

KHOS-312H Cells | 300447**General information****Description**

KHOS-312H is a human osteosarcoma cell line derived from bone cancer. This cell line is part of a group of KHOS-derived osteosarcoma models, which includes KHOSNP and KHOS-240S, among others. Like other osteosarcoma cell lines, KHOS-312H is used extensively in cancer research to study the biology of osteosarcomas, particularly their genetic and molecular characteristics, and to evaluate potential therapeutic agents. The KHOS-312H cell line is known for its resistance to certain targeted kinase inhibitors, such as those affecting the PI3K-Akt-mTOR pathway, making it an essential model for studying drug resistance mechanisms in osteosarcoma.

One of the significant features of the KHOS-312H cell line is its utility in high-throughput screening for anticancer drugs. In large-scale screening studies, KHOS-312H has been tested against a wide array of compounds, including both FDA-approved drugs and investigational agents. These studies have revealed that KHOS-312H shows varying degrees of sensitivity and resistance to different classes of anticancer drugs, helping researchers to map the molecular landscape of osteosarcoma response to treatment. Notably, the cell line's resistance to mTOR inhibitors has been particularly highlighted, suggesting a potential need for combination therapies or novel agents to overcome this challenge.

Organism Human**Tissue** Bone**Disease** Osteosarcoma**Synonyms** KHOS-321H, KHOS312H, KHOS321H**Characteristics****Age** 13 years**Gender** Female**Ethnicity** Caucasian**Morphology** Fibroblast-like**Growth properties** Monolayer, adherent**Regulatory Data****Citation** KHOS-312H (Cytion catalog number 300447)

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Biosafety level	1
NCBI_TaxID	9606
CellosaurusAccession	CVCL_2545

Biomolecular Data

Tumorigenic	No
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Handling

Culture Medium	EMEM (MEM Eagle), w: 2 mM L-Glutamine, w: 2.2 g/L NaHCO ₃ , w: EBSS (Cytion article number 820100a)
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Supplements	Supplement the medium with 10% FBS and 1% NEAA
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Dissociation Reagent	Accutase
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Subculturing	Remove the old medium from the adherent cells and wash them with PBS that lacks calcium and magnesium. For T25 flasks, use 3-5 ml of PBS, and for T75 flasks, use 5-10 ml. Then, cover the cells completely with Accutase, using 1-2 ml for T25 flasks and 2.5 ml for T75 flasks. Let the cells incubate at room temperature for 8-10 minutes to detach them. After incubation, gently mix the cells with 10 ml of medium to resuspend them, then centrifuge at 300xg for 3 minutes. Discard the supernatant, resuspend the cells in fresh medium, and transfer them into new flasks that already contain fresh medium.
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Seeding density	1×10^4 cells/cm ²
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Fluid renewal	2 to 3 times per week
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Post-Thaw Recovery	After thawing, plate the cells at 5×10^4 cells/cm ² and allow the cells to recover from the freezing process and to adhere for at least 24 hours.
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Freeze medium	As a cryopreservation medium, we use complete growth medium (including FBS) + 10% DMSO for adequate post-thaw viability, or CM-1 (Cytion catalog number 800100), which includes optimized osmoprotectants and metabolic stabilizers to enhance recovery and reduce cryo-induced stress.
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Thawing and Culturing Cells

1. Confirm that the vial remains deeply frozen upon delivery, as cells are shipped on dry ice to maintain optimal temperatures during transit.
2. Upon receipt, either store the cryovial immediately at temperatures below -150°C to ensure the preservation of cellular integrity, or proceed to step 3 if immediate culturing is required.
3. For immediate culturing, swiftly thaw the vial by immersing it in a 37°C water bath with clean water and an antimicrobial agent, agitating gently for 40-60 seconds until a small ice clump remains.
4. Perform all subsequent steps under sterile conditions in a flow hood, disinfecting the cryovial with 70% ethanol before opening.
5. Carefully open the disinfected vial and transfer the cell suspension into a 15 ml centrifuge tube containing 8 ml of room-temperature culture medium, mixing gently.
6. Centrifuge the mixture at $300 \times g$ for 3 minutes to separate the cells and carefully discard the supernatant containing residual freezing medium.
7. Gently resuspend the cell pellet in 10 ml of fresh culture medium. For adherent cells, divide the suspension between two T25 culture flasks; for suspension cultures, transfer all the medium into one T25 flask to promote effective cell interaction and growth.
8. Adhere to established subculture protocols for continued growth and maintenance of the cell line, ensuring reliable experimental outcomes.

Incubation Atmosphere

37°C , 5% CO_2 , humidified atmosphere.

Shipping Conditions

Cryopreserved cell lines are shipped on dry ice in validated, insulated packaging with sufficient refrigerant to maintain approximately -78°C throughout transit. On receipt, inspect the container immediately and transfer vials without delay to appropriate storage.

Storage Conditions

For long-term preservation, place vials in vapor-phase liquid nitrogen at about -150 to -196°C . Storage at -80°C is acceptable only as a short interim step before transfer to liquid nitrogen.

Quality Control & Molecular Analysis

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Sterility

Mycoplasma contamination is excluded using both PCR-based assays and luminescence-based mycoplasma detection methods.

To ensure there is no bacterial, fungal, or yeast contamination, cell cultures are subjected to daily visual inspections.