

ProductInformation

Preparation of Auto-Mod[™] Powdered Media Product Nos. D 5280, M 0769, and R 7755

Sigma's AUTO-MOD™ powdered media are specially formulated to withstand the temperatures and conditions required for autoclaving. The same recommendations as with all powdered media for protection against atmospheric moisture and the preparation of concentrated media apply to AUTO-MOD™ media.

PROCEDURE

Medium supplements can be added on the basis of their thermostability, i.e., heat stable supplements can be added prior to autoclaving and heat labile supplements must be added aseptically after autoclaving. The storage conditions and shelf life of the supplemented medium will be determined by the type of supplement added.

- Measure out 90% of final volume of tissue culture grade water.
- While gently stirring the water, add the powdered AUTO-MOD™ medium. Stir until dissolved.
- Rinse original package with a small amount of water to remove all traces of powder. Add to solution.
- The pH for RPMI-1640, (Product No. R 7755), should be adjusted to 4.0 before autoclaving.
- Add final volume of tissue culture grade water. Since sodium bicarbonate and L-glutamine solutions will be added after autoclaving, subtract these volumes from the final volume before adding to medium.
- Autoclave medium at 121°C (250°F) at 15 psi for 15 minutes. Note: Autoclaves vary in performance. Validation of each system is recommended.
- Medium should be promptly removed from the autoclave to avoid extended heating and evaporation.
- After cooling medium to 15-20°C, add required amounts of sterile Sodium Bicarbonate Solution, 7.5% w/v (Product No. S 8761) and of L-Glutamine, using either a sterile 200 mM solution (Product No. G 7513) or γ-irradiated L-glutamine (Product No. G 6392) for each liter of final volume of medium being prepared. See the following table for appropriate amounts.
- If necessary, adjust the pH using sterile 1 N NaOH or 1 N HCl.
- Store liquid medium refrigerated at 2-8°C and in the dark.

SODIUM BICARBONATE AND L-GLUTAMINE ADDITION TABLE

Recommended sodium bicarbonate and L-glutamine concentrations for 1X (single strength) AUTO-MOD™ powdered media using 7.5% Sodium Bicarbonate (NaHCO₃) Solution, (Product No. S 8761), and 200 mM sterile L-Glutamine Solution, (Product No. G 7513):

Auto-Mod™ Medium	Product Number	NaHCO₃ 7.5% Solution (ml/L)	L-Glutamine 200 mM (ml/L)	L-Glutamine γ-Irradiated (g/L)					
					DME	D 5280	49.3	20.0	0.584
					MEM	M 0769	29.3	10.0	0.292
RPMI-1640	R 7755	26.7	10.25	0.3					