CLONING AND EXPRESSION

Cell Culture Media Serum Free Optimized CHO Media

For successful culture of Chinese Hamster Ovary Cells

The expression of recombinant proteins has increased in importance for both research and biopharmaceutical manufacturing applications. An increasing number of mammalian-derived proteins require post-translational modifications for full biological function. The popular Chinese Hamster Ovary (CHO) cell line is frequently used to provide these necessary modifications. Sigma's CHO media has been optimized to promote high levels of recombinant protein expression. Additionally, because the formulation is serum-free, it eliminates the concerns associated with the detrimental effects of animal-derived components.

Sigma has devoted a major portion of its cell culture development efforts to optimizing media for biopharmaceutical production that is chemically defined as well as completely free of animal components.

Features & Benefits

- Contains no serum or animal components
- Outperforms the competition by consistently producing more viable cells than competing serum-free formulations
- Increases in protein productivity up to 500% per cell have been observed
- Cells grow at a rapid pace with little or no adaptation required

CHO Medium, Animal Component-free

Designed for maximum productivity in suspension cultures, CHO-PF-AF can support viable densities up to 8 x 10^6 cells/ml in fed-batch culture systems and can increase recombinant protein productivity on a per-cell basis by 500% or more over competitive formulations (Fig. 1). Contains recombinant human insulin, Pluronic[®] F-68, and plant-based hydrolysates.

CHO DHFR Medium, Animal Component-free

This latest addition to the growing line of animal component-free CHO media from Sigma-Aldrich has been designed to provide outstanding cell growth and recombinant protein productivity in Chinese hamster ovary (CHO) cells that are engineered to contain the dihydrofolate reductase (DHFR) amplification system. Contains recombinant human insulin, Pluronic[®] F-68, and plant-based hydrolysates.

CHO Medium for Attached Cells, Animal Component-free

The latest CHO medium from Sigma-Aldrich has been designed to support the growth of CHO cells in adherent culture systems. Maintains attachment of cells for extended periods and promotes outstanding recombinant protein production (Fig. 2). Contains recombinant growth factors and hormones, and plant-based hydrolysates.

CHO Medium Chemically-defined, Animal Component-free

This newly developed chemically-defined CHO medium provides excellent cell growth and productivity without the presence of hydrolysates or other undefined materials. Designed to meet the needs of manufacturers seeking to optimize down-stream processing as well as researchers studying nutritional and physiological controls of cellular processes. Contains recombinant human insulin and Pluronic F-68.

Product Code	Description	Size
<u>C 5467</u>	CHO Medium Liquid, Protein-free, Animal Component-free, cell culture tested	1 L 6 x 1 L
<u>C 8862</u>	CHO CHFR Medium Animal Component-free	1 L 6 x 1 L
<u>C 8730</u>	CHO Medium Liquid, Animal Component-free for Adherent Cultures, cell culture tested	1 L 6 x 1 L
<u>C 4726</u>	CHO Medium Liquid, Chemically-defined Animal Component-free, cell culture tested	500 ml 6 x 500 ml 1 L 6 x 1 L
<u>C 1707</u>	CHO Serum-free Medium without L-Glutamine	1 L 6 x 1 L



Figure 1. Cell growth and IgG production in a proprietary CHO cell line were compared under suspension culture conditions in <u>C 5467</u> medium or a leading competitor's proteinfree medium. CHO cells were seeded at 50,000 cells/ml in spinner flasks. Samples were taken daily to analyze cell growth and protein production.



Figure 2. CHO Medium.

Cell growth and IgG production in a proprietary CHO cell line were compared under adherent culture conditions in <u>C 8730</u> medium or DME/F12 with 10% FBS in T-flasks. Spent medium samples were collected daily for analysis of IgG production. The attached cells were trypsinized and counted for cell growth. The data indicates cell growth in CHO medium was not as high as DME/F12 with 10% FBS, but production of recombinant IgG was much greater.

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- 1) 100% A
- 2) 100% B
- 3) 100% C
- 4) 50% A, 50% B
- 5) 50% B, 50% C
- 6) 50% A, 50% B
- 7) 67% A, 17% B, 17% C
- 8) 67% B, 17% C, 17% A
- 9) 67% C, 17% A, 17% B
- 10) 33% A, 33% B, 33% C

Figure 1:

Following the initial screening of the 6 media contained in this kit, researchers can perform additional blending experiments with the three best performing media. This diagram and table represents the recommended blends for this initial set of experiments for media A. B. and C.

Components

Free

CHO Medium, Animal Component-Free CHO DHFR- Medium, Animal Component-

CHO Medium, Chemically Defined, Animal Component-Free

CHO Medium 4, Animal Component-Free

CHO Medium 5, Animal Component-Free

CHO Medium 6, Chemically Defined, Animal Component-Free

CHO Kit 1

Faster, easier medium optimization is now in your hands

Researchers have long realized that individual Chinese hamster ovary (CHO) cell clones have highly specific nutritional needs in order to reach maximal cell growth and productivity. To help researchers optimize their CHO systems, we designed CHO Kit 1.

CHO Kit 1 (Product Code CH0001) from Sigma-Aldrich is the latest addition to the fastest growing line of animal component-free media in the industry. This novel kit provides researchers a convenient format in which they can rapidly screen multiple formulations to identify which formulations work best for their particular clones. It also provides detailed protocols utilizing Design-of-Experiment (DOE) methodology to allow researchers to take the three best performing media and perform blending experiments to further optimize media specific for their particular CHO systems.

The kit consists of six diverse animal component-free media designed to maximize cell growth and recombinant protein productivity in a wide variety of CHO cell clones. Two of these six media are chemically defined and consist of differing amounts of amino acids, vitamins, salts, trace elements, recombinant human insulin, Pluronic[®] F-68, and other organic compounds. The remaining four media contain plant-based protein hydrolysates in addition to the above list of compounds. These six media, as well as any media blends derived from them, are designed to meet the raw material needs of the biopharmaceutical industry and are scalable under cGMP.

CHO Kit 1 from Sigma-Aldrich provides the following benefits:

- Six unique animal component-free media, including two chemically defined formulations, designed for maximum recombinant protein production in CHO cells
- Convenient format allows for rapid screening of multiple media to select optimal formulations
- Simple yet powerful blending experiments (using DOE methodology) lets researchers quickly perform further media optimization experiments
- Detailed instructions for various methods of data analysis are included
- Don't want to perform the data analysis yourself? E-mail your normalized data to cellculture@sial.com and we'll do it for you! At no additional charge!

Product Code	Description	Size
<u>CH0001</u>	CHO Kit 1, Animal Component-Free	1 kit

Recombinant Protein oning and **Expression**

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