

Product Information

Catalog. No.	Product Name	Format	Stock Conc.	Storage on Arrival	Thawing Instructions	Storage Once Thawed
ax3701	ARE Hepatocytes	≥5 million cells/vial	N/A	Dry Ice	Follow protocol	Liquid nitrogen
ax3702	ARE Hepatocytes, CYP2D6 Overexpressing	≥5 million cells/vial	N/A	Dry Ice	Follow protocol	Liquid nitrogen
ax3705	ARE Hepatocyte Thawing Medium	50 mL	N/A	Blue Ice	N/A	4°C for 3 months
ax3710	ARE Hepatocyte Maintenance Medium + Supplement	500 mL	N/A	Medium: Blue Ice Supplements: Dry Ice	N/A	4°C for 3 months; For long-term storage, store Medium at +4°C and Supplements at -80°C

Recommendations

- Always count the number of viable cells after thawing
- Recommended culture vessel coating: Type I Collagen
- Recommended cell culture media: ARE Hepatocyte Thawing Medium & ARE Hepatocyte Maintenance Medium
- Recommended seeding density: 20,000 30,000 viable cells/cm2
- Recommended centrifugation speed: 90 x g for 5 min
- Recommended days in culture before assay: 3 days



Preparation of the Reagent

Hepatocyte Thawing Medium

This is ready-to-use for thawing Axol Hepatocytes. No additional supplements are required.

Hepatocyte High Performance Medium

The Hepatocyte High Performance Medium is designed for the optimal culture and endpoint measurement of Hepatocytes. In order to obtain Hepatocyte High Performance Medium, add the entire contents of supplement A (5 mL) and L-Glutamine (5 mL) provided, to the basal medium. Due to the manufacturing process the medium may appear opaque but this does not affect the performance of the cells.

Thawing & Plating

- Coat the culture vessels with Type I Collagen or use pre-coated culture vessels.
- Dispense 50 mL of ARE Hepatocyte Thawing Medium into a 50 mL sterile conical tube and warm the medium to 37°C.
- Thaw the cells quickly in a 37°C water bath until just prior to complete thawing. Do not shake the vial when thawing, as vigorous movements will damage the cells.
- Wipe the outside of the vial with 70% ethanol.
- Gently resuspend the cells and slowly add to 50 mL of pre-warmed Thawing Medium.
- Rinse the cryovial twice with 1 mL of Thawing Medium to ensure all the cells are transferred.
- Centrifuge the cells at 90 x g at room temperature for 5 min.
- Carefully remove the supernatant and gently resuspend in 2 mL of pre-warmed ARE Hepatocyte Maintenance Medium and perform a cell count.
- Dilute the cells into the required volume of pre-warmed Maintenance Medium.
- Seed cells into the culture vessel (coated with Type I Collagen) at the recommended seeding density.
- Once the cells have attached (after 24 h), replace the Maintenance Medium.
- Optional Step: a Matrigel overlay can be added to maintain ARE Hepatocytes for at least 18 days after plating.
- Frequency of media changes: Every 2 3 days.
- We recommend the cells be cultured for 3 days in Maintenance Medium before proceeding with endpoint assays. The cells should reach confluency within 5 days after plating.
- ARE Hepatocytes can be maintained for at least 7 days after plating.

Assay-Ready-Expanded-Hepatocytes



Passaging

ARE Hepatocytes can be plated at subconfluency (50% confluency; 10,000-15,000 viable cells /cm²) in Maintenance Medium and the cells will proliferate to confluency.

We do not recommend passaging ARE Hepatocytes as they are ready for use. Further passaging will lead to de-differentiation of the hepatocytes and loss of biological functionality.

Usage Statement

Our products are intended for research use only and are not to be used for any other purpose, which includes but is not limited to, unauthorized commercial uses, in vitro diagnostic uses, ex vivo or in vivo therapeutic uses or any type of consumption or application to humans. The cells should not be expanded further or re-frozen.

Assay-Ready-Expanded-Hepatocytes



Got any questions? Need help with the protocol?

Contact Axol Technical Support at support@axolbio.com

Or

call +44 (0) 1223 751051