FastGro[™]

Fully Chemically Defined FBS Replacement

Cat. No. 092640049, 100 mL; Cat. No. 092640054, 500 mL

Fetal bovine serum (FBS) is widely used as a serumsupplement 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:

FastGro Synthetic, imal Free, Chemically

- 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



APPLICATION NOTE

FastGro with Vero Cells

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 cm²)	Final Cell Density (per cm²)	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

Growth of Vero cells in FastGrosupplemented 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 cm² 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.







APPLICATION NOTE

Common Recommendations for Primary Cell Cultures

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

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.

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 on the back page 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	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	40.45%	Williams' Medium E	EGF (human, recombinant), 50 ng/mL, optimal/ beneficial	Insulin (recombinant human), 5 μg/mL, essential
	10-15%			Hydrocortisone, 0.5 μg/mL, essential
Primary Keratinocytes	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	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	10%	DMEM high glucose	EGF (human, recombinant), 50 ng/mL, optimal/ beneficial	Insulin (recombinant human), 0.5 μg/mL, essential



MP BIOMEDICALS

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