

## RY(ham) Yoshida Sarcoma Cells | 500415

<b>Description</b>	This cell line was established as in vitro cell line from a Yoshida Ascites sarcoma induced in Sprague-Dawley rats.
<b>Organism</b>	Rat
<b>Disease</b>	Sarcoma
<b>Metastatic site</b>	Ascites
<b>Cell type</b>	Fibroblast
<b>Growth properties</b>	Adherent
<b>Citation</b>	RY(ham) Yoshida Sarcoma (Cytion catalog number 500415)
<b>Biosafety level</b>	1
<b>NCBI_TaxID</b>	10116
<b>CellosaurusAccession</b>	CVCL_1D05
<b>Culture Medium</b>	RPMI 1640, w: 2.0 mM stable Glutamine, w: 2.0 g/L NaHCO <sub>3</sub> (Cytion article number 820700a)
<b>Supplements</b>	Supplement the medium with 10% FBS
<b>Dissociation Reagent</b>	Accutase

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**Subculturing** Gather the suspension cells in a 15 ml tube and gently wash the adherent cells with PBS lacking calcium and magnesium (use 3-5 ml for T25 flasks and 5-10 ml for T75 flasks). Apply Accutase (1-2 ml for T25 flasks, 2.5 ml for T75 flasks) ensuring full coverage of the cell layer. Allow the cells to incubate at room temperature for 10 minutes. Following incubation, combine and centrifuge both the suspension and adherent cells. After centrifugation, carefully resuspend the cell pellet and transfer the cell suspension into new flasks containing fresh medium.

**Seeding density** 1 to  $2 \times 10^4$  cells/cm<sup>2</sup>

**Post-Thaw Recovery** After thawing, the cells should be handled at a cell concentration of  $5 \times 10^5$  cells/ml and allowed to recover for at least 48 hours.

**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.

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### 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.

### 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.

### 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.