

Human Melanocytes

Catalog No.	Product Name	Product quantity	Short-term Storage	Long-term Storage	Thawing Instructions
ax3529	Melanocytes (Single	500,000	Liquid nitrogen	Liquid nitrogen	See below
	Donor, Juvenile)	cells/vial			
ax3530	Melanocytes (Single	500,000	Liquid nitrogen	Liquid nitrogen	See below
	Donor, Adult)	cells/vial			
ax3531	Melanocyte Growth	500 mL	4°C for 6	-20°C for 6	Thaw at 4°C or
	Medium		weeks	months	RT
ax3532	Melanocyte	250 mL	4°C for 6	-20°C for 6	Thaw at 4°C or
	Differentiation		weeks	months	RT
	Medium				
ax3542	Melanocyte Assay	250 mL	4°C for 6	-20°C for 6	Thaw at 4°C or
	Medium		weeks	months	RT
ax0044	Axol Unlock	25 mL	Aliquot and	-80°C	Thaw at 4°C
			store at -80°C		

Lot-specific information such as donor details and passage number are stated in the Certificate of Analysis for each product.

Recommendations:

Always count the number of viable cells after thawing

Recommended culture vessel coating: Not required

Recommended cell culture medium: Axol Melanocyte Growth Medium for initial culture

Recommended seeding density: 4,000 <u>viable</u> cells/cm²

Recommended centrifugation speed: 200 x g for 5 min

Thawing & Plating:

- Thaw the cells quickly in a 37°C water bath until just prior to complete thawing.
- Wipe the outside of the vial with 70% ethanol.
- Gently resuspend the cells and take an aliquot to perform a cell count.
- Slowly dilute the cells into the required volume of pre-warmed **Melanocyte Growth Medium** (must be at least 10 mL so that the concentration of DMSO is less than 1%).
- Rinse the cryovial with 1 mL of pre-warmed Melanocyte Growth Medium to ensure all of the cells are transferred.
- Seed cells into the culture vessel at the recommended seeding density.
- Once the cells have attached (after 6-24 h), replace the culture medium with fresh, prewarmed Melanocyte Growth Medium.
- Frequency of media changes: Every 2 days

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Passaging:

Passage when the culture reaches: 80% confluent
Recommended passaging reagent: Axol Unlock

• When the cells have detached from the culture vessel, dilute out the passaging reagent with pre-warmed **Melanocyte Growth Medium** and centrifuge the cells at 200 x g for 5 min.

It is important that the cells are centrifuged in order to remove the passaging reagent before plating the melanocytes.

- Remove the supernatant and resuspend in 1-2 mL of pre-warmed Melanocyte Growth Medium.
- Perform a cell count to determine the number of viable cells.
- Dilute the cells into the required volume of pre-warmed **Melanocyte Growth Medium**.
- Seed cells into the culture vessel at the recommended seeding density.

Endpoint Assays:

- Our human Melanocyte Assay Medium has a lower concentration of growth factors and so reduces proliferation of the melanocytes. In the absence of growth factor stimuli, the melanocytes are more responsive to experimental conditions.
- Melanocytes should be cultured initially in Melanocyte Growth Medium until 95% confluent.
- Replace the culture medium with **Melanocyte Assay Medium**. The melanocytes can be maintained in **Assay Medium** for up to 1 week.

Differentiation:

- Our human Melanocyte Differentiation Medium has been optimized to promote melanocyte differentiation and increased melanin production.
- Melanocytes should be cultured initially in Melanocyte Growth Medium until 95% confluent.
- Replace the culture medium with **Melanocyte Differentiation Medium**. The melanocytes will undergo differentiation and will be fully differentiated in 5 days.

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.

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