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TECHNICAL INFORMATION

Catalog Number: 1680849
HAT (50X Solution)

Description: This media is selective for growth of cells with a functioning HGPRT mechanism. The main application is for the selective growth of hybridoma cells for monoclonal antibody production. Aminopterin blocks the synthesis of DNA by inhibiting dihydrofolate reductase.³ Cells that lack the ability to utilize the salvage pathway for nucleotide synthesis are eliminated. Cells that possess hypoxanthine-guanine phosphoribosyl transferase (HPRTase) and thymidine kinase (TK) enzymes can utilize the salvage pathway if supplied with hypoxanthine and thymidine.^{1,2} The purpose of this media is two-fold:

- Selectively kill unfused myeloma cells that are well adapted to tissue culture and would otherwise outgrow any hybridomas produced.
- Eliminate any myeloma-myeloma hybridomas that lack HPRTase. HPRTase positive spleen-spleen hybridomas, although not sensitive to aminopterin, are normally short-lived in culture.

After the selection is complete (approximately 10-14 weeks), aminopterin is diluted from the culture by several passages of the cells in hypoxanthine-thymidine (HT) supplemented medium (approximately 2-3 weeks) before transfer into normal hybridoma growth medium.

References:

- Freshney, R.I., *Culture of Animal Cells: A Manual of Basic Technique*, 3rd Ed., John Wiley and Sons, Inc., pp. 389-391 (1994).
- Harlow, E. and Lane, D., *Antibodies: A Laboratory Manual*, Cold Spring Harbor Laboratory, p. 280 (1988).
- Kennet, R.H., *Meth. Enzymol.*, W.B. Jakoby and I.H. Pastan (eds.), Academic Press, v. LVII, p. 352 (1979).
- Littlefield, J.W., *Science*, v. **145**, 709 (1964).

Formulation (as 50X):

Components	mg/L	Mol. Wt.	Mol. (mM)
Other			
Aminopterin 2H ₂ O	9.53000	476.4	0.02
Hypoxanthine, Na	910.70	180.08	5.06
Thymidine	193.80000	242.2	0.80

Availability:

HT:

Catalog Number	Description	Size
1680949	50X HT concentrate	100 ml