

Enhance Serum-Free Insect Cell Culture with GIBCO® Media

- Maximize cell growth and recombinant protein yields
- Protein-free formulations optimized for specific cell types
- Convenient pre-adapted cells save time
- Custom products and packaging available
- Easy to scale up to large volume cultures

Product	Optimized For	Applications
Drosophila-SFM	Suspension <i>Drosophila melanogaster</i> cells (D.Mel-2, Schneider S2 cells)	Growth and maintenance medium for adherent or suspension culture.
Express Five® SFM	Suspension BTI-TN-5B1-4 insect cells	Growth and maintenance of cells used for the baculovirus expression vector system (BEVS) for adherent or suspension culture. Large-scale production of recombinant protein expressed by BEVS.
Sf-900 II SFM†	Suspension Sf9, Sf21 (<i>Spodoptera frugiperda</i>), TN368 cells (<i>Trichoplusia ni</i>)	Growth and maintenance of cells used for BEVS for adherent or suspension culture. Large-scale production of recombinant protein expressed by BEVS.
D.Mel-2 Cells, Adapted to Drosophila SFM		
High Five™ Cells, Adapted to Express Five® SFM		
Sf9 Cells, Adapted to Sf-900 II SFM		
Sf21 Cells, Adapted to Sf-900 II SFM		

■ Protein-Free Media ■ Pre-Adapted Cells

† Drug Master File available

Note: Cell lines from different sources, and different clones of the same cell line, may have highly specific nutritional requirements and may therefore prefer one medium over another. More than one medium formulation (if available) should be evaluated to determine the best option.

Sf-900 II SFM

Protein-Free Formulation

GIBCO® Sf-900 II SFM is a protein-free medium optimized to support and maximize Sf9 and Sf21 cell growth in suspension applications, as well as recombinant gene expression using both baculovirus and stable insect expression systems. This medium provides excellent results in the production of a variety of recombinant proteins (figures 1 and 2) and will support Sf9 cell growth to densities of 8 to 10 × 10⁶ cells/ml.

Sf9 and Sf21 Cells

For your convenience, we offer Sf9 and Sf21 cells that are pre-adapted to suspension growth in Sf-900 II SFM and commonly used for expression of recombinant proteins using the Baculovirus Expression System (BEVS). Sf9 is a clonal isolate derived from the parental *Spodoptera frugiperda* cell line IPLB-Sf21-AE. Sf21 is an isolate from *Spodoptera frugiperda* ovarian cells.

Sf9 Cell Growth After 15 Passages

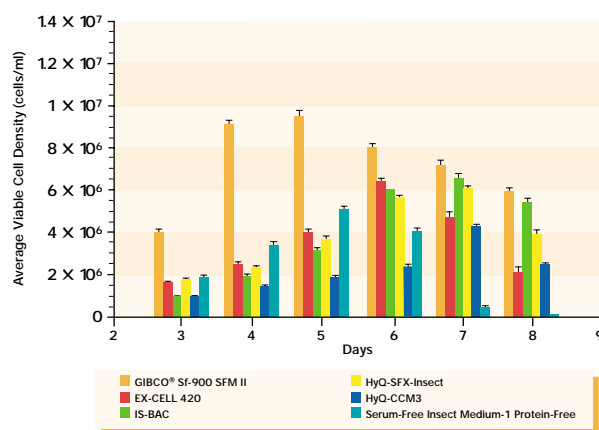


Figure 1. Sf9 cells expanded in TNM-FH medium plus 10% FBS, and then adapted into each of the test media. After 15 passages in the test media, cell growth curves were determined by seeding 30 ml cultures in 125 ml shake flasks (8 replicates for each medium) at 400,000 cells/ml, and counting viable cell number on each day post-seed.

SFM

Serum-Free Media

GIBCO® Serum-Free Media do not require supplementation with serum, but may contain discrete proteins or bulk protein fractions.

PFM

Protein-Free Media

GIBCO® Protein-Free Media contain no proteins, but may contain plant or yeast hydrolysates. Many are animal-origin-free.

CDM

Chemically-Defined Media

GIBCO® Chemically-Defined Media contain no proteins, hydrolysates, or components of unknown composition. These media are animal-origin-free and all components have a known chemical structure.

- Completely defined system eliminates variability
- Consistent performance improves reproducibility
- Decrease possibility of contamination by adventitious agents
- Save time with simplified purification and downstream processing

Total β -gal Produced per Cell

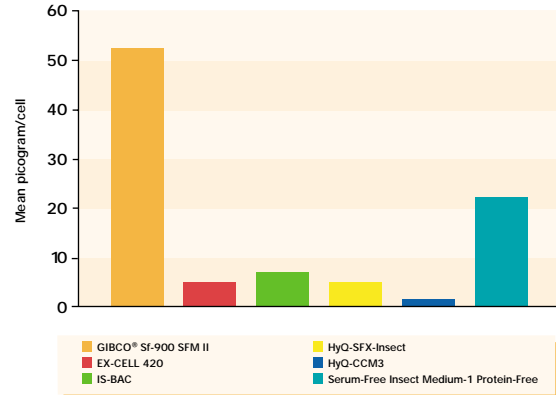


Figure 2. Sf9 cells expanded in TNM-FH medium plus 10% FBS, and then adapted into each of the test media. After 13 passages in the test media, 30 ml cultures in 125 ml shake flasks (3 replicates for each media) were seeded at 600,000 cell/ml, incubated for 24 hours and then infected at an MOI= 3 with AcMNPV expressing a recombinant β -galactosidase. Samples were taken daily and counted for viable cells and assayed for β -gal activity (combined media and cell samples). The maximum per ml β -gal production (day 4 or 5 post-infection) was divided by the maximum viable cell density observed post-infection, to obtain a per cell β -gal productivity value.

Express Five® SFM

Protein-Free Formulation

This unique medium eliminates problems associated with the culturing of the BTI-TN-5B1-4 (High Five™) cell line. It is optimized for serum-free growth in monolayer and suspension culture, boosting insect cell growth to densities exceeding 5×10^6 cells/ml and population doubling times of 16 to 30 hours. It demonstrates expression of recombinant proteins that is equal or superior to that of other serum-free formulations.

High Five™ Cells

For your convenience, we offer High Five™ cells, pre-adapted to Express Five® SFM. Excellent for expressing recombinant proteins, High Five™ cells are derived from *Trichoplusia ni* embryo cells.

Drosophila-SFM

Protein-Free Formulation

Optimized for the growth of *Drosophila melanogaster* (D.Mel) cells, protein-free Drosophila-SFM outperforms other commercially available media for culturing D.Mel-2 cells, reaching peak cell densities of $> 10^7$ cells/ml in suspension culture. Cells grown in other media adapt quickly and easily to this medium. The absence of protein due to FBS supplementation promotes easier downstream purification of functional protein.

D.Mel cells can be transfected for stable or transient expression of recombinant proteins. To prevent lipid interference, transfection of D.Mel cells is best performed in Drosophila-SFM.

D.Mel-2 Cells

For your convenience, we offer D.Mel-2 cells pre-adapted to Drosophila-SFM. These cells are isolated from late-stage *Drosophila melanogaster* embryos, and are used for transient or stable expression of recombinant proteins (figure 3).

Growth of D.Mel-2 Cells

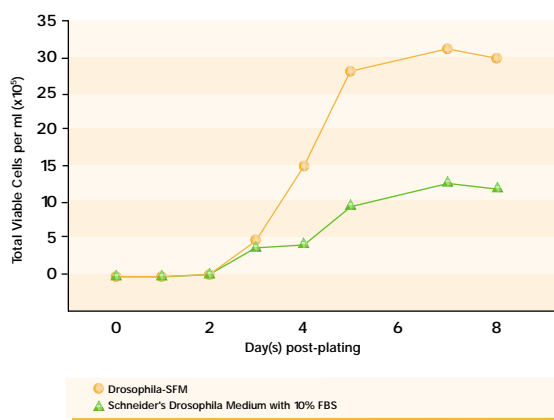


Figure 3. Early passage *Drosophila melanogaster* cells were seeded in 125 ml shaker flasks at a density of 3×10^5 viable cells per ml in 35 ml media. The flasks were incubated at 27°C and rotated at 135 rpm. Samples were taken daily and evaluated for viable cell density with the use of trypan blue.

The cells grown in Drosophila-SFM reached significantly higher densities than cells in Schneider's Drosophila Medium supplemented with 10% FBS. Maximal growth rate was found to be during days 3 and 4.

Cellfectin® Reagent for High-Efficiency Insect Transfection

Invitrogen Cellfectin® Reagent is formulated and tested for optimal transfection of insect cells. Transfection with Cellfectin® Reagent leads to consistent and efficient transfection of D.Mel-2, S2, Sf9, Sf21, and High Five™ cells when using the Baculovirus Expression System (BEVS) or stable insect expression plasmids (figure 4).

BaculoDirect™ Baculovirus Expression System

The Invitrogen BaculoDirect™ Expression System is the fastest and easiest method for generating recombinant baculovirus. The BaculoDirect™ System utilizes the strong polyhedrin promoter for high-level expression and a counter-selection cassette for easy isolation of parent-free viral stock without the need for plaque purification. From the initial transfection to the isolation of the baculovirus stock, the BaculoDirect™ System requires only eight hours of hands-on time—less than half the time required with traditional baculovirus systems.

Sf9 Cells Transfected with Cellfectin® Reagent

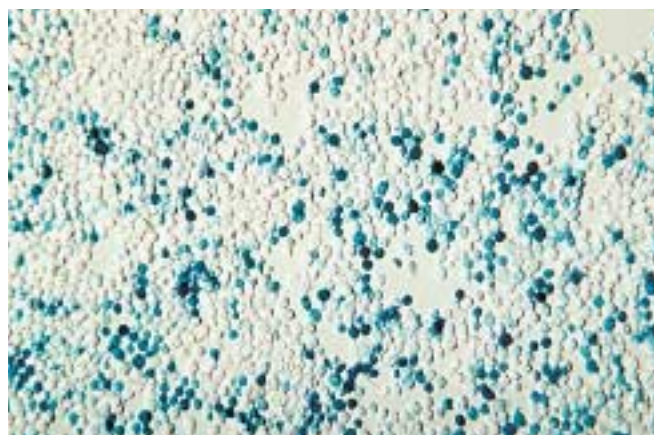


Figure 4. Cellfectin® Reagent for transfection of insect cells. 9×10^5 Sf9 cells were plated/well in a 6-well plate on Sf-900 II SFM. One hour later, the cells were transfected with bacmid DNA coding for the β -glucuronidase gene, using Cellfectin® Reagent. The cells were stained with X-glucuronide 72 hours post-transfection to detect β -gluc expression.

Custom Production and Packaging

When you need a unique formulation or special packaging, our Custom Product Services team can modify GIBCO® catalog media formulations and packaging to meet your particular requirements.

Media are available in different formats for easy scale-up to meet the needs of various levels of product development: R&D, process development, pilot plant, and manufacturing.

We can produce volumes as small as a few liters to > 30,000 liters, or > 100,000 liters in dry format. In addition, we offer large media bag packaging options up to 500 liters.

The Custom Product Services team can also assess feasibility and provide options for formulation design, testing, and packaging for your proprietary formulations.

For information call 1-800-955-6288, Ext. 46966.

References

Weiss, S.A., Godwin, G.P., Gorfien, S.F., and Whitford, W.G. (1993) *Insect Cell Culture Engineering* (Goosen, M.F.A., Daugulis, A.J., and Faulkner, P., eds.) Marcel Dekker, Inc., NY.

Weiss, S. A., Godwin, G. P., Gorfien, S.F., and Whitford, W. G. (1995) *Methods in Molecular Biology* (Richardson, C.D., ed.) **39**, Humana Press, Totowa, NJ.

Sf-900 II SFM

Godwin, G. and Whitford, W. (1993) *FOCUS*® **13**, 44.

Express Five® SFM

Godwin, G., Danner, D., and Gorfien, S. (1995) *FOCUS*® **17**, 58.

Ordering Information

Description	Catalog No.	Size
Sf-900 II SFM (1X), liquid Contains L-glutamine.	10902-096	500 ml
	10902-088	1,000 ml
	10902-104	6 × 1,000 ml (case)
	10902-070	10 L
Sf-900 II SFM (1X), liquid (w/o Met and Cystine) Contains L-glutamine.	21012-026	500 ml
Sf-900 Medium (1.3X), liquid Contains L-glutamine.	10967-032	100 ml
Sf9 Cells	11496-015	1.5 ml
Sf21 Cells	11497-013	1.5 ml
Express Five® SFM (1X), liquid	10486-025	1,000 ml
High Five™ Cells	B855-02	3 × 10 ⁶ cells/ml
Drosophila-SFM (1X), liquid	10797-017	500 ml
	10797-025	1,000 ml
D.Mel-2 Cells	10831-014	1.5 ml
Cellfectin® Reagent	10362-010	1 ml
BaculoDirect™ Baculovirus Expression Kit	12562-013	5 reactions
BaculoDirect™ Baculovirus Transfection Kit	12562-039	5 reactions



www.invitrogen.com

Corporate Headquarters: Invitrogen Corporation • 1600 Faraday Avenue • Carlsbad, California 92008 U.S.A.
Tel: 1 760 603 7200 • Tel (Toll Free): 1 800 955 6288 • Toll Free Fax: 1 800 331 2286 • E-mail: tech_service@invitrogen.com
European Headquarters: Invitrogen Ltd • 3 Fountain Drive • Inchinnan Business Park • Paisley PA4 9RF, UK
Tel: + 44(0) 141 814 6100 • Fax: + 44(0) 141 814 6260 • E-mail: eurotech@invitrogen.com



These products are for research use, and where appropriate, as raw material components in further cell culture manufacturing applications. They are not intended for human or animal diagnostic, therapeutic, or other clinical uses, unless otherwise stated. © 2003 Invitrogen Corporation PAS03-063MS Part No. 332-032443G