# FetalClone III

### **HYCLONE SERA**

HyClone<sup>™</sup> FetalClone<sup>™</sup> III is an economical alternative to FBS delivering both cost-effectiveness and high performance. Commonly used in cell culture and bioprocessing applications, FetalClone III is a supplement to enrich cell culture performance. With high lot-to-lot consistency and low endotoxin and hemoglobin levels, FetalClone products have demonstrated performance with a variety of cell lines, including hybridomas, CHO, BHK-21, NSO, MRC-5, and Vero cells (Fig 1).

#### Key features of FetalClone III include

- The most widely applicable of our FetalClone family of products
- Designed to include fibroblasts in its applications
- Improved cell growth performance relative to FBS control of US origin
- Offering the same basic formulation found in FetalClone II, with additional growth factors and supplements

# Specifications

FetalClone III is filtered through three sequential 100 nm (0.1  $\mu$ m) pore size-rated filters. Before dispensing, each lot of serum is pooled to ensure uniformity and consistency between bottles (true pool technology).

#### **Cell lines**

FetalClone III engineered serum has demonstrated performance with a variety of cell lines (Table 1). Figures 2 show the results of growth studies comparing FetalClone III with an FBS control on AIF, CHO-K1, BHK-21, NSO, MRC-5, and Vero cell lines.

# Product handling

### Storage and handling

Sera should be stored at -10°C or lower. Once thawed, sera should be stored at 2°C to 8°C. To maintain quality, a conservative storage recommendation for sera at this temperature is no longer than six weeks. If the serum needs to be stored longer than six weeks after opening, it is recommended to aliquot the serum into convenient volumes and refreeze.



**Fig 1.** FetalClone engineered serum products show equivalent performance to FBS at a reduced cost and without limitation in supply.

#### Thawing

Remove serum from -10°C (or lower) storage and place in a refrigerator overnight at 2°C to 8°C. Transfer the serum to a 37°C water bath, agitate periodically to mix the solutes concentrated at the bottom of the container. Do not hold the serum at 37°C any longer than necessary after thawing. Thawing serum in a bath above 40°C without mixing can denature the concentrated proteins in the bottom of the container and precipitates might form in the bottle. Thawing serum at higher temperatures is not recommended.

Alternatively, bottles can be placed directly from -10°C (or lower) storage into a 37°C water bath. Bottles should be agitated to enhance mixing and thawing. Turbidity and flocculent material might be present after thawing or after prolonged storage.



Table 1. Cell lines successfully cultured with FetalClone III engineered serum

Cell line	Description	Cell line	Description	Cell line	Description
MRC-5	Human lung fibroblast	HEL	Human lung fibroblast	PC-12	Murine adrenal gland
Mv 1Lu	Mink lung	Jurkat Human thymoma			pheochromocytoma
MDCK	Canine kidney	T-lymphocyte		COS	African green monkey kidney
CRFK	Feline kidney cortex	K562	Human bone marrow		fibroblast
VERO	African green monkey kidney	AtT-20	Murine pituitary tumor	Sp2/0-Ag14	Murine myeloma
WI-38	Human lung fibroblast	HEK-293	Human kidney	Sp2/0-Ag14	Murine hybridoma
ACHN	Human kidney renal cell carcinoma	EL-4 6.1	Murine thymoma	FOX-NY	Murine myeloma
786-O	Renal cell carcinoma	CHO-K1	Chinese hamster ovary	FOX-NY	Murine hybridoma
SAOS-2	Human bone osteosarcoma	JEG3	Human choriocarcinoma	ВНК	Baby hamster kidney fibroblast
U2-OS	Human primary osteogenic sarcoma	L929	Murine fibrosarcoma	NIH/3T3	Murine embryonic fibroblast
Hep 3B2	Human liver epithelial carcinoma	PLB-985	Human hystiocytic lymphoma	U-937	Human histiocytic lymphoma
P3X63 Ag8.653	Murine hybridoma	NS-1	Murine myeloma	P3	Murine myeloma
P3X63 Ag8.653	Murine myeloma	NS-1	Murine hybridoma	P3	Murine hybridoma
MDBK	Bovine kidney	Dg44/CHO-K1	Chinese hamster ovary	HeLa	Human epithelial adenocarcinoma
U9-IIIB	Human histiocytic lymphoma	C3H/10T1/2	Murine embryonic fibroblast	Hep-2	Human epithelial adenocarcinoma
1.6 1.4 1.2 O to 0.8 0.6 0.4 0.2		1.4 1.2 1.0 0.8 0.6 0.4 0.2 0.2		1.2 1.0 0.8 0.4 0.4 0.2	
MR	C-5 cell growth study	Ver	o cell growth study	1	BHK-21 cell growth study
1.8		1.2		1.4	
1.6		1.0		1.2	
1.2		0.8		1.0	
8.0 8 G		9.0 Pation — — — — — — — — — — — — — — — — — — —		8.0 Gatio	
℃ <sup>0.8</sup>		0.4			
0.4		0.2		0.4	

AIF cell growth study

Fig 2. Cell growth comparison studies, FBS control vs FetalClone III.

CHO-K1 cell growth study

Experience indicates that regardless of the method used to thaw serum, it is critical that it is mixed during the thawing process to prevent the formation of gradients and subsequent precipitation. Because of differences in thawing rates of different components, serum will form a gradient if it is not mixed as it thaws. If serum is allowed to remain in such a gradient state, precipitation is likely to occur. Handle bottles that have been stored in freezer carefully. Avoid large temperature shifts and protect the serum from exposure to light. Wear protective clothes and equipment. Storage requirements are listed on the product label.

### General culture recommendations

Supplementation of classical media such as Dulbecco's Modified Eagle's Medium (DMEM) is recommended at a range between 5% and 10% FetalClone III to support culture of a wide variety of cell lines and applications.

NSO cell growth study

# Quality control testing

FetalClone III is assayed for gamma globulin, alkaline phosphatase, lactate dehydrogenase, glutamic pyruvic transaminase (SGPT), glutamic oxaloacetic transaminase (SGOT), pH, total protein, albumin, blood urea nitrogen, creatinine, total bilirubin, sodium, potassium, calcium, chloride, inorganic phosphorous, osmolality, iron, total iron binding capacity (TIBC), percent saturation, glucose, and IgG. Assays are subject to change without notice.

Quality control test specifications are listed in Table 2.

Table 2. Test specifications

Endotoxin (Limulus amebocyte lysate gel clot assay)	≤ 10 EU/mL
Hemoglobin (spectrophotometric)	≤ 20 mg/dL
Sterility testing (current USP)	
Bacteria and fungi	No growth
Virus Testing (9 CFR 113.53)	
Fluorescent Antibody	
Bluetongue	Not detected
Bovine adenovirus	Not detected
Bovine parvovirus	Not detected
Bovine respiratory syncytial virus	Not detected
Bovine viral diarrhea virus	Not detected
Rabies	Not detected
Reovirus	Not detected
Cytopathogenic agents (e.g., IBR)	Not detected
Cytopathogenic agents (e.g., IBR)	Not detected
Mycoplasma	
Large volume, direct culture	Not detected
Hoechst DNA stain	Not detected
Certificate of suitability	Included

### Related products

### FetalClone I and FetalClone II

HyClone FetalClone I and FetalClone II complete the FetalClone product family. Each FetalClone product is optimized for particular cell lines and production platforms. Consult the respective data file for information on differences in formulation, cell lines supported, and application specificities.

#### Classical media

HyClone classical media are manufactured using ISO 9001and ISO 13485-certified processes. All raw material components have passed strict quality control testing to ensure the appropriate level of quality. The classical media are hydrated using process water and have undergone 0.1 µm sterile filtration.

Classical media that have been system tested with FetalClone III to confirm product efficacy and homogeneity are:

- Dulbecco's Modified Eagle's Medium (DMEM)
- Minimal essential medium (MEM)
- · Eagle's minimal essential medium (EMEM)
- RPMI 1640 medium

### HyClone phosphare buffered saline (PBS)

Our PBS products are manufactured according to cGMP guidelines using ISO 9001-certified processes. The products have full traceability and documented origin of all formula ingredients.

### HyClone trypsin protease

Our trypsin protease is derived from porcine pancreas and is gamma irradiated prior to hydration and filling. The product is formulated without calcium and magnesium.

### Ordering information

Product	Size	Product code
FetalClone III	100 mL	SH30109.02
	500 mL	SH30109.03
FetalClone III Heat Inactivation	100 mL	SH30109.02HI
	500 mL	SH30109.03HI
FetalClone III Irradiated	500 mL	SH30109.03IR
Related products		
FetalClone I	100 mL	SH30080.02
	500 mL	SH30080.03
FetalClone I Heat Inactivation	100 mL	SH30080.02HI
	500 mL	SH30080.03HI
FetalClone I Irradiated	500 mL	SH30080.03IR
FetalClone II	100 mL	SH30066.02
	500 mL	SH30066.03
FetalClone II Heat Inactivation	100 mL	SH30066.02HI
	500 mL	SH30066.03HI
FetalClone II Irradiated	500 mL	SH30066.03IR

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