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ProductInformation

HYBRIDOMA STARTER PACK

Product Number **H 4387**
Storage Temperature –0 °C

Product Description

The hybridoma starter pack is designed to give the researcher an opportunity to sample the reagents required for hybridoma production in a small pack size prior to committing to the purchase of larger packs of the individual HAT, HT, aminopterin, and 8-azaguanine components. PEG may also be purchased separately in 5 gram quantities or in solution.

The hybridoma starter pack includes two vials each of:

- H 0262, Hypoxanthine-Aminopterin-Thymidine [HAT] 50X; when reconstituted to 10 ml each vial contains 5 x 10^{-3} M hypoxanthine, 2 x 10^{-5} M aminopterin, and 8 x 10^{-4} M thymidine.
- H 0137, Hypoxanthine-Thymidine [HT] 50X; when reconstituted to 10 ml each vial contains 5 x 10^{-3} M hypoxanthine and 8 x 10^{-4} M thymidine.
- A 5159, Aminopterin 50X; when reconstituted to 10 ml each vial contains 2 x 10^{-5} M aminopterin.
- A 5284, 8-Azaguanine 50X; when reconstituted to 10 ml each vial contains 6.6 x 10^{-3} M 8-azaguanine.
- P 2906, Polyethylene Glycol [PEG], MW 3000-3700; 2 grams solid PEG.

HAT, HT, Aminopterin, and 8-Azaguanine are supplied as lyophilized, γ-irradiated powders. PEG is offered as an autoclaved, sterile solid.

Precautions and Disclaimer

For laboratory use only. Not for drug, household, or other uses.

Preparation Instructions

To reconstitute, add 10 ml of sterile tissue culture medium to the lyophilized products and rotate gently until dissolved.

When 10 ml of the 50X concentrates of HAT, HT, aminopterin, and 8-azaguanine are diluted to 500 ml in tissue culture medium, the final working concentrations of hypoxanthine, aminopterin, thymidine, and 8-azaguanine are 100 μM, 0.4 μM, 16 μM, and 0.13 μM respectively. The 50X concentrate may be added directly to sterile tissue culture medium if proper aseptic procedure is followed during its reconstitution. Melt the PEG by autoclaving at 121 °C for 5 minutes. Dilute to 30-50% using sterile, pre-warmed serum-free medium (e.g. for a 50% solution add 2 ml to the 2 g vial of PEG). The pH of the solution should be slightly alkaline (pink, not orange, or purple). Adjustment of the pH with sterile NaOH may be required.

Storage/Stability

Prior to reconstitution, store lyophilized products at –0 °C. PEG may be stored at room temperature. After reconstitution, store all products at 2-8 °C for 1-2 weeks. Prolonged storage of reconstituted products is NOT recommended.

Product Profile

HAT and Aminopterin have been tested in cell culture in the medium supplemented with 10% Fetal Bovine Serum. In the presence of either 1X HAT or 1X aminopterin, the growth of HPRTase negative cells was inhibited. Under the same culture conditions, the growth of HPRTase positive cells was not affected.

References

1. Jakoby, W.B. and Pastan, I.H. (1979). Methods in Enzymology, 58, 345.
2. Kennett, R.H., McKearn, T.J. and Bechtol, K.B. (1980). Monoclonal Antibodies, Hybridomas: A New Dimension in Biological Analysis, [Plenum Press, New York], 365.
3. Mishell, B.B. and Shiigi, S.M. (1980). Selected Methods in Cellular Immunology, [H.W. Freeman and Co., San Francisco], 337.

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