CELLMAX®
Hollow Fiber
Cell Culture System

- Monoclonal Antibody Production
- Lymphocyte Expansion
- Secreted Protein Production
- Endothelial Cell Culture

SPECTRUMLABS.COM

cell culture

CELLMAX® DUO Hollow Fiber Cell Culture System

Content

Introduction

Conventional Cell Culture Technologies
Cell culture is a laboratory tool used primarily for the production of cells and cell products. For many applications, conventional cell culture technologies prove inefficient, cumbersome, costly and labour-intensive, all too often hindering rapid progress in the laboratory.

Two-Dimensional Cultures
Cells have a natural tendency to grow in three dimensions. Two dimensional substrates, such as T-flasks and roller bottles, are not designed for layered cultures. As cells pile up, cells in lower layers can quickly perish from slow diffusion of oxygen, nutrients and metabolic waste.

Nutrient medium must be frequently replaced, and cell populations must be minimized to prevent overcrowding and layering. Medium replacement causes sudden, frequent changes in microenvironment immediately adjacent to the cells. These changes often prevent the accumulation of beneficial cell-secreted growth factors.

Suspension Cultures
In suspension cultures, cells are continuously agitated in nutrient medium. Some cells are too fragile to tolerate this environment. Culture density is also severely limited, as a high-density culture would not permit adequate agitation, feeding and oxygenation of cells due to a higher viscosity of the medium.

In order to continually divide and function, most cells also require costly medium comprised of 5 – 20% serum proteins. Moreover, this technology requires that the desired cell secreted products be concentrated from very large volumes of medium.

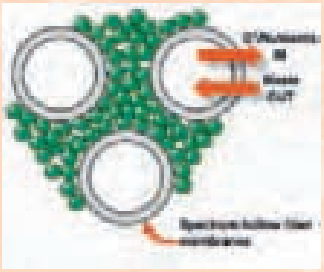
CellMax® is superior to all cell culture systems. Except one . . .

To create a better way to grow cells ***in vitro***, we took the ***in vivo*** process as our model. Of all *in vitro* cell culture technologies, nothing comes closer to duplicating ideal, *in vivo* growth conditions than CellMax®.

CELLMAX® DUO Hollow Fiber Cell Culture System

We duplicated the *in-vivo* process as closely as possible to produce the optimal pericellular environment for cell growth as well as maximal secretion and accumulation of cell products. The result is artificial capillary cell culture technology using hollow fibers. The best realized in the CellMax® Hollow Fiber Cell Culture System.

CELLMAX®, Hollow Fiber Cell Culture Technology

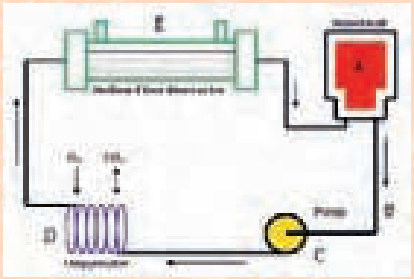


Initiation of Culture
In the CellMax®, cells are grown on and around a network of artificial capillaries encased in a cartridge shell. Cells suspended in tissue culture medium are inoculated into the extra capillary space of the cartridge and settle onto the outer surface on the capillaries as shown in the figure. Cells remain in the extra-capillary space during culture, protected from the shear of the rapidly perfusing, continually circulating medium.

Nutrient supply
Nutrients and oxygen in the circulating medium readily diffuse through the capillary walls to nourish the cells. Metabolic waste products similarly diffuse away from the cells and are diluted in the circulating medium.

Oxygenation
To remain viable, cells require a significant amount of oxygen. The entire CellMax® System operates in a standard CO_2 incubator where oxygen from the incubator atmosphere diffuses through the walls of the system's silicone tubing.

System description



Referring to the figure, culture medium is drawn from the medium reservoir bottle (A) through silicone tubing (B). The adjustable pump compresses the thick walled tubing (C) causing the medium to flow through the oxygenator (D) where oxygen and carbon dioxide are exchanged.
From the oxygenator the medium flows into the hollow fiber bioreactor (E) through the lumen of each hollow fiber within the hollow fiber bundle where it perfuses the cells growing on the outside of the fibers with fresh nutrients and oxygen and removes metabolic waste products. There is no direct medium flow within the extra capillary space between the capillaries. The medium is then returned to the reservoir bottle.

CellMax® DUO System

System Description
The CellMax® Duo base consists of a two-position pump station and bottle holders to mount up to two bioreactor modules and reservoir bottles simultaneously.

As the pump motor rotates, a cam on the motor shaft forces the Teflon pump bars to depress the thick-walled pump tubing on the hollow fiber bioreactor module. This action forces culture media to flow from the media reservoir bottle through the gas permeable silicone flow path tubing into the hollow fiber bioreactor module.

The Teflon pump bars are asymmetric to allow different flow curves to be generated. One side of the Teflon pump bar has a section cut out to allow a smaller contact surface with the tubing. This reduces the overall displacement of the tubing to allow smaller flow rates to be achieved. The CellMax® Duo system is normally operated inside a humidified, temperature-controlled CO_2 incubator. The pH of the recirculating media is regulated via diffusion of CO_2 through the walls of the silicone tubing. For most cells, a CO_2 setting of 5-10% provides a media pH that is optimum for cell growth.

The CellMax® Duo can also be used with CO_2-independent media. This allows the researcher to support the growth of most cell lines using a standard 37°C incubator or warm room.

ECU Operation
The ECU is connected to the DUO Base by the flat motor cable, and controls the speed of the pump motor. The ECU is placed outside the incubator for easy visualization of the control panel during operation. The ECU contains a magnetic plate on the back of the enclosure to attach to the wall of the incubator. The ECU is activated by moving the rocker switch on the front of the ECU to the ON position. The RED LED on the rocker switch ensures that the ECU is on. The variable speed dial on the front of the ECU can adjust the flow rate.

CELLMAX® DUO Hollow Fiber Cell Culture System

The CellMax® Duo System includes
- CellMax® Duo Base
- Electronic Control Unit (ECU) and Power supply
- Power Cord
- Teflon Pump Bars (2)
- Instruction Manual

Specifications:
- Dimensions: 25,4 x 33,0 x 16,5 cm
- Weight: 2,27 kg
- Power: 100 – 240 VAC, 50/60 Hz Input 0,8 A
- Flow rates: 5 - 120 ml/min (w/o bioreactor)
- Drive Motor: Brushless DC Motor
- Environment: 20 - 42° C, 100% Relative Humidity
  CO_2 Incubator (Base only)

CellMax® Hollow Fiber Bioreactors

A low cost alternative to conventional Cell Culture Methods: one CellMax® Bioreactor equals the yield of 100 T-Flasks!

Unlike traditional cell culture methods, T-flasks, roller bottles, and two compartment bioreactors, CellMax Hollow Fiber Bioreactors provide a three dimensional space for *in vitro* cell culture. Hollow fibers are tubular membranes with pore sizes ranging from 10kD to 0.3µm. Cells grow on and around the large surface area provided by the network of hollow fibers. When perfused with culture media, the hollow fibers allow oxygen and nutrients to be supplied to the cells while metabolic waste products are eliminated. The process increases the accumulation of the cell-secreted growth factors required for optimal growth. Due to the system's efficient delivery of media and removal of waste, the process uses less culture media than other methods. Cultures can be sustained for 6 months or more while maintaining high production yields and titers.

Applications
- Monoclonal Antibody Production
- Recombinant Protein Production
- Stem Cell Expansion
- Endothelial Cell Shear Stress Research
- In Vitro Pharmacology Studies
- Lymphocyte Expansion
- Blood Brain Barrier Modeling
- Bioartifical Liver, Pancreas & Kidney Devices
- Retroviral Vector Production

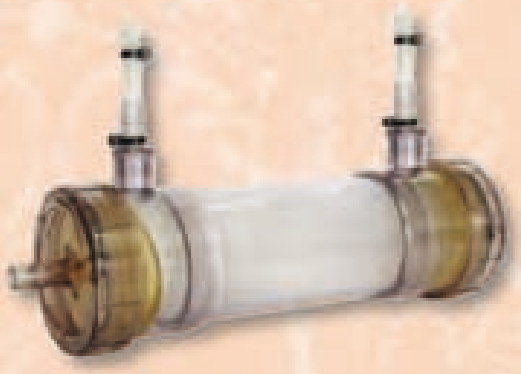
CellMax Hollow Fiber Bioreactors
- 4 Membrane Chemistries including:
  - Regenerated Cellulose (RC)
  - Polysulphone (PS)
  - Polypropylene (PP)
  - Polyethylene (PE)
- ProNectin® F coated PP and PE membranes for enhanced cell adherence
- Surface area ranging from 100 cm^2 to 1.6 m^2
- Animal Derived Component Free
- Fully Assembled and Preflushed
- Irradiated and Ready-to-Use
- 100% Integrity test

Clave® Aseptic Needle Free Access Port
- Reduces the risk of cell culture contamination
- Provides easy access to the ICS for monitoring glucose and lactic acid levels
- Provides easy access to the ECS for Cell Inoculation or Protein Harvest

CELLMAX® DUO Hollow Fiber Cell Culture System

ZYMAX® Hollow Fiber Bioreactors For Large Scale Cell Culture Applications

The Zymax® hollow fiber bioreactor offers the advantage of three dimensional cell growth for large cell culture applications. Cells are inoculated into the extra capillary space of the hollow fibers while culture media is circulated through the lumen. The porosity of the fibers allows nutrients and oxygen to perfuse the culture while CO_2 and metabolic wastes are removed. Based on the mammalian circulatory system the unique Zymax® bioreactor provides much denser cell growth with better viability than other conventional cell culturing systems such as flasks and roller bottles.



Standard specifications

Membrane Type	Fiber ID	Pore Size
Regenerated Cellulose (RC)	0,2 mm	30 kD
Polysulfone (PS)	0,2 mm	10 kD
Polysulfone (PS)	0,2 mm	30 kD

Housing: Clear Polysulfone
Potting material: Polyurethane
Side port connections: 3/8 inch Hose Barb (HB) or Female Luer Lock (FLL)
Surface area: up to 3,1 m²

Standard Bioreactors

Product number*	Membrane	Pore size	Inlet/Outlet connections
Z71R-102-01N	RC	30 kD	½ inch HB
Z71R-102-01S	RC	30 kD	¼ inch HB
Z81R-102-01N	RC	30 kD	½ inch HB
Z81R-102-01S	RC	30 kD	¼ inch HB
Z71S-102-01N	PS	10 kD	½ inch HB
Z71S-102-01S	PS	10 kD	¼ inch HB
Z81S-102-01N	PS	10 kD	½ inch HB
Z81S-102-01S	PS	10 kD	¼ inch HB
Z73S-102-01N	PS	30 kD	½ inch HB
Z73S-102-01S	PS	30 kD	¼ inch HB
Z83S-102-01N	PS	30 kD	½ inch HB
Z83S-102-01S	PS	30 kD	¼ inch HB

* Product numbers ending with "N" are NON irradiated (sterile) and with "S" are irradiated (sterile)
The Zymax® bioreactors are available with standard product specifications but also with custom designed options.
Please contact Spectrum Europe for your custom design solutions: info@spectrumeurope.nl or call +31 76 5719 419

CellMax® Applications

• **Monoclonal Antibody Production:** The CellMax® Artificial Capillary system is the method of choice for the *in vitro* production of 50 mg to several grams of monoclonal antibody. A cellulosic fiber with a MWCO of 30 kD is used. This permits the inhibitory factor TGF beta to freely diffuse away while trapping the higher molecular weight immunoglobulins along with the hybridoma cells in the extra capillary space. Cell density can become higher than 10^8 per ml and antibody concentration is in the range of 0,5 mg to 5 mg per ml. Harvest volume is only 10 to 20 ml each cartridge will produce antibody for up to 4-6 months.

Pore size of hollow fibers: large pore size allows MAb to diffuse out • small pore size keeps in MAb and metabolic waste products • hybridoma-specific pore size keeps MAb in ECS and allows waste to be removed

Pore size chart:

Molecular Weight cutoff	10 kD	30 kD	0.3 µm pores
IgG retention (146 kD)	Yes	Yes	No
IgM retention (970 kD)	Yes	Yes	No
TGF-BETA retention (21 kD)	Yes	No	No

CELLMAX® DUO Hollow Fiber Cell Culture System

• **Secreted Protein Production:** Current methods for the production and harvest of secreted biologicals such as growth factors, recombinant proteins and antibodies involve the use of inefficient *in-vitro* culture systems or animals. CellMax® Hollow Fiber Cell Culture Systems permit the culture of large numbers of cells (up to 5 x 10^{10} cells) within a small volume extra capillary space (10 to 100 ml). Selection of appropriate fiber MWCO can trap the secreted product in this small volume while allowing lower molecular weight nutrients and waste products to diffuse into the circulating medium. Both total concentration of product and product secreted per cell can be increased by a factor of 10 or more using the CellMax® system.

• **Lymphocyte Expansion:** Repeated culture and harvest of primary lymphocytes can be achieved with the CellMax® Hollow Fiber Cell Culture System. Modules are available for the expansion of lymphocytes up to 10^{10} per module. This method can be used to produce and collect significant amounts of lymphocytic growth factors. CellMax® modules have also been used to increase the efficiency of viral transductions.

• **Endothelial Cell Culture:** Spectrum has developed several Hollow Fiber Cell Culture Cartridges specifically designed for the culture and study of endothelial cells under flow. The system provides a means to evaluate endothelial cell functions in a more physiological environment when compared to other methods. As a result, typical cell morphology is maintained, allowing the investigator to generate results that more closely reflect the *in vivo* situation.

** Please ask Spectrum Europe for your specific application.*

Ordering information

HOLLOW FIBER CELL CULTURE SYSTEM

	Description
CMMAX-DUO	CellMax® DUO - two cartridge capacity - CE certified Power: 100 – 240 volt, 50/60 Herz, Input 0,8 A Flow rate: 5 – 120 ml/min (11 settings) Environment: 20 – 42° C, 100% Relative Humidity CO_2 Incubators (Base only) Positive pressure pump with variable flow rates for efficient delivery of nutrients and waste removal. Due to the compact design the CellMax® DUO Pump requires less space than traditional cell culture techniques and can be placed in an incubator. The cool running motor does not affect incubator temperature.

CELLMAX® HOLLOW FIBER BIOREACTORS

	Type	Membrane	MWCO	Area (cm^2)	ID (µm)	OD (µm)	ECS (ml)
430-011	RC	Cellulosic	30 kD	1.700	200	216	12
430-021	RC	Cellulosic	30 kD	10.000	200	216	60
400-004	RC	Cellulosic	20 kD	16.000	192	200	100
430-013	PS	Polysulfone	30 kD	1.300	200	320	12
430-023	PS	Polysulfone	30 kD	10.000	200	320	60
430-010	PS	Polysulfone	10 kD	1.700	200	280	12
430-020	PS	Polysulfone	10 kD	10.000	200	280	60
410-025	PP	Coated Polypropylene	0,2 µm	100	480	630	1,5
400-025	PP	Polypropylene	0,2 µm	100	480	630	1,5
420-015	PE	Coated Polyethylene	0,3 µm	123	380	430	2,3
400-014	PE	Polyethylene	0,3 µm	123	380	430	2,3
420-007	PP	Coated Polypropylene	0,5 µm	423	480	630	7
400-012	PE	Polyethylene	0,3 µm	1.500	380	430	12

CELLMAX® DUO Hollow Fiber Cell Culture System

CELLMAX® SYSTEM ACCESSORIES

	Description	
010-006	Replacement Flowpath, Ready-to-Use standard flowpath for hollow fiber bioreactors < 1 m^2 S.A.	
010-007	Replacement Flowpath, Ready-to-Use flowpath for increased oxygenation for hollow fiber bioreactors > 1 m^2 S.A.	
100-020	Reservoir Cap, 33 mm, Polysulfone cap for 500 ml & 1 L glass bottles	
100-025	Reservoir Cap, 38 mm, Polysulfone cap for 500 ml & 1 L square plastic bottle	
100-035	Reservoir Cap, 45 mm used with new Gibco® bottle	
100-145	CellMax® Starter Kit This kit should enable the researcher to immediately begin hollow fiber bioreactor cell culture	Includes: - Non-sterile Fitting Female Luer, W 3/16" HB (25) - Non-sterile Fitting, Male Luer x 3/16" HB (25) - Sterile Syringe 10 cc Luer Lock (6) - Sterile Syringe 60 cc (4) - Non-sterile Glass Bottle & Cap, 125 ml (1) - Sterile Needle 15 gauge blunt (6) - Sterile Swabs Alcohol 70% (60) - Sterile Square Petg. Bottle 125/38-430 (1) - Sterile Square Petg. Bottle 250 ml 38-430 (1) - Sterile Plug Male/female Luer Lock (4) - Sterile Syringe 30 cc (6)

MediaKap® & MediaKap® Plus HF Filters

Sterile Filtration of Culture Medium w/ Serum

For fast sterile filtration of media

While MediaKap® Filters are designed to efficiently sterilize and clarify culture medium or buffer solution by means of a 0.2 µm DynaFibre® membrane, MediaKap Plus Filters utilize an advanced DynaFibre® membrane that further increases filtration efficiency and dramatically reduces the time required to filter serum-enriched medium. Both can be operated by gravity or under pressure by a peristaltic pump or pressure vessel. Sterile MediaKap and MediaKap Plus Filters are available in 5 different sizes that filter the designated volume (2 , 5, 10, 25 or 50 liter) in 15 - 20 minutes. Each is available with the option of a filling bell that protects the sterile filling environment and reduces the risk of contamination.

Specifications		Components	
2 Filter Types:	MediaKap & MediaKap Plus	HF Membrane:	Mixed cellulose ester
5 Volume Sizes:	2, 5, 10, 25 and 50 liter	HF Vent:	Polypropylene
Pore Rating:	0.2 µm	Potting Material:	Polyurethane
Membrane SA:	refer to Ordering Information	Housing:	Clear polysulfone
Inlet/Outlet Conn.:	refer to Ordering Information	End-caps:	Clear & pigmented polysulfone
Packaging:	Gamma irradiated	Filling Bell:	PVC (if present)

Performance Specifications

Filter Size & Type	Water Flow @ 10 psig	DMEM w/o serum	DMEM w/ serum	Filter Size & Type	Water Flow @ 10 psig	DMEM w/o serum	DMEM w/ serum
MediaKap-2	400 ml/min	0.2 – 2 L	-	MediaKap-2 Plus	400 ml/min	5 L	0.2 – 2 L
MediaKap-5	750 ml/min	2 – 5 L	-	MediaKap-5 Plus	750 ml/min	10 L	2 – 5 L
MediaKap-10	1000 ml/min	5 – 10 L	-	MediaKap-10 Plus	1000 ml/min	20 L	5 – 10 L
MediaKap-25	1400 ml/min	10 – 25 L	-	MediaKap-25 Plus	1400 ml/min	50 L	10 – 25 L
MediaKap-50	2000 ml/min	25 – 50 L	-	MediaKap-50 Plus	2000 ml/min	100 L	25 – 50 L

CELLMAX® DUO Hollow Fiber Cell Culture System

Ordering information

Part No.	Description	Pore Rating	Surface Area	IN / OUT Conn	IRR	Filling Bell	Qty/Pkg
ME2M-02B-12S	MediaKap®-2	0.2 µm	35 cm^2	FLL / 1/4HB	YES	YES	12
ME2M-020-18S	MediaKap®-2	0.2 µm	35 cm^2	FLL / 1/4HB	YES	NO	18
ME2M-05B-12S	MediaKap®-5	0.2 µm	70 cm^2	FLL / 1/4HB	YES	YES	12
ME2M-050-18S	MediaKap®-5	0.2 µm	70 cm^2	FLL / 1/4HB	YES	NO	18
ME2M-10B-12S	MediaKap®-10	0.2 µm	100 cm^2	1/4HB / 1/4HB	YES	YES	12
ME2M-25B-06S	MediaKap®-25	0.2 µm	185 cm^2	VarHB / VarHB	YES	YES	6
ME2M-50B-03S	MediaKap®-50	0.2 µm	440 cm^2	VarHB / VarHB	YES	YES	3
MP2M-02B-12S	MediaKap-2 Plus	0.2 µm	35 cm^2	FLL / 1/4HB	YES	YES	12
MP2M-020-18S	MediaKap-2 Plus	0.2 µm	35 cm^2	FLL / 1/4HB	YES	NO	18
MP2M-05B-12S	MediaKap-5 Plus	0.2 µm	70 cm^2	FLL / 1/4HB	YES	YES	12
MP2M-050-18S	MediaKap-5 Plus	0.2 µm	70 cm^2	FLL / 1/4HB	YES	NO	18
MP2M-10B-12S	MediaKap-10 Plus	0.2 µm	100 cm^2	1/4HB / 1/4HB	YES	YES	12
MP2M-25B-06S	MediaKap-25 Plus	0.2 µm	185 cm^2	VarHB / VarHB	YES	YES	6
MP2M-50B-03S	MediaKap-50 Plus	0.2 µm	440 cm^2	VarHB / VarHB	YES	YES	3

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