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Zebrafish Testicular Feeder Cells (ZtA6-12)

Cat.No.	**Unit**
T2611	1x10^6 cells / 1.0 ml

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Name	Zebrafish Testicular Feeder Cells (ZtA6-12)
Description	ZtA6 cells are derived from spontaneous tumor-like hypertropheied testis isolated from albino-type (alb-1/alb-1) zebrafish. Clones were analyzed for the expression of the Sertoli cell marker (Sox9a), the germ cell marker (Vas), and the Wilm's tumor suppressor marker (WT1). 12 clones were derived and have distinctive properties and are available at abm.
Organism	Fish
Tissue	Testes
Donor History	Zebrafish (D. rerio)
Growth Properties	Adherent, epithelial and fibroblast-like
Cell Type	Stable Cell Lines
Unit	1x10^6 cells / 1.0 ml
Storage Condition	Vapor phase of liquid nitrogen, or below -130°C.
Shipping Conditions	Ship with dry ice.
Product Format	Frozen
Intended Use	This product is intended for laboratory research use only. It is not intended for any animal or human therapeutic use, any human or animal consumption, or any diagnostic use.
BioSafety	II
Certificate of Analysis	For batch-specific test results, refer to the applicable certificate of analysis that can be found at www.abmgood.com.
Growth Conditions	**PriCoat™ T25 Flasks (G299) coated with Gelatin Coating Solution (0.1%) (TM063) are required for optimal cell adhesion and growth.** PriGrow XIII Medium (TM013) + 2 mM L-Glutamine (G275) + 100 µg/mL Kanamycin sulfate + 800 µM $CaCl_2$ + 200 µg/mL L-arginine +20 µg/mL aspartic acid + 15 µg/µl L-histidine- HCl + 72.5 µg/mL L-lysine-HCl + 20 µg/mL L-proline + 0.5% w/v Bovine serum albumin + 10mM HEPES + 10 IU/mL human chorionic gonadotropin + 10 IU/mL pregnant mare's serum gonadotropin + 3% FBS (TM999). Sterilize the media by filtration (0.2 µM filter), store at 4°C, and should be used within 2 weeks. May add 1% Penicillin/Streptomycin Solution (G255) if desired. Incubate cells at 38.0°C, **no CO_2. Use only Accutase and Gelatin coated dishes.**

	To prepare as feeder cells: Treat with 10 µg/ml mitomycin C in L-15 for 5 hours before plating for use as feeder cells for further experimentation.
Unpacking and Storage Instructions	1. Visually examine the packaging containers for signs of leakage or breakage. 2. Immediately transfer frozen cells from dry ice packaging to a temperature below -130°C, preferably in liquid nitrogen vapor phase storage, until ready for use. To ensure the highest level of viability, thaw the vial and initiate culture as soon as possible upon receipt. If continued storage is desired, the vial should only be stored below -130°C or in liquid nitrogen vapor phase. Do not store at -70°C, as it will result in loss of viability.
Thawing Protocol	1. Thaw cells quickly in a 37°C water bath while agitating gently (maximum 2 minutes). The vial cap should be kept above the water level to minimize the risk of contamination. 2. Decontaminate the vial by spraying and wiping the exterior of the vial with 70% ethanol. From this point onwards, all operations should be strictly carried out inside a biological safety cabinet using aseptic conditions. 3. Transfer the cell suspension into a 15ml sterile conical tube containing 5ml of pre-warmed, complete growth media. Centrifuge cells at 125xg for 5-7 minutes. 4. Aspirate the supernatant without disturbing the cell pellet. Re-suspend the cell pellet in the recommended pre-warmed, complete growth media and dispense into a T25 culture flask. 5. Incubate the cells at the recommended conditions.
Subculture Protocol	Volumes given below are for a T75 flask; proportionally increase or decrease the volume as required per culture vessel size. Subculture cells once the culture vessel is 80% confluent. 1. Aspirate the culture media, and add 2-3ml of pre-warmed Accutase to the culture vessel. 2. Observe the cells under a microscope to confirm detachment (typically within 2-10 minutes). Cells that are difficult to detach can be put in 37°C, for several minutes to facilitate detachment. 3. Neutralize Accutase by adding an equal volume of the complete growth media into the culture vessel. 4. Transfer the culture suspension into a sterile centrifuge tube, and centrifuge at 125xg for 5 minutes. The actual centrifuge duration and speed may vary depending on the cell type. 5. Aspirate the supernatant, and re-suspend the pellet with pre-warmed fresh complete growth media. Add appropriate aliquots of the cell suspension to new culture vessels, as desired. 6. Incubate the cells at the recommended conditions.
Cryopreservation	We recommend using serum-free CryoGuard™ Freezing Media (TM078) or, if serum is preferred, Cryopreservation Medium (TM024).
Material Citation	If use of this material results in a scientific publication, please cite the material in the following manner: Applied Biological Materials Inc, Cat. No. T2611.
Warranty	**abm** warrants that cell lines shall be viable upon initiation of culture for a period of thirty (30) days after shipment and that they shall meet the specifications on the applicable **abm** Material Product Information sheet, certificate of analysis, and/or catalog description. Such thirty (30) day period is referred to herein as the "Warranty Period".

Disclaimer	1. Sale of this item is subjected to the completion of a Material Transfer Agreement (MTA) by the purchasing individual/institution for each order. If you have any questions regarding this, please contact us at licensing@abmgood.com. 2. All test parameters provided in the CoA are conducted using **abm**'s standardized culture system and procedures. The stated values may vary under the end-user's culture conditions. Please verify that the product is suitable for your studies by referencing published papers or ordering RNA (0.5 µg, Cat.# C207, $450.00) or cell lysate (100 µg, Cat.# C206, $600.00) to perform preliminary experiments, or alternatively use our Gene Expression Assay Service (Cat# C138). All sales are final. 3. We recommend live cell shipments for ease of cell transfer and this option can be requested at the time of ordering. Please note that the end-user will need to evaluate the feasibility of live cell shipment by taking into account the final destination's temperature variation and its geographical location. In addition, we thoroughly test our cell lines for freeze-thaw recovery. If frozen cells were received and not recovered in your lab under the exact, specified conditions (using recommended culture vessel, media, additional supplements, and atmospheric conditions), a live cell replacement is possible at a cost (plus shipping). 4. All of **abm**'s cell biology products are for research use ONLY and NOT for therapeutic/diagnostic applications. **abm** is not liable for any repercussions arising from the use of its cell biology product(s) in therapeutic/diagnostic application(s). Please contact a technical service representative for more information. 5. **abm** makes no warranties or representations as to the accuracy of the information on this site. Citations from literature and provided for informational purposes only. **abm** does not warrant that such information has been shown to be accurate. 6. **abm** warrants that cell lines shall be viable upon initiation of culture for a period of thirty (30) days after shipment and that they shall meet the specifications on the applicable **abm** Material Product Information sheet, certificate of analysis, and/or catalog description. Such thirty (30) day period is referred to herein as the "Warranty Period."
Depositor	National Institute of Genetics (ROIS/NIG)
QC	1) Phagocytic activity was analyzed via the ability to internalize polystyrene beads; 2) RT-PCR was used to assess the presence or absence of WT1, Vas, and Sox9a markers; 3) Functionality test was performed to determine the ability to support male germ cells when co-cultured as feeder cells.
Application	Research Use Only.