Serum-free Cell Culture

SERUM-FREE CELL CULTURE

The use of animal sera as a supplement for sustaining cell cultures continues to be widespread primarly because of unquestioned adherence to cited methods, perceived economy and convenience, and observable growth. However, such factors do not exactly equate to proper cell growth and function. Sera is comprised of more than 200 known components and an indeterminant number of unknown complexes. Thus, it is a complex composition which varies depending on the age, gender, nutritional state, and overall health of the donor animal. Additionally, sera is a common source of detrimental factors such as mycoplasma, viruses, prions, bacterial mitogens, hormones, extraneous protein, growth factors and proteases. The presence of any factor can directly affect the cell's growth rate, characteristics, adherence and genetic stability.

Serum-free cell culture offers significant advantages over more commonly employed serum supplemented methods. Chemically controlled systems eliminate the risk of the aforementioned detrimental contaminants and will substantially reduce variation from lot-to-lot yielding improved growth and reproducibility. Consequently, such systems offer better economy over the long-term maintenance of a given cell line as fewer antibiotics are needed and regular sampling of sera is eliminated. In addition, serum-free conditions permit detailed investigation into unique processes such as receptor activation, growth factor and cytokine interactions, and apoptosis cascades. These type of studies are extremely difficult with serum supplemented systems. Finally, comparative studies of murine hybridomas demonstrate comparable growth rates and consistently up to 20% more antibody secretion under serum-free conditions.

Serum-Free Culture Media

The following defined CELLect™ liquid and powdered media are recommended for serum-free cell culture and are ideal for use with any of ICN's serum replacement or serum reduction products.

Basal Medium Eagle (BME)

	Catalog No.	Quantity
1X BME Liquid with Earle's salts; w/o L-glutamine	1200254	500 ml
1X BME Liquid with Earle's salts and L-glutamine	1200354	500 ml
BME Powder with Earle's salts and L-glutamine; w/o sodium bicarbonate	1000120 1000122	10x1 liter 1x10 liter
BME Powder with Hanks' salts and L-glutamine; wo sodium bicarbonate	1003120 1003122	10x1 liter 1x10 liter

See page 5 for an entire list of available BME media.

DMEM/F-12 Media

	Catalog No.	Quantity	
1X DMEM/F-12 Liquid with sodium pyruvate, phenol red and pyroxidine HCl; w/o HEPES buffer and L-glutamine	1246754	500 ml	
DMEM/F-12 Powder with phenol red and pyroxidine HCl; w/o L-glutamine, sodium pyruvate and sodium bicarbonate	1046920 1046922	10x1 liter 1x10 liter	
DMEM/F-12 Powder with phenol red, pyroxidine HCl and sodium pyruvate; w/o L-glutamine and sodium bicarbonate	1046820 1046822	10x1 liter 1x10 liter	
BioRich 1™ Powder with L-glutamine	1047120 1047122	10x1 liter 1x10 liter	

F-10 Nutrient Medium

	Catalog No.	Quantity	
1X F-10 Liquid w/o L-glutamine	1240249	100 ml	
, ,	1240254	500 ml	
1X F-10 Liquid with L-glutamine	1240349	100 ml	
	1240354	500 ml	
1X F-10 Liquid with 20 mM HEPES;	1240449	100 ml	
w/o L-glutamine and sodium bicarbonate	1240454	500 ml	
10X F-10 Liquid w/o L-glutamine and sodium	1440049	100 ml	
bicarbonate	1440054	500 ml	
F-10 Powder with L-glutamine; w/o sodium	1040120	10x1 liter	
bicarbonate	1040122	1x10 liter	
	1040124	1x50 liter	
F-10 Powder with L-glutamine; w/o sodium	1040220	10x1 liter	
bicarbonate and hypoxanthine	1040222	1x10 liter	

F-12 Nutrient Medium

	Catalog No.	Quantity	
1X F-12 Liquid with L-glutamine	1242354	500 ml	
1X F-12 Liquid w/o L-glutamine	1242249 1242254	100 ml 500 ml	
1X F-12 Liquid, Kaighn's Modification with L-glutamine	1242454	500 ml	
F-12 Powder with L-glutamine; w/o sodium bicarbonate	1042120 1042122	10x1 liter 1x10 liter	

Iscove's Modification of Dulbecco's Medium (IMDM)

	Catalog No.	Quantity
1X IMDM Liquid with L-glutamine, BSA, transferrin and lecithin (soybean)	1235854	500 ml
1X IMDM Liquid with L-glutamine, 25 mM HEPES and 2.52 g/L sodium bicarbonate; w/o BSA, transferrin and lecithin (soybean)	1235954	500 ml
IMDM Powder with L-glutamine; w/o BSA,	1035720	10x1 liter
transferrin, lecithin and sodium bicarbonate	1035722	1x10 liter
IMDM Powder with L-glutamine; BSA, transferrin	1035520	10x1 liter
and lecithin; w/o sodium bicarbonate	1035522	1x10 liter

NCTC 135 Medium

	Catalog No.	Quantity	
1X NCTC 135 Liquid with L-glutamine	1291354	500 ml	

RPMI 1640 Medium

	Catalog No.	Quantity	
1X RPMI 1640 Liquid w/o L-glutamine	1260249	100 ml	
pH 6.9-7.2	1260254	500 ml	
1X RPMI 1640 Liquid w/o L-glutamine	1265249	100 ml	
pH 7.2-7.4	1265254	500 ml	
1X RPMI 1640 Liquid with L-glutamine	1260349	100 ml	
	1260354	500 ml	
1X RPMI 1640 Liquid with 20 mM HEPES;	1260449	100 ml	
w/o sodium bicarbonate and L-glutamine	1260454	500 ml	
1X RPMI 1640 Liquid with 25 mM HEPES and 4.75 g/L NaCl; w/o L-glutamine	1260554	500 ml	
1X RPMI 1640 Liquid with 25 mM HEPES,	1260649	100 ml	
4.75 g/L NaCl and L-glutamine	1260654	500 ml	
1X RPMI 1640 Liquid Dutch Modification w/o L-glutamine	1260954	500 ml	
10X RPMI 1640 Liquid w/o L-glutamine and	1460049	100 ml	
sodium bicarbonate	1460054	500 ml	
RPMI 1640 Powder with L-glutamine;	1060120	10x1 liter	
w/o sodium bicarbonate	1060122	1x10 liter	
	1060124	1x50 liter	
RPMI 1640 Powder with L-glutamine and	1060520	10x1 liter	
25 mM HEPES; w/o sodium bicarbonate	1060522	1x10 liter	

Serum-free Insect Medium

	Catalog No.	Quantity
1X SFIM Liquid with L-glutamine	2720154	500 ml
SFIM Powder with glutamine	1127220 1127222	10x1 liter 1x10 liter

Serum-free Mammalian Medium

	Catalog No.	Quantity
1X SFMM Liquid with L-glutamine	2011054	500 ml
1X SFMM Liquid with L-glutamine; w/o phenol red	2012054	500 ml

Serum-free Virus Production Medium

	Catalog No.	Quantity
1X SFVM Liquid with L-glutamine	2730154	500 ml
SFVM Powder with glutamine	1127320	10x1 liter
·	1127322	1x10 liter

Waymouth's MB 752/1 Medium

	Catalog No.	Quantity
1X Liquid MB 752/1 w/o L-glutamine	1252254	500 ml
MB 752/1 Powder with L-glutamine; w/o sodium bicarbonate	1052120 1052122	10x1 liter 1x10 liter

Serum Replacements and Serum Extenders

ICN's serum replacements address the deficiencies, limitations and difficulties associated with the use of whole serum. Researchers worldwide agree that ICN's innovative products- TCHTM, TCMTM, TM-235TM, and VaxMaxTM- are superior for maintaining the consistent, long-term culture of a vast range of cell types including Vero, CHO, SP2/O, HeLa, DU145, MCF-7, human and murine hybridomas, LNcap, and HepG2.

The use of serum in cell culture continues widespread primarily because of unquestioned adherence to cited methods and perceived economy and convenience. Additionally, observable growth is often misinterpretted as proper cell function. But, the fact is that the composition of serum varies depending on the age, gender and overall health of the animal. Likewise, the collection time and seasonal factors can affect the quality of serum. Hence, components such as steroid hormones, growth factors and bacterial mitogens will vary from lot-to-lot. Serum is also a potential source of contaminants like mycoplasma, prions, viruses, and toxins. Any of which may affect the growth rate, phenotype, adherence and genetic stability of *in vitro* cultured cells.

- TCHTM a defined serum replacement formulated for supporting a vast range of human and mammalian cell lines.
- TCMTM a defined serum replacement specially formulated for supporting mammalian cell lines.
- TM-235™ a defined serum replacement similar to TCM™ specially formulated for supporting fastidious cell lines.
- VaxMaxTM a serum extender offering improved economy by reducing the total serum requirement for most cell cultures.
- ITS™ Premix a convenient supplement that reduces the total serum requirement for many cell cultures.

ICN's serum replacements and extenders eliminate the detrimental factors and variation associated with serum. They are free of detectable hormones, growth factors, cytokines, glucocorticoids, cell adhesion molecules and phenol red. Additionally, they are completely free of proteases, extraneous protein and infectious agents. Moreover, ICN's serum-free products offer the advantage of investigating unique processes not possible with serum including the cell regulation and adaptation to surface matrices devoid of serum artifacts, signal transduction receptor studies, the detailed investigation of cytokines and growth factors, drug discovery studies, and apoptosis mechanisms.

TCHTM is specially formulated for the propagation of human cell cultures and for the production of high purity cell secreted proteins. In addition, it has proven effective for culturing many other mammalian cell types needing no serum or additional animal protein. Similarly, TCMTM is a universal serum substitute specially formulated for supporting a vast range of mammalian cells. An insulin-free formulation is also available. TM-235TM offers the same advantages as both TCHTM and TCMTM but is specially formulated for fastidious cell lines with higher nutrient requirements. All three formulations offer superior performance and growth for the long-term maintenance of anchorage-dependent and suspension cell types.

ICN offers two serum extenders, VaxMax[™] and ITS[™] Premix. VaxMax™ is a serum-free convenient formulation that reduces the total serum requirement for most cell cultures. Cells types such as MDBK, MDCK, vero, CRFK, and swine testicle have demonstrated remarkable yields when supplemented with VaxMax™. Furthermore, it enhances production yields of veterinary vaccines while simultaneously reducing expense. VaxMax[™] is typically used with 0.5-1% serum. ITS[™] Premix also reduces the total serum requirement while stimulating proliferation for many cell types including rat contractile heart cells and human colon mucosal epithelial cells. This convenient preparation contains insulin, transferrin and selenium, and it requires as little as 2% serum for supporting diploid and heteroploid proliferation at equivalent or better growth rates.

TM-235™ - Total Serum Replacement

A universal serum replacement that supports a broad range of fastidious cells Completely free of virus, mycoplasma and other infectious agents Contains no growth factors, steroid hormones, extraneous proteins or phenol red Endotoxin - < 0.5 EU/ml

Supplied as a 50X concentrate Total protein (diluted to 1X) - 820 µg/ml Protein origin - bovine (U.S.)

Recommended storage - 4 - 8°C; stable for at least 7 months

Product	Catalog No.	Quantity	
TM-235 TM	2040026	20 ml	
	2040049 2040022	100 ml 2x100 ml	

TCH™ - Total Serum Replacement

A universal serum replacement that supports human and mammalian cells Completely free of virus, mycoplasma and other infectious agents Contains no growth factors, steroid hormones, extraneous proteins or phenol red Endotoxin - < 0.5 EU/ml

Supplied as a 50X concentrate Total protein (diluted to 1X) - 650 µg/ml Protein origin - human

Recommended storage - 4 - 8°C; stable for at least 7 months

Drug Master on file with the FDA

Product	Catalog No.	Quantity	
TCH TM	2010026	20 ml	
	2010049	100 ml	
	2010022	2x100 ml	

VaxMax[™] - Serum Extender

A defined serum extender that reduces the total serum requirement for most cell cultures

Completely free of virus, mycoplasma and other infectious agents

Contains no growth factors, steroid hormones, extraneous proteins or phenol red

Endotoxin - < 0.5 EU/ml Supplied as a 50X concentrate

Total protein (diluted to 1X) - 310 ug/ml

Protein origin - bovine (U.S.)

Recommended storage - 4 - 8°C; stable for at least 7 months

Product	Catalog No.	Quantity
VaxMax TM	2050049	100 ml
	2050054	500 ml

TCM™ - Total Serum Replacement

A universal serum replacement that supports mammalian cells Completely free of virus, mycoplasma and other infectious agents Contains no growth factors, steroid hormones, extraneous proteins or phenol red Endotoxin - < 0.5 EU/ml

Supplied as a 50X concentrate Total protein (diluted to 1X) - 650 µg/ml

Protein origin - bovine (U.Ś.)

Recommended storage - 4 - 8°C; stable for at least 7 months

Drug Master on file with the FDA

Product	Catalog No.	Quantity	
TCM TM	2020026	20 ml	
	2020049	100 ml	
	2020022	2x100 ml	
TCM™, Insulin-free	2060026	20 ml	

ITS™ Premix- Serum Extender

A serum extender that reduces the total serum requirement for many cells Completely free of virus, mycoplasma and other infectious agents Contains insulin, transferrin and selenium

Each vial supplements 5 liters of medium

Requires as little as 2% serum to yield equivalent or better growth rates Recommended storage - 4 - 8°C

Product	Catalog No.	Quantity	
ITS™ Premix	2001227	1 vial	