



CDM-HD SERUM REPLACEMENT

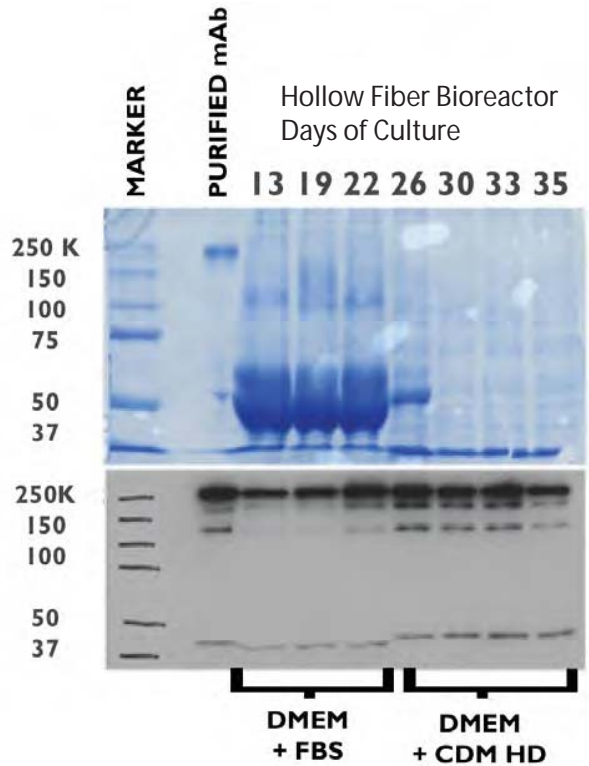
Chemically Defined Medium for High Density Cell Culture

- Purification Simplified
- Protein-Free
- Chemically Defined
- Use like Serum



CDM-HD Serum Replacement is a chemically defined, protein free serum replacement that permits any basal medium such as **DMEM** or **RPMI** to be used without serum. **CDM-HD** is designed specifically for the culture of cells at high density and optimized for use in our hollow fiber bioreactor systems. Secreted products such as monoclonal antibodies and recombinant proteins are free of contaminating proteins from the medium and can be purified using simplified protocols, increasing net yield in many cases. CDM HD provides lot-to-lot consistency and is an economical replacement for serum. It is available as a dry powder to make up one liter and is used at a concentration of 10%. The cost of CDM-HD is half that of serum giving you all the benefits of a chemically defined, protein free medium without the high price.

FEATURES	BENEFITS
Chemically defined	lot-to-lot consistency
Protein free	purification of secreted products simplified
Animal component free	regulatory compliance simplified, no agents of unknown origin such as mycoplasma or viruses
Dry powder	long shelf life
Store at 4 degrees	simple storage conditions

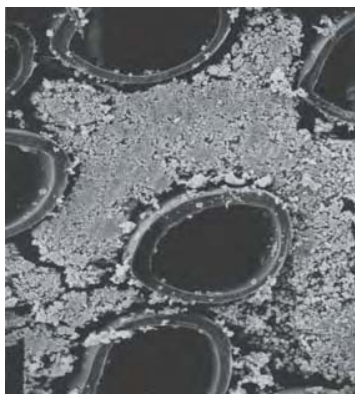


The beneficial effects of serum supplementation are limited by several disadvantages: serum is an ill-defined mixture and often contains adventitious agents and by-products like bacterial endotoxins and other immunogenic contaminants. Furthermore, batch qualities vary and the costs can be high. Sera are a major source of viral contaminants that often do not produce cytopathic effects or morphological changes but once present are almost impossible to remove from cultures. Some important advantages in the use of totally defined media are the avoidance of risks of contamination and immunogenic stimuli and lot-to-lot consistency. Possible contaminants include viruses, prions and mycoplasma which highly impair pharmacologic use of culture derived products. Taking things one-step further a totally defined, animal component free, protein free medium will not contain any proteins nor any materials derived from animals and will have high lot-to-lot consistency. Purification of the desired secreted product is facilitated and regulatory compliance is made much simpler.

Optimized for Hollow Fiber Bioreactors

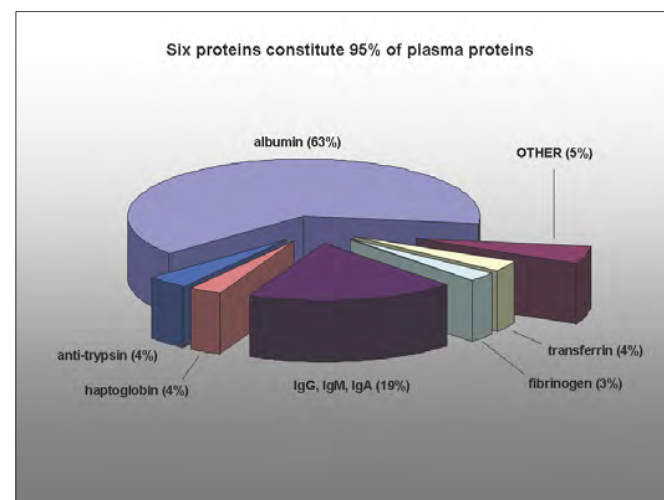
- Increased buffering capacity maintains optimum conditions for protein and antibody production.
- No detergents or other cell membrane stabilizing agents that sometimes interfere with assays or precipitations.
- Composition balanced for high-density culture.

Hollow fiber cell culture is an ideal method for the production and collection of secreted products from mammalian cells as large numbers of cells can be supported in a small space and the secreted product is concentrated to a high degree. The high surface area to volume ratio that hollow fiber bioreactors provide (100cm²/ml or more) and the extreme high flux rates of fibers from FiberCell Systems can allow cells to grow at 100X the density found in flask culture (10⁸ vs. 10⁶/ml) and is in fact one of the only cell culture methods that permits cell cultures at densities similar to those found in vivo. High cell density allows cells to be grown in reduced serum environments (as low as 2% vs. 10% for conventional culture methods) and more easily adapted to



ABOVE: Cross-section of hollow fibers in FiberCell[®] Systems hollow fiber bioreactor. High-density cell growth is shown in extracapillary space.

commercially available serum free mediums. However, these mediums can be expensive and sometimes optimal for only a limited number of cell types. The concept of CDM-HD is simple: produce a universal serum substitute that can take advantage of the simplified serum requirements for cells grown in hollow fiber bioreactor systems. CDM-HD represents a true breakthrough in cell culture ideology and technology. It is the first time that a specific culture medium takes advantage of the growth properties of cells to simplify and optimize that medium for those specific growth conditions.



Above: Various components of serum. The major component, BSA binds many substances non-specifically and can be an overwhelming protein burden in some purification schemes, immunoglobulins can interfere with specific antibody purifications and the "other" can have many undefined effects. None of these characteristics will be reproducible from lot to lot.

Instructions for use: Reconstitute the contents of one bottle of CDM-HD to make a total volume of 1 liter. Adjust pH to no higher than 6.5 using 1N NaOH. Sterile filter, do not autoclave. Add to any basal medium at a concentration of 10%. When using with FiberCell hollow fiber bioreactors add 10% fetal calf serum to the cell inoculum when loading cells to provide attachment factors. No adaptation is required though you may see a reduction in glucose uptake rate for the first day or two of use. CDM-HD is stable for at least 1 month as a concentrate when stored at 4 degrees and away from light.

Notes:

- CDM-HD will not require any adaptation but is optimized for cells growing at high density. For best results switch to CDM-HD after cells have reached a high density inside the FiberCell hollow fiber bioreactor, typically after one week of culture or so.
- CDM-HD does not contain any carbon source. When working with a low glucose medium such as RPMI it is important to supplement the glucose concentration to 4.5 grams per liter.
- CDM-HD is protein free and does not contain any cell attachment factors. When inoculating cells in medium containing CDM-HD add 10% FBS to the cell inoculum only. This will provide the required attachment factors for adherent cells. This is not required for suspension cells though hybridoma cell lines should be treated as adherent cells.
- CDM-HD can be used in other culture systems such as spinner culture and roller culture. For these applications add plurionic F60 or some other cell membrane protecting surfactant.
- CDM-HD is protein free. The secreted protein of interest may be the only significant protein present in your cell culture supernatant. You should re-evaluate your purification protocols as entire steps can sometimes be eliminated, increasing yield. Keep in mind that CDM-HD is protein free and contains no ferritin so the free iron levels will be higher than in standard mediums. Pay attention to any chelating agents that may be part of your buffers or purification protocol.