
Also Available for CHO Cells:

IS CHO-V™

Catalog No. 9197

A serum-free medium with only non-mammalian origin components. Formulated for dihydrofolate reductase (dhfr) selection.

IS CHO-V-GS™

Catalog No. 9198

Similar to IS CHO-V™, but designed for use with the GS selection system.

IS CHO-CD™

Catalog No. 91119

A chemically-defined medium designed for use with dihydrofolate reductase (dhfr), Glutamine Synthetase (GS) or other selection systems.



IrvineScientific®

2511 Daimler Street, Santa Ana, California 92705-5588

Telephone: 1 949 261 7800 • 1 800 437 5706

Fax: 1 949 261 6522 • www.irvinesci.com

PN 40630 Rev. 1

IS CHO™ MEDIUM

Catalog #91109

1 L Liquid

DESCRIPTION

IS CHO™ medium is a serum-free medium optimized for the production of recombinant proteins in Chinese Hamster Ovary (CHO) cells.

INTENDED USE

For further manufacturing use. IS CHO is a growth medium for the production of recombinant proteins in CHO cells.

FORMULA

IS CHO medium is provided without L-Glutamine to extend shelf life and allow the use of L-Glutamine feeding strategies. The recommended L-Glutamine concentration to add is 8 mM (40 mL per liter of a 200 mM L-Glutamine solution, Catalog #9317). The medium should be used without L-Glutamine if the GS selection system is being used. IS CHO medium has been designed without Hypoxanthine and Thymidine for use with the dihydrofolate reductase (dhfr) and other selection systems. IS CHO medium contains protein hydrolysates. This medium contains no antibiotics or antimycotics.

PRECAUTIONS

Handle using aseptic techniques to avoid contamination.

Storage: Store at 2-8° C, protected from light. Do not use after the assigned expiration date.

Indications of Deterioration: Do not use if cloudy or if solution precipitates.

INSTRUCTIONS FOR USE

Adaptation

I. Direct Adaptation from serum-supplemented media to IS CHO.

In most cases, CHO cells may be subcultured from a serum-supplemented medium (e.g., Ham's F-12/DME + 10% FBS) directly into IS CHO.

1. Dispense IS CHO medium into a culture vessel and equilibrate to 37° C and 5% CO₂.
2. Passage CHO cells from serum-supplemented culture into IS CHO at 3 x 10⁵ viable cells/mL. It is important that cells be in the logarithmic phase of growth with at least 90% viability before passaging.
3. Incubate cultures at 37° C and 5% CO₂ until the viable cell density reaches 1 x 10⁶ cells/mL.
4. Subculture into fresh IS CHO medium at 2 x 10⁵ cell/mL starting density.
5. Maintain cells in IS CHO for several passages, subculturing twice weekly to allow complete adaptation and assure optimum performance.

II. Sequential adaptation from serum-supplemented media to IS CHO.

Sequential adaptation may be used if direct adaptation is troublesome.

1. Dispense the original serum-supplemented medium and IS CHO medium in a 3:1 ratio into an appropriate culture vessel and equilibrate to 37° and 5% CO₂.
2. Passage CHO cells from serum-supplemented culture into 3:1 IS CHO at 3 x 10⁵ viable cells/mL. It is important that cells be in the logarithmic phase of growth with at least 90% viability before passaging.
3. Incubate cultures at 37° C and 5% CO₂ until the viable cell density reaches 1 x 10⁶ cells/mL.
4. Subculture at 3 x 10⁵ cells/mL starting density into fresh medium prepared in a 2:1 ratio of original serum-supplemented medium to IS CHO medium.
5. Repeat steps 3 and 4 with sequential dilution ratios of 1:1, 1:2, 1:4 and 0:1 of the original serum-supplemented medium and IS CHO. If the cells look unhealthy or the growth rate declines significantly at a particular step of adaptation, maintain the cells for an additional passage in the media ratio of the previous step before subculturing into the next ratio.
6. Maintain cells in IS CHO for several passages, subculturing twice weekly to allow complete adaptation and assure optimum performance.

CRYOPRESERVATION

Viable cell banks may conveniently be created by freezing cells in 90% IS CHO + 10% DMSO. No other additions are necessary.

Freezing

1. Use cultures that are in logarithmic growth with high viabilities (> 85%).
2. Centrifuge cells for 5 minutes at 200 g.
3. Resuspend in cold (2-8° C) 90% IS CHO, 10% DMSO to a density of 1 x 10⁷ viable cells/mL.
4. Aliquot into sterile cryovials.
5. Gradually lower the temperature of the vials to below -80° C at a rate of -1° C/minute.
6. Store vials in liquid nitrogen freezer.

Thawing

1. Thaw frozen vial rapidly in a 37° C water bath.
2. Transfer the cell suspension to a culture flask with fresh IS CHO medium to achieve an initial cell density of 3 x 10⁵ viable cells/mL.
3. Incubate cultures at 37° C and 5% CO₂ until the viable cell density reaches 1 x 10⁶ cells/mL.
4. Subculture into fresh IS CHO medium at 2 X 10⁵ cells/mL starting density.

QUALITY ASSURANCE

All testing results are reported on a lot specific Certificate of Analysis, which is available upon request.

IS CHO™ is a trademark of Irvine Scientific.
