to *in vivo* tumors.
4. Research results will be more comparable with that from *in vivo*.
5. Cost-saving right away.

Use Restrictions

These cells are distributed for research purposes only. BioMedicure recommends that individuals contemplating commercial use of any enriched CancerStemCell™ products first contact BioMedicure for potential licensing agreement. Buyers are the second party end users of this product. Any

Information Sheet for BioMedicure's Products

distribution of this product to a third party is strictly prohibited.

Biosafety Level: 1

Appropriate safety procedures should always be used with this material. Laboratory safety is discussed in the following publications: *Biosafety in Microbiological and Biomedical Laboratories*, 4th ed. HHS Publication No. (CDC) 93-8395. U.S. Department of Health and Human Services, Centers for Disease Control and Prevention. Washington DC: U.S. Government Printing Office: 1999.

The entire text is available online at www.cdc.gov/od/ohs/biosfty/bmbl4/bmbl4toc.htm

Handling Procedure for Frozen Cells

To insure the highest level of viability, thaw the vial and initiate the culture as soon as possible upon receipt. If upon arrival, continued storage of the frozen culture is necessary, it should be stored in liquid nitrogen vapor phase and not at -70°C. Storage at -70°C will result in loss of viability.

SAFETY PRECAUTION: BioMedicure highly recommends that protective gloves and clothing always be used and a full face mask always be worn when handling frozen vials. *It is important to note that some vials leak when submersed in liquid nitrogen and will slowly fill with liquid nitrogen. Upon thawing, the conversion of the liquid nitrogen back to its gas phase may result in the vial exploding or blowing off its cap with dangerous force creating flying debris.*

1. Thaw the vial by a gentle agitation in a 37°C water bath. To reduce the possibility of contamination, keep the O-ring and cap out of the water. Thawing should be rapid (within 2 minutes).
2. Remove the vial from the water bath as soon as the contents are thawed, and decontaminate by dipping in or spraying with 70% ethanol. All of the

operations from this point on should be carried out under strict aseptic conditions.

3. Transfer the vial contents to a centrifuge tube containing 9.0 ml complete growth medium and spin at 125 x g for 8 to 10 minutes.
4. Re-suspend cell pellet with a complete growth medium (see the specific lot information for the recommended dilution ratio). *It is important to avoid excessive alkalinity of the medium during recovery of the cells. It is suggested that, prior to the addition of the vial contents, the culture vessel containing the growth medium be placed into the incubator for at least 15 minutes to allow the medium to reach its normal pH (7.0 to 7.6)*
5. Incubate the culture at 37°C in a suitable incubator with 95% humidity. A 5% or 0% CO₂ in air atmosphere is used according to the medium you are using.
6. Since this product is enriched with CancerStemCell™, cells will grow in most, if not all, complete media.

Subculturing Procedure

Volumes used in this procedure are for 75 cm² flask; proportionally reduce or increase amount of dissociation medium for culture vessels of other sizes.

1. Remove and discard culture medium.
2. Briefly rinse the cell layer with 0.25% (w/v) Trypsin-EDTA solution to remove all traces of serum which contains trypsin inhibitors.
3. Add 1.0 ml of Trypsin-EDTA solution to flask and observe cells under an inverted microscope until cell layer is dispersed (usually within 5 to 10 minutes).

Note: To avoid clumping, do not agitate the cells by hitting or shaking the flask while waiting for the cells to detach. Cells that are difficult to detach may be placed at 37°C to facilitate the dispersal process.

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4. Add 6.0 to 8.0 ml of complete growth medium and aspirate cells by gentle pipetting.
5. Add appropriate aliquots of the cell suspension to new culture vessels. Subcultivation Ratio is 1:3 to 1:5 depending on cell numbers per flask used for subcultures.
6. Incubate cultures at 37°C with 95% humidity.

Note: For more information on enzymatic dissociation and subculturing of cells consult Chapter 10 in *Culture of Animal Cells, a manual of Basic Technique* by R. Ian Freshney, 3rd edition, published by Alan R. Liss, N.Y., 1994.

Medium Change

Two to three times weekly.

Complete Growth Medium

The base medium for this product can be any complete medium, including but not limited to various Dulbecco's Modified Eagle's Medium, RPMI-1640 Medium, Eagle's Minimum Essential Medium or Leibovitz's L-15 Medium since this human colorectal adenocarcinoma enriched CancerStemCell™ has the capability to adjust to the new environment, even extreme conditions with fetal bovine serum at a final concentration ranging from 2.0-10.0%.

Cyroprotectant Medium

Complete culture medium described above supplemented with 5% (v/v) DMSO.

Additional Information

Additional product and technical information can be obtained from the catalog references and the BioMedicure web site at www.biomedicure.com, or by email to csc@biomedicure.com.

BioMedicure Warranty

The viability of BioMedicure products is warranted for 30 days from the date of shipment. If you feel there is a problem with this product, contact Technical Services by phone at 858-586-1888 or by email to csc@biomedicure.com.

Disclaimers

This product is intended for laboratory research purposes only. It is not intended for use in any regulatory area or subjects including humans.

While BioMedicure uses reasonable efforts to include accurate and up to date information on this product sheet, BioMedicure makes no warranties or representations as to its accuracy. Citations from scientific literature and patents are provided for informational purposes only. BioMedicure does not warrant that such information has been confirmed to be accurate.

This product is sent with the condition that you are responsible for its safe storage, handling and use. BioMedicure is not liable for any damages or injuries arising for receipt and/or use of this product. While reasonable effort is made to insure authenticity and reliability of strains on deposit, BioMedicure is not liable for damages arising from the misidentification or misrepresentation of cultures.

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