NEW

FastGro™

Fully Chemically Defined FBS Replacement

Thomas Scientific

Unleash the Possibilities

Animal-free, chemically defined FBS replacement to promote cell growth and consistency

MP Biomedicals

Cell Biology Research Solutions

MP Biomedicals offers a full range of products to meet all of your needs in cell biology, spanning many areas of research from cell structure, organization, function and metabolism, to life cycle. Our products include cell culture media, media supplements, reagents, kits and solutions for cell separation, detachment, signalling, proliferation, growth, apoptosis, staining, and many more areas of current topics of cell biologists. Our time-tested and high quality products can provide the right tools for all your research needs.

NEW!

FastGro™, Fully Chemically Defined FBS Replacement for Cell Culture

Fetal bovine serum (FBS) is widely used as a serum-supplement for in vitro cell culture media, providing an undefined mixture of nutrients for healthy cell culture growth, such as proteins, attachment factors, growth factors, amino acids, trace elements, vitamins, lipids, and hormones. However, due to its undefined nature and the variation of animals, FBS can lead to unexpected and undesired stimulations of cells, not to mention the biorisk of animal protein or pathogen contamination, such as bovine spongiform encephalopathy (BSE).

To avoid these concerns, MP Bio is pleased to launch FastGro™, a fully chemically defined FBS replacement for cell culture use. This unique product allows culturing a wide range of cells in vitro without the use of serum or any animal or human derived compound. All components in FastGro™ are highly purified and identified chemical compounds, ensuring:

- **Chemically defined nature without lot-to-lot variations**
- **No animal or human derived materials or compounds**
- **No interference with hormones or growth factors**
- **Elimination of the risk of contaminants – viruses, mycoplasma, prions, etc.**
- **Wide range of cell culture practices**
- **Storage in the refrigerator, and no need for thawing before use**

Description	Pack Size	Mfr. No.	Thomas No.
FastGro™, Fully Chemically Defined FBS Replacement for Cell Culture	100 mL	092640049	CHM00G916
	500 mL	092640054	CHM00G915

The Vero cell strain used in the following experiments comes from a flask of VERO IM 153 on the 153rd passage acquired from ATCC. This strain was divided into two sub-lines, one propagated in Fetal Bovine Serum (FBS)-supplemented medium and the other sub-line proliferated in FastGro™-supplemented serum-free medium.

Growth evaluation in FBS-supplemented versus FastGro™-supplemented medium

This study was performed with Vero cells adapted over long-term culture in serum-free, FastGro™-supplemented medium. Growth dynamics were compared to those of the sub-line cultured in FBS-supplemented medium. The results are from a 6-day culture period in T25 flasks. Cell numbers were determined at the end of the 144-hour culture period (Figure 1).

Medium	Seeding Cell Density (per cm^2)	Final Cell Density (per cm^2)	Cell Multiplication Index
Williams Medium E + 10% FBS	20,000	184,000	9.20
Williams Medium E + 10% FastGro™	20,000	197,000	9.85

Figure 1

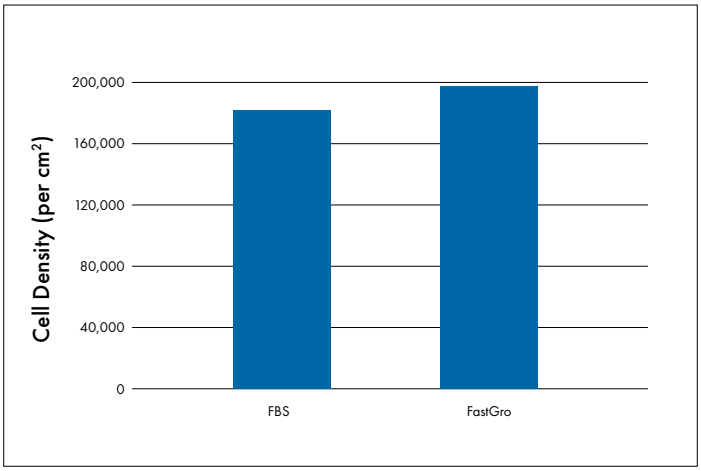
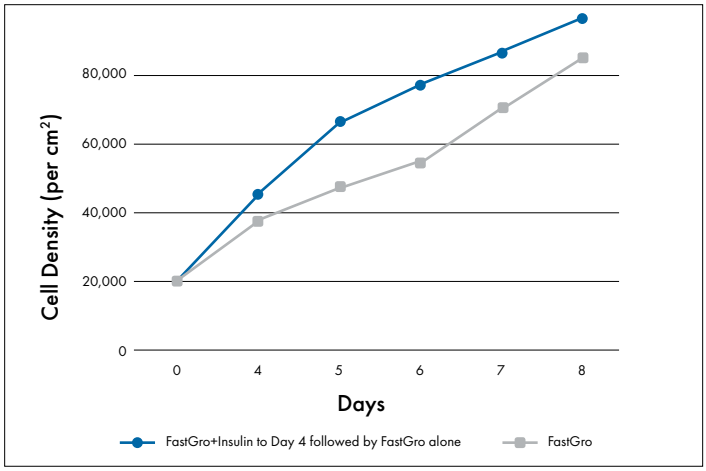


Figure 2



Growth of Vero cells in FastGro™- supplemented medium on microcarriers

In this study, a Vero strain was adapted over 3 passages to serum-free growth in FastGro™-supplemented Williams Medium E grown on microcarriers (200 mg cytodex/100 mL corresponding to 1200 cm^2 growth area) in small-scale laboratory bioreactors. To test for potential positive effects of insulin, the hormone was added to the culture medium (1.25 mg/L) of one group from day 0 through day 4, then switched back to Williams E + FastGro™ alone.

As can be observed, in both conditions the Vero cells showed a constant growth over the entire experimental period (Figure 2). The addition of insulin during the initial growth phase (4 days) caused a growth enhancement during the logarithmic growth phase, albeit the two experimental groups showed only a slight difference (< 10%) in the final cell numbers (< 10%). These results imply that a short mitogenic stimulus by insulin during the initial growth phase may be sufficient for generating a sustained growth benefit over the whole batch culture period.

The following application note pertains to a variety of cell types used for serum-free primary cell culture methods.

Absence of Serum Attachment Factors

Preliminary step - Coating of the culture surface

In defined culture conditions, the treatment of the culture surface with an adequate coating strategy is crucial. Crude preparations of extracellular matrices (ECM), such as mouse sarcoma extracts (e.g. matrigel) or extracted collagen preparations are commonly used to coat culture surfaces. However, the undefined nature, as well as the presence of animal-derived compounds, renders their use problematic for many applications. If animal-derived material will not pose a problem, an overnight treatment of the plastic cell culture surfaces with a small amount of FBS may be considered as a 'quick-fix'. This method is cost-effective and efficient, but represents a step back from the fully-defined culture environment concept.

Today, recombinant and defined coating kits are available that mimic the attachment properties of ECM proteins through the use of biosynthetic signaling peptides derived from fibronectin, laminin, collagen, E-cadherin, vitronectin, etc.

Absence of Enzyme Inhibitors from Serum

Dissociation enzyme

There are essentially two methods for initiating primary cultures: outgrowth from a primary explant or via enzymatic disaggregation. In the latter method, the starting tissue is digested using proteolytic enzyme cocktails, such as dispase, collagenase and trypsin. Care must be taken to neutralize/deactivate any remaining proteolytic activity before seeding the cells, particularly when trypsin is employed. The use of standard trypsin preparations can become problematic in the absence of serum, which contains trypsin inhibitors. In serum-deprived conditions, tryptic activity must be inactivated after the cell dissociation process. This can be achieved by using an efficient trypsin inhibitor, such as soybean trypsin inhibitor.

As an alternative to trypsin, the use of Accutase™ is highly recommended because it does not require deactivation. This recombinant, non-mammalian enzyme has been efficiently used for multiple types of primary cultures, including primary smooth muscle cells, primary human endothelial cells, and primary chick neuronal cells.

Absence of Binding by Serum Proteins

Use of Antibiotics

Antibiotics, like many other compounds, bind to the plasma proteins of serum, in particular to the albumin fraction. Thus, the same concentration of antibiotics will exhibit a much higher biological activity in serum- and albumin-free conditions, resulting in deleterious implications to cell growth. Streptomycin is particularly deleterious, as it is known to interfere at the level of protein synthesis in mammalian cells. In instances where 'antibiotic-free culture' is deemed unworkable, the use of gentamicin is suggested at a concentration of 50 mg/L.

Cell-type-specific Recommendations

Primary cell cultures have various requirements, depending on the tissue of origin. For the purposes of this application note, we will not discuss the primary cell culture procedures that differ vastly from one cell type to the other. Generally, we recommend applying the 'conventional' techniques for the isolation of primary cells of the desired type, replacing the serum source with the addition of FastGro. This will satisfy the nutritional requirements of most, if not all cell types. For the majority of mammalian cell cultures, nutritional requirements vary only slightly quantity-wise. More demanding cell types, such as hepatocytes, require higher nutrient concentrations.

Growth factor and hormone requirements often differ significantly between cell types. The table below lists the cell culture media preparations that we recommend when FastGro is used as a replacement for animal serum. The growth factor and hormone concentrations indicated are recommended for optimal cellular development and proliferation with respect to each of the indicated cell types.

Recommended cell culture media set-up using FastGro™ for select major primary cell culture types

PRIMARY CELL CULTURE TYPE	FASTGRO CONCENTRATION	RECOMMENDED BASAL MEDIUM	RECOMMENDED GROWTH FACTOR, FINAL CONCENTRATIONS & REQUIREMENT	RECOMMENDED HORMONES, FINAL CONCENTRATIONS & REQUIREMENT
Primary Kidney Cultures	FastGro™ 10%	DMEM high glucose / F-12	EGF (human, recombinant), 50 ng/mL, optimal/ beneficial	Insulin (recombinant human), 0.5 µg/mL, essential
				Hydrocortisone, 0.1 µg/mL, essential
				Triiodo-L- thyronine, 10 pg/mL, essential
				Epinephrine, 0.5 µg/mL, essential
Primary Hepatocytes	FastGro™ 10-15%	Williams' Medium E	EGF (human, recombinant), 50 ng/mL, optimal/ beneficial	Insulin (recombinant human), 5 µg/mL, essential
				Hydrocortisone, 0.5 µg/mL, essential
Primary Keratinocytes*	FastGro™ 10%	DMEM/F-12 (1:3 ratio)	EGF (human, recombinant), 0.125 ng/mL, optimal/ beneficial	Bovine Pituitary Extract (BPE), 4 µL/mL, essential
				Hydrocortisone, 5 µg/mL, essential
				Epinephrine, 0.5 µg/mL, essential
Primary Cardiomyocytes	FastGro™ 10%	Claycomb Medium	EGF (human, recombinant), 5 ng /mL, optimal/ beneficial	T3 (triodo-L-thyronine), 1 ng/mL (1.5 nM), essential
			bFGF (human, recombinant), 5 ng /mL, optimal/ beneficial	Insulin (recombinant human), 5 µg/mL, essential
Neuronal Cells	FastGro™ 10%	DMEM high glucose	EGF (human, recombinant), 50 ng/mL, optimal/ beneficial	Insulin (recombinant human), 0.5 µg/mL, essential

Recommendations for optimal cell culture performance with use of FastGro:

- As a basal medium, select DMEM/F12, Williams' medium E or Ham's F12.
- Avoid the use of antibiotics, as FastGro lacks albumins, and all added antibiotics will remain free and active, increasing risk of cytotoxicity. If antibiotics must be used, choose gentamicin over pen-strep.
- We advise utilizing a gentle dissociation reagent for cell detachment, such as Accutase, while avoiding the use of trypsin. Since Accutase is an enzyme derived from tropical shellfish, it works optimally at 25°C, but decays quickly at higher temperatures.
- When refreshing the medium, we advise retaining approximately 25% old (conditioned) medium and adding 75% fresh prepared medium. Valuable factors produced and excreted by the cultured cells can be collected, filtered, and frozen from old media for future use.

NEW

All Synthetic FBS Replacement

FastGro™

Fully Chemically Defined FBS Replacement for Cell Culture

- Chemically defined nature without lot-to-lot variations
- No animal or human derived materials or compounds
- No interference with hormones or growth factors
- Elimination of the risk of contaminants – viruses, mycoplasma, prions, etc.
- Wide range of cell culture practices
- No thawing necessary – Store in the refrigerator
- Eliminates biorisk and increases cell culture consistency

HaCaT cells in medium enriched with 10% FastGro

HaCaT cells in medium enriched with 10% FBS

Outrgrow your wildest cell growth
No bull inside, all you need is FastGro

MP Biomedicals™

One Call. One Source. A World of Life Science Products.

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- Cell Biology
- Culture Growth Media
- FastPrep® Sample Prep
- Immunology
- Molecular Biology
- Adsorbents
- Biochemicals
- Fine Chemicals
- Labware
- Dosimetry
- Research Diets
- SafTest™ Food Quality
- Diagnostics
- Drugs of Abuse
- Infectious Disease
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