

## ATDC5 Cells | 305427

### General information

#### Description

ATDC5 is a murine chondrogenic cell line derived from mouse teratocarcinoma cells and is widely used as an in vitro model for studying chondrogenesis and cartilage development. This cell line undergoes sequential chondrogenic differentiation, mimicking in vivo processes such as cellular condensation, the expression of early chondrocytic markers like type II collagen and aggrecan, and the transition to hypertrophic chondrocytes, marked by type X collagen expression and matrix mineralization. Due to its ability to proliferate and differentiate efficiently, ATDC5 serves as a valuable model for exploring molecular mechanisms related to skeletal development, especially endochondral ossification.

ATDC5 cells have been extensively used to study the influence of various growth factors, hormones, and transcription factors on chondrogenesis. For instance, transforming growth factor-beta (TGF- $\beta$ ) has been shown to promote early chondrogenic differentiation by modulating the expression of extracellular matrix components like fibronectin. Similarly, bone morphogenetic proteins (BMPs), particularly BMP-2, -4, and -7, play a critical role in promoting different stages of chondrocyte differentiation in ATDC5. Moreover, the activation of transient receptor potential vanilloid 4 (TRPV4) channels in these cells, combined with hyaluronan, has been demonstrated to enhance the expression of key chondrogenic markers such as SOX9 and Aggrecan, further supporting their utility in cartilage tissue engineering studies.

This cell line has been instrumental in proteomics research as well, showing that ATDC5 cells can synthesize major cartilage extracellular matrix (ECM) components like aggrecan and type II collagen, along with the proper post-translational modifications required for cartilage function. Its capacity to recapitulate crucial ECM biosynthesis events makes ATDC5 an indispensable model for studying cartilage formation and related pathologies.

#### Organism

Mouse

#### Tissue

Embryo

#### Disease

Teratocarcinoma

#### Metastatic site

Not applicable (derived from mouse embryonic teratocarcinoma; non-metastatic model)

#### Applications

Chondrogenesis research; cartilage development and endochondral ossification; chondrocyte differentiation (type II collagen, aggrecan, SOX9 expression); BMP-2/-4/-7 and TGF- $\beta$  signaling in chondrocytes; osteoarthritis modeling; cartilage tissue engineering; proteoglycan biosynthesis; TRPV4 channel biology in cartilage

#### Synonyms

ATDC-5

### Characteristics

#### Breed/Subspecies

129

#### Age

Embryo

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<b>Gender</b>	Male
<b>Morphology</b>	Polygonal
<b>Cell type</b>	Chondrocyte precursor cells
<b>Growth properties</b>	Adherent

## Regulatory Data

<b>Citation</b>	ATDC5 (Cytion catalog number 305427)
<b>Biosafety level</b>	1
<b>NCBI_TaxID</b>	10090
<b>CellosaurusAccession</b>	CVCL_0225
<b>GMO Status</b>	No genetic modification; wildtype murine teratocarcinoma-derived chondrogenic cell line

## Biomolecular Data

## Handling

<b>Culture Medium</b>	DMEM:Ham's F12 (1:1), w: 3.1 g/L Glucose, w: 2.5 mM L-Glutamine, w: 15 mM HEPES, w: 0.5 mM Sodium pyruvate, w: 1.2 g/L NaHCO <sub>3</sub> (Cytion article number 820400a)
<b>Supplements</b>	Supplement the medium with 5% FBS
<b>Dissociation Reagent</b>	Accutase

**Subculturing** For routine adherent cell culture: Aspirate the old culture medium from the adherent cells, and wash them with PBS to remove any remaining medium. After aspirating the PBS, add the appropriate volume of Accutase solution based on the culture vessel size (e.g., 1 ml for a T25 flask, 3 ml for a T75 flask) and incubate at room temperature or 37°C for 5-10 minutes, or until the cells detach. Monitor detachment under a microscope, and gently tap the vessel if necessary to release the cells. Once detached, add complete medium to inactivate the Accutase, gently resuspend the cells, and transfer an aliquot of the cell suspension into a new culture vessel containing fresh medium. Place the vessel in an incubator set to 37°C with 5% CO<sub>2</sub>, and change the medium every 2-3 days.

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**Seeding density**  $2 \times 10^4$  cells/cm<sup>2</sup>

**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.

### 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 x 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<sub>2</sub>, 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.

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### 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

### 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.