



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.<sup>3</sup> 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.<sup>1,2</sup>

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.

### Formulation (as 50X):

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

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