THE UNIVERSITY OF WESTERN ONTARIO
BIOHAZARDOUS AGENTS REGISTRY FORM
Approved Biohazards Subcommittee: March 27, 2009
Biosafety Website: www.uwo.ca/humanresources/biosafety/

This form must be completed by each Principal Investigator holding a grant administered by the University of Western Ontario or in charge of a laboratory/facility where the use of Level 1, 2 or 3 biohazardous agents is described in the laboratory or animal work proposed. The form must also be completed if any work is proposed involving animals carrying zoonotic agents infectious to humans or involving plants, fungi, or insects that require Public Health Agency of Canada (PHAC) or Canadian Food Inspection Agency (CFIA) permits.

This form must ~~also~~ be updated at least every 3 years or when there are changes to the biohazards being used.

Containment Levels will be established in accordance with Laboratory Biosafety Guidelines, 3rd edition, 2004, Public Health Agency of Canada (PHAC) or Containment Standards for Veterinary Facilities, 1st edition 1996, Canadian Food Inspection Agency (CFIA).

Completed forms are to be returned to Occupational Health and Safety, (OHS), (Support Services Building, Room 4190) for distribution to the Biohazard Subcommittee. For questions regarding this form, please contact the Biosafety Officer at extension 81135 or biosafety@uwo.ca. If there are changes to the information on this form (excluding grant title and funding agencies), contact Occupational Health and Safety for a modification form. See website: www.uwo.ca/humanresources/biosafety

PRINCIPAL INVESTIGATOR: FRANÇOIS LAGUGNÉ-LABARTHET
SIGNATURE:
DEPARTMENT: chemistry
ADDRESS: 1151 Richmond Street
PHONE NUMBER: 519 661 2111 ext 81006
EMERGENCY PHONE NUMBER(S): 519 471 6956
EMAIL: flagugne@uwo.ca

Location of experimental work to be carried out: Building(s) chemistry Room(s) 124

*For work being performed at Institutions affiliated with the University of Western Ontario, the Safety Officer for the Institution where experiments will take place must sign the form prior to its being sent to the University of Western Ontario Biosafety Officer (See Section 12.0, Approvals).

FUNDING AGENCY/AGENCIES: CRC : Canada Research Chair
GRANT TITLE(S): CRC tier 2 in Photonics and Nanoscale Ionics

PLEASE ATTACH A BRIEF DESCRIPTION OF YOUR WORK THAT EXPLAINS THE BIOHAZARDS USED AND HOW THEY WILL BE USED. PROJECTS SUBMITTED WITHOUT A SUMMARY WILL NOT BE REVIEWED.

Names of all personnel working under Principal Investigators supervision in this location:

Christine Séguin + Amanda Leclaire	(PI: LAGUGNÉ)
Jessica McLachlan	(PI: NORTON)
Zhag Mengni	(PI: DING)
Collin BONDUELLE	(PI: GILLIES)

*** DESCRIPTION MUST BE ATTACHED TO THIS FORM OR PROJECT WILL NOT BE REVIEWED***

1.0 Microorganisms

1.1 Does your work involve the use of microorganisms or biological agents of plant or animal origin (including but not limited to viruses, prions, parasites, bacteria)? O YES ⊘NO
If no, please proceed to Section 2.0

Do you use microorganisms that require a permit from the CFIA? O YES O NO
If YES, please give the name of the species. _____
What is the origin of the microorganism(s)? _____
Please describe the risk (if any) of escape and how this will be mitigated:

_____
_____
_____

Please attach the CFIA permit.
Please describe any CFIA permit conditions:

_____
_____
_____

1.2 Please complete the table below:

Name of Biological agent(s)*	Is it known to be a human pathogen? YES/NO	Is it known to be an animal pathogen? YES/NO	Is it known to be a zoonotic agent? YES/NO	Maximum quantity to be cultured at one time? (in Litres)	Source/ Supplier	PHAC or CFIA Containment Level
	O Yes O No	O Yes O No	O Yes O No			O 1 O 2 O 3
	O Yes O No	O Yes O No	O Yes O No			O 1 O 2 O 3
	O Yes O No	O Yes O No	O Yes O No			O 1 O 2 O 3
	O Yes O No	O Yes O No	O Yes O No			O 1 O 2 O 3

*Please attach a Material Safety Data Sheet or equivalent from the supplier.

2.0 Cell Culture

2.1 Does your work involve the use of cell cultures? ⊘YES O NO
If no, please proceed to Section 3.0

2.2 Please indicate the type of primary cells (i.e. derived from fresh tissue) that will be grown in culture in the table below

Cell Type	Is this cell type used in your work?	Source of Primary Cell Culture Tissue	AUS Protocol Number
Human	O Yes ⊘No		Not applicable
Rodent	O Yes ⊘No		
Non-human primate	O Yes ⊘No		
Other (specify)	O Yes ⊘No		

2.3 Please indicate the type of established cells that will be grown in culture in the table below.

Cell Type	Is this cell type used in your work?	Specific cell line(s)*	Supplier / Source
Human	☑ Yes O No	T24, A431	CEDARLANE LABORATORIES
Rodent	☑ Yes O No	CRCIZ	CEDARLANE LABORATORIES
Non-human primate	O Yes ☑ No		
Other (specify)	O Yes ☑ No		

*Please attach a Material Safety Data Sheet or equivalent from the supplier. (For more information, see www.atcc.org)

2.4 For above named cell types(s) indicate PHAC or CFIA containment level required ☑ 1 O 2 O 3

3.0 Use of Human Source Materials

3.1 Does your work involve the use of human source materials? O YES ☑ NO
If no, please proceed to Section 4.0

3.2 Indicate in the table below the Human Source Material to be used.

Human Source Material	**Source/Supplier /Company Name**	**Is Human Source Material Known to Be Infected With An Infectious Agent? YES/NO**	**Name of Infectious Agent (If applicable)**	**PHAC or CFIA Containment Level (Select one)**
Human Blood (whole) or other Body Fluid		O Yes O No		O 1 O 2 O 3
Human Blood (fraction) or other Body Fluid		O Yes O No		O 1 O 2 O 3
Human Organs or Tissues (unpreserved)		O Yes O No		O 1 O 2 O 3
Human Organs or Tissues (preserved)		O Yes O No		O 1 O 2 O 3

4.0 Genetically Modified Organisms and Cell lines

4.1 Will genetic modifications be made to the microorganisms, biological agents, or cells described in Sections 1.0 and 2.0? O YES ☑ NO If no, please proceed to Section 5.0

4.2 Will genetic modification(s) involving plasmids be done? O YES, complete table below O NO

Bacteria Used for Cloning *	Plasmid(s) *	Source of Plasmid	Gene Transfected	Describe the change that results

* Please attach a Material Data Sheet or equivalent if available.

4.3 Will genetic modification(s) involving viral vectors be done? O YES, complete table below O NO

Virus Used for Transduction *	Vector(s) *	Source of Vector	Gene Transfected	Describe the change that results

* Please attach a Material Safety Data Sheet or equivalent.

4.4 Will genetic sequences from the following be involved?

- HIV O YES, please specify _____ O NO
- HTLV 1 or 2 or genes from any Level 1 or Level 2 pathogens O YES, specify _____ O NO
- SV 40 Large T antigen O YES O NO
- E1A oncogene O YES O NO
- Known oncogenes O YES, please specify _____ O NO
- Other human or animal pathogen and or their toxins O YES, please specify _____ O NO

4.5 Will virus be replication defective? O YES O NO

4.6 Will virus be infectious to humans or animals? O YES O NO

4.7 Will this be expected to increase the containment level required? O YES O NO

5.0 Human Gene Therapy Trials

5.1 Will human clinical trials be conducted using the viral vector in 4.0? O YES O NO
If no, please proceed to Section 6.0 If YES attach a full description of the make-up of the virus.

5.2 Will virus be able to replicate in the host? O YES O NO

5.3 How will the virus be administered? _____

5.4 Please give the Health Care Facility where the clinical trial will be conducted:_____

5.5 Has human ethics approval been obtained? O YES, number: _____ O NO O PENDING

6.0 Animal Experiments

6.1 Will live animals be used? O YES O NO If no, please proceed to section 7.0

6.2 Name of animal species to be used_____

6.3 AUS protocol # _____

6.4 Will any of the agents listed be used in live animals O YES, specify: _____ O NO

*** DESCRIPTION MUST BE ATTACHED TO THIS FORM OR PROJECT WILL NOT BE REVIEWED***

7.0 Use of Animal species with Zoonotic Hazards

7.1 Will any of the following animals or their organs, tissues, lavages or other body fluids including blood be used?

- Pound source dogs O YES ~~O NO~~
- Pound source cats O YES ~~O NO~~
- Cattle, sheep or goats O YES ~~O NO~~
- Non-human primates O YES, please specify species _____ ~~O NO~~
- Wild caught animals O YES, please specify species & colony # _____ ~~O NO~~
- Birds O YES ~~O NO~~
- Others (wild or domestic) O YES, please specify _____ ~~O NO~~

8.0 Biological Toxins

8.1 Will toxins of biological origin be used? ✓ YES ~~O NO~~ If no, please proceed to Section 9.0

8.2 If YES, please name the toxin(s) Phalloidin
Please attach information, such as a Material Safety Data Sheet, for the toxin(s) used.

8.3 What is the LD_{50} (specify species) of the toxin 2 mg/kg (mouse)

8.4 How much of the toxin is handled at one time*? 5 mL of ~~0.022 mM solution~~ 6.6 mM solution. 7.8 mg/L

8.5 How much of the toxin is stored*? 3 mL of 6.6 mM solution. (7.8 mg/L)

*For information on biosecurity requirements, please see:
http://www.uwo.ca/humanresources/docandform/docs/healthandsafety/biosafety/Biosecurity_Requirements.pdf

9.0 Insects Requiring CFIA Permits

9.1 Do you use insects that require a permit from the CFIA? O YES ~~O NO~~
If no, please proceed to Section 10.0

9.2 If YES, please give the name of the species. _____

9.3 What is the origin of the insect? _____

9.4 What is the lifestage of the insect? _____

9.5 What is your intention? O Initiate and maintain colony, give location: _____
O "One-off" use, give location: _____

9.6 Please describe the risk (if any) of escape and how this will be mitigated:
_____
_____
_____

9.7 Please attach the CFIA permit.

9.8 Please describe any CFIA permit conditions:
_____
_____
_____

*** DESCRIPTION MUST BE ATTACHED TO THIS FORM OR PROJECT WILL NOT BE REVIEWED***

10.0 Plants Requiring CFIA Permits

10.1 Do you use plants that require a permit from the CFIA? O YES ⊘ NO
If no, please proceed to Section 11.0

10.2 If YES, please give the name of the species. _____

10.3 What is the origin of the plant? _____

10.4 What is the form of the plant (seed, seedling, plant, tree...)? _____

10.5 What is your intention? O Grow and maintain a crop O "One-time" use

10.6 Do you do any modifications to the plant? O YES O NO
If yes, please describe: _____
_____

10.7 Please describe the risk (if any) of loss of the material from the lab and how this will be mitigated:
_____
_____
_____

10.8 Is the CFIA permit attached? O YES O NO

10.9 Please describe any CFIA permit conditions:
_____
_____
_____

11.0 Import Requirements

11.1 Will any of the above agents be imported? O YES, please give country of origin_____
If no, please proceed to Section 10.0 ⊘ NO

11.2 Has an Import Permit been obtained from HC for human pathogens? O YES O NO

11.3 Has an import permit been obtained from CFIA for animal or plant pathogens? O YES O NO

11.4 Has the import permit been sent to OHS? O YES, please provide permit # _____ O NO

12.0 Training Requirements for Personnel Named on Form

All personnel named on the above form who will be using any of the above named agents are required to attend the following training courses given by OHS:

- Biosafety
- Laboratory and Environmental/Waste Management Safety
- WHMIS (Western or equivalent)
- Employee Health and Safety Orientation

As the Principal Investigator, I have ensured that all of the personnel named on the form who will be using any of the biohazardous agents in Sections 1.0 to 9.0 have been trained.

SIGNATURE _____

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13.0 Containment Levels

11.1 For the work described in sections 1.0 to 9.0, please indicate the highest HC or CFIA Containment Level required. ☑ 1 ○ 2 ○ 3

13.2 Has the facility been certified by OHS for this level of containment?

○ YES, permit # if on-campus_____

○ NO, please certify

☑ NOT REQUIRED for Level 1 containment

14.0 Procedures to be Followed

14.1 As the Principal Investigator, I will ensure that this project will follow the Western Biosafety Guidelines and Procedures Manual for Containment Level 1 & 2 Laboratories (and the Level 3 Facilities Manual for Level 3 projects). I will ensure that UWO faculty, staff and students working in my laboratory have an up-to-date Hazard Communication Form, found at http://www.wph.uwo.ca/

SIGNATURE _____ Date: 17/04/2009

15.0 Approvals

UWO Biohazard Subcommittee: SIGNATURE: _____

Date: _____

Safety Officer for Institution where experiments will take place: SIGNATURE: _____

Date: _____

Safety Officer for University of Western Ontario (if different from above): SIGNATURE: _____

Date: _____

Approval Number: _____ Expiry Date (3 years from Approval): _____

Special Conditions of Approval:

*** DESCRIPTION MUST BE ATTACHED TO THIS FORM OR PROJECT WILL NOT BE REVIEWED***

Material Safety Data Sheet

Revision Number: 1.2
Revision Date: 31-Jul-2006

Product and Company Identification

Product Name: fluorescein phalloidin
Catalog Number: F432
Unit Size: 300 U
Manufacturer/Supplier: Molecular Probes, Inc.
29851 Willow Creek Road, Eugene, OR 97402-9132, USA
For US and Canada, Toll-Free Phone: 1-800-438-2209 · Fax: 1-800-438-0228
Phone: (541)465-8300 · Fax: (541)335-0305 · Web: http://probes.invitrogen.com
Technical Assistance: (541)335-0353 · E-mail: probestech@invitrogen.com

Composition / Information on Ingredients

- fluorescein phalloidin

Molecular Formula: $C_{56}H_{60}N_{10}O_{15}S_2$
Molecular Weight: 1177.27
CAS Number/Name: 84313-38-2 / Phalloidin, 7-[3-[4-[[[[2-(3',6'-dihydroxyspiro[isobenzofuran-1(3H),9'-[9H]xanthen]-3-yl)hydrazino]thioxomethyl]amino] methyl]-2-methyl-1,3-dithiolan-2-yl]-L-alanine]-

Hazards Identification

Emergency Overview: Toxic in contact with skin. Harmful if swallowed. Harmful by inhalation.

Potential Health Effects

Inhalation: May be harmful if inhaled.
Ingestion: Harmful if swallowed.
Skin: Toxic in contact with skin.
Eyes: Risk of serious damage to eyes.
Chronic Exposures: See Emergency Overview above.
Target Organs: Liver. Kidney. Central nervous system (CNS). Blood. Respiratory system.

First Aid Measures

Potentially harmful. Avoid prolonged or repeated exposure. Wash thoroughly after handling. If eye or skin contact occurs, wash affected area with water for 15 minutes and seek medical advice. If inhaled, move individual to fresh air and seek medical advice. If swallowed, seek medical advice.

Fire Fighting Measures

Use dry chemical powder or appropriate foam extinguisher.

Accidental Release Measures

Use appropriate protective equipment and methods to clean up spilled substances promptly. Absorb spill onto an appropriate material. Collect and dispose of all waste in accordance with applicable laws.

Handling and Storage

Store at <= -20°C. Protect material from long-term exposure to light; may be exposed to light for short periods of time.

Exposure Controls / Personal Protection

Wear appropriate gloves, protective clothing and eyewear and follow safe laboratory practices.
ACGIH/OSHA Permissible Exposure Limit Data: Not determined

Physical and Chemical Properties

Form:	Solid
Odor:	Not determined
Solubility in Water:	Moderate
Specific Gravity:	Not determined
pH:	Not determined
Boiling Point:	Not determined
Melting Point:	Not determined
Flash Point:	Not determined
Vapor Pressure:	Not determined

Stability and Reactivity

Thermal Decomposition: No decomposition if used according to specifications.
Dangerous Reactions: No dangerous reactions identified.
Dangerous Products of Decomposition: No dangerous decomposition products identified.

Toxicological Information

RTECS Number:	None known
Toxicity:	We are not aware of any toxicity data for this product.
Health Hazards:	See Potential Hazards below.
Potential Hazards:	This material is a phalloidin derivative and as such is potentially toxic. Phalloidin has toxicity data: LD50: 2 mg/kg, intraperitoneal, mouse.
Carcinogenicity:	Not listed by NTP, IARC or OSHA.

Ecological Information

Do not allow product to reach ground water, water course, or sewage system.

Disposal Considerations

Consult local, state or national regulations for proper disposal.

Transport Information

Hazard Class: 6.1
Identification Number: UN3287
Packing Group: II
Proper Shipping Name (Technical Name): Toxic solid, organic, n.o.s. (phalloidin)

Regulations

US Toxic Substances Control Act (TSCA): Not listed
US Other: Not applicable
EEC EINECS Number: Not identified
EEC Risk Statements: None identified
Other Country Regulations: R-phrase(s) R24 - Toxic in contact with skin R20 - Harmful by inhalation R22 - Harmful if swallowed S-phrase(s) S45 - In case of accident or if you feel unwell, seek medical advice immediately (show label where possible) S36/37 - Wear suitable protective clothing and gloves S24/25 - Avoid contact with skin and eyes

Other Information

This material is sold for research purposes only and is not required to appear on the TSCA inventory. It is not intended for food, drug, household, agricultural or cosmetic use. Its use must be supervised by a technically qualified individual experienced in handling potentially hazardous chemicals. The above information is correct to the best of our knowledge. Users should make independent decisions regarding completeness of the information based on all sources available. Molecular Probes shall not be held liable for any damage resulting from handling or contact with the above product.

Cell Biology

ATCC® Number:	CRL-1772™ Order this Item	Price:	$256.00
Designations:	C2C12		
Biosafety Level:	1	Shipped:	frozen
Medium & Serum:	See Propagation	Growth Properties:	adherent myoblast
Organism:	*Mus musculus* (mouse)	Morphology:	PHOTO

Source:
Tissue: muscle
Strain: C3H
Cell Type: myoblast;

Permits/Forms:
In addition to the MTA mentioned above, other ATCC and/or regulatory permits may be required for the transfer of this ATCC material. Anyone purchasing ATCC material is ultimately responsible for obtaining the permits. Please click here for information regarding the specific requirements for shipment to your location.

Related Cell Culture Products

Applications:
transfection host(technology from amaxa Roche FuGENE® Transfection Reagents)

Comments:
This is a subclone (produced by H. Blau, et al) of the mouse myoblast cell line established by D. Yaffe and O. Saxel. [22903]
The C2C12 cell line differentiates rapidly, forming contractile myotubes and producing characteristic muscle proteins. [22953]
Treatment with bone morphogenic protein 2 (BMP-2) cause a shift in the differentiation pathway from myoblastic to osteoblastic. [23427]
Tested and found negative for ectromelia virus (mousepox).

Propagation:
ATCC complete growth medium: The base medium for this cell line is ATCC-formulated Dulbecco's Modified Eagle's Medium, Catalog No. 30-2002. To make the complete growth medium, add the following components to the base medium: fetal bovine serum to a final concentration of 10%.
Temperature: 37.0°C
Protocol: IMPORTANT - DO NOT ALLOW CULTURES TO BECOME CONFLUENT.
Cultures must not be allowed to become confluent as this will deplete the myoblastic population in the culture.
Myotube formation is enhanced when the medium is supplemented with 10% horse serum instead of fetal bovine serum.

Subculturing:
1. Remove and discard culture medium.
2. Briefly rinse the cell layer with 0.25% (w/v) Trypsin- 0.53 mM EDTA solution to remove all traces of serum which contains trypsin inhibitor.
3. Add 2.0 to 3.0 ml of Trypsin-EDTA solution to flask and observe cells under an inverted microscope until cell layer is dispersed (usually within 5 to 15 minutes).
   Note: To avoid clumping do not agitate the cells by hitting or shaking the

flask while waiting for the cells to detach. Cells that are difficult to detach may be placed at 37°C to facilitate dispersal.
4. Add 6.0 to 8.0 ml of complete growth medium and aspirate cells by gently pipetting.
5. Add appropriate aliquots of the cell suspension to new culture vessels. Inoculate at a cell concentration between 1.5 X 10 exp5 and 1.0 X 10 exp6 viable cells/75 cm2.
6. Incubate cultures at 37°C.

Medium Renewal: Every two to three days

Preservation:

Freeze medium: Complete growth medium supplemented with 5% (v/v) DMSO
Storage temperature: liquid nitrogen vapor phase

Related Products:

Recommended medium (without the additional supplements or serum described under ATCC Medium):ATCC 30-2002
recommended serum:ATCC 30-2020

References:

22903: Yaffe D, Saxel O. Serial passaging and differentiation of myogenic cells isolated from dystrophic mouse muscle. Nature 270: 725-727, 1977. PubMed: 563524
22953: Blau HM, et al. Plasticity of the differentiated state. Science 230: 758-766, 1985. PubMed: 2414846
23427: Katagiri T, et al. Bone morphogenetic protein-2 converts the differentiation pathway of C2C12 myoblasts into the osteoblast lineage [published erratum appears in J Cell Biol 1995 Feb;128(4):following 713]. J. Cell Biol. 127: 1755-1766, 1994. PubMed: 7798324
28236: Chow YH, et al. Improvement of hepatitis B virus DNA vaccines by plasmids coexpressing hepatitis B surface antigen and interleukin-2. J. Virol. 71: 169-178, 1997. PubMed: 8985336
32828: Kessler PD, et al. Gene delivery to skeletal muscle results in sustanined expression and systemic delivery of a therapeutic protein. Proc. Natl. Acad. Sci. USA 93: 14082-14087, 1996. PubMed: 8943064
33069: Hsu DK, et al. Identification of a murine TEF-1-related gene expressed after mitogenic stimulation of quiescent fibroblasts and during myogenic differentiation. J. Biol. Chem. 271: 13786-13795, 1996. PubMed: 8662936

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The Mammalian Cell Culturing Facility in room 124 of the chemistry building will be used to culture mammalian cells. The users of the facility will maintain their cells cultures, and eventual plate them onto a variety of surfaces to further study the cells using different types of microscopy (Atomic force microscopy, confocal microscopy, Reman microscopy ect.). Cell staining will also take place in the lab.

Cell Biology

ATCC® Number:	CRL-2592™ Order this Item	Price:	$338.00
Designations:	A431NS	Depositors:	K Hirabayashi
Biosafety Level:	1	Shipped:	frozen
Medium & Serum:	See Propagation	Growth Properties:	adherent
Organism:	*Homo sapiens* (human)	Morphology:	epithelial

Source:
Organ: skin
Tissue: epidermis
Disease: epidermoid carcinoma

Permits/Forms:
In addition to the MTA mentioned above, other ATCC and/or regulatory permits may be required for the transfer of this ATCC material. Anyone purchasing ATCC material is ultimately responsible for obtaining the permits. Please click here for information regarding the specific requirements for shipment to your location.

Related Cell Culture Products

DNA Profile (STR):
Amelogenin: X
CSF1PO: 11,12
D13S317: 9,13
D16S539: 12,14
D5S818: 12,13
D7S820: 10
THO1: 9
TPOX: 11
vWA: 15,17

Age: 85 yrs

Gender: female

Comments:
A431NS was derived from the A431 cell line (ATCC CRL-1555) in 1997 by repeated subculturing to select cells that detached from the substrate easily. The polyinosinic-polycytidylic acid/cationic liposome complex (LIC) inhibits cancer cell growth by the induction of apoptosis. [51526]
LIC has strong cytotoxic effects on the A431NS cell line but neither polyinosinic-polycytidylic acid alone nor cationic liposome alone have antiproliferation effects. While cell sensitivity to drugs tend to be changeable, the A431NS cell line has a stable sensitivity to LIC. The cell line can be used to test the antiproliferation activity of LIC.

Propagation:
ATCC complete growth medium: The base medium for this cell line is ATCC-formulated Dulbecco's Modified Eagle's Medium, Catalog No. 30-2002. To make the complete growth medium, add the following components to the base medium: fetal bovine serum to a final concentration of 10%.
Temperature: 37.0°C

Subculturing:
Protocol: Remove medium and rinse with PBS. Remove the PBS completely and add fresh 0.25% trypsin, 0.03% EDTA solution. Allow the flask to sit at room temperature (or 37C) until the cells detach. Centrifuge the cell suspension at 1000 rpm for 10 minutes, resuspend the pellet in fresh medium, aspirate and dispense

	into new flasks. **Subcultivation Ratio:** A subcultivation ratio of 1:3 to 1:6 is recommended **Medium Renewal:** Every 2 to 3 days
Preservation:	**Freeze medium:** Complete growth medium 95%; DMSO, 5% **Storage temperature:** liquid nitrogen vapor temperture
Related Products:	Recommended medium (without the additional supplements or serum described under ATCC Medium):ATCC 30-2002 parental cell line:ATCC CRL-1555 recommended serum:ATCC 30-2020
References:	51526: Hirabayashi K, et al. Inhibition of cancer cell growth by polyinosinic-polycytidylic acid/cationic liposome complex: a new biological activity. Cancer Res. 59: 4325-4333, 1999. PubMed: 10485480

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Cell Biology

ATCC® Number:	HTB-4™ Order this Item	Price:	$268.00
Designations:	T24	Depositors:	C O'Toole
Biosafety Level:	1	Shipped:	frozen
Medium & Serum:	See Propagation	Growth Properties:	adherent
Organism:	*Homo sapiens* (human)	Morphology:	epithelial
Source:	**Organ:** urinary bladder **Disease:** transitional cell carcinoma		
Cellular Products:	tumor specific antigen		
Permits/Forms:	In addition to the MTA mentioned above, other ATCC and/or regulatory permits may be required for the transfer of this ATCC material. Anyone purchasing ATCC material is ultimately responsible for obtaining the permits. Please click here for information regarding the specific requirements for shipment to your location.		
	Related Cell Culture Products		
Applications:	transfection host (Roche FuGENE® Transfection Reagents technology from amaxa)		
Tumorigenic:	N0		
Antigen Expression:	HLA A1, A3, B18, Bw35, Cw4, DRw2, Dw4		
DNA Profile (STR):	Amelogenin: X CSF1PO: 10,12 D13S317: 12 D16S539: 9 D5S818: 10,12 D7S820: 10,11 THO1: 6 TPOX: 8,11 vWA: 17		
Cytogenetic Analysis:	hypodiploidy to hypopentaploidy; stemline 86; 2 to 4 telocentrics; 3 to 4 minutes, hypotetraploid to hypertetraploid with abnormalities including dicentrics, breaks, pulverization, minutes and telocentric markers		
Isoenzymes:	AK-1, 1 ES-D, 1 G6PD, B GLO-I, 1 Me-2, 1-2 PGM1, 1 PGM3, 1		
Age:	81 years		
Gender:	female		
Ethnicity:	Caucasian		

Comments:	Leukocytes and sera from patients with transitional cell carcinoma were cytotoxic to T24 and related lines. Cells have a 19 hour generation time. Contains the ras (H-ras) oncogene.
Propagation:	**ATCC complete growth medium:** The base medium for this cell line is ATCC-formulated McCoy?s 5a Medium Modified, Catalog No. 30-2007. To make the complete growth medium, add the following components to the base medium: fetal bovine serum to a final concentration of 10%. **Temperature:** 37.0°C
Subculturing:	**Subcultivation Ratio:** A subcultivation ratio of 1:3 to 1:8 is recommended **Medium Renewal:** 2 to 3 times per week Remove medium, and rinse with 0.25% trypsin, 0.03% EDTA solution. Remove the solution and add an additional 1 to 2 ml of trypsin-EDTA solution. Allow the flask to sit at room temperature (or at 37C) until the cells detach. Add fresh culture medium, aspirate and dispense into new culture flasks.
Preservation:	Culture medium, 95%; DMSO, 5%
Related Products:	purified DNA:ATCC HTB-4D recommended serum:ATCC 30-2020
References:	21849: O'Toole CHuman bladder cancer lines: HLA Class I and Class II antigen expression and susceptibility to cytostatic and cytotoxic effects in vitro*In:* O'Toole CIn vitro models for cancer researchvol. IVBoca Raton, FLCRC Presspp. 103-125. 22365: O'Toole C, et al. Cellular immunity to human urinary bladder carcinoma. I. Correlation to clinical stage and radiotherapy. Int. J. Cancer 10: 77-91, 1972. PubMed: 4196436 22443: Williams BY, Schonbrunn A. Bombesin receptors in a human duodenal tumor cell line: binding properties and function. Cancer Res. 54: 818-824, 1994. PubMed: 8306345 22510: Bubenik J, et al. Cellular and humoral immune responses to human urinary bladder carcinomas. Int. J. Cancer 5: 310-319, 1970. PubMed: 5452065 22536: Fogh J, et al. Absence of HeLa cell contamination in 169 cell lines derived from human tumors. J. Natl. Cancer Inst. 58: 209-214, 1977. PubMed: 833871 22539: Fogh J, et al. One hundred and twenty-seven cultured human tumor cell lines producing tumors in nude mice. J. Natl. Cancer Inst. 59: 221-226, 1977. PubMed: 327080 22849: Bubenik J, et al. Established cell line of urinary bladder carcinoma (T24) containing tumour-specific antigen. Int. J. Cancer 11: 765-773, 1973. PubMed: 4133950 23226: Pollack MS, et al. HLA-A, B, C and DR alloantigen expression on forty-six cultured human tumor cell lines. J. Natl. Cancer Inst. 66: 1003-1012, 1981. PubMed: 7017212 23256: Carey TE, et al. Cell surface antigens of human malignant melanoma: mixed hemadsorption assays for humoral immunity to cultured autologous melanoma cells. Proc. Natl. Acad. Sci. USA 73: 3278-3282, 1976. PubMed: 1067619 24381: Fogh J. Cultivation, characterization, and identification of human tumor cells with emphasis on kidney, testis, and bladder tumors. Natl. Cancer Inst. Monogr. 49: 5-9, 1978. PubMed: 571047

25065: Bellet D, et al. Malignant transformation of nontrophoblastic cells is associated with the expression of chorionic gonadotropin beta genes normally transcribed in trophoblastic cells. Cancer Res. 57: 516-523, 1997. PubMed: 9012484
26316: Bubenik J, et al. Cellular immunity to renal carcinomas in man. Int. J. Cancer 8: 503-513, 1971. PubMed: 5137312
32266: Bender CM, et al. Inhibition of DNA methylation by 5-Aza-2'-deoxycytidine suppresses the growth of human tumor cell lines. Cancer Res. 58: 95-101, 1998. PubMed: 9426064
33025: Ponton A, et al. The CD95 (APO-1/Fas) receptor activates NF-kappaB independently of its cytotoxic function. J. Biol. Chem. 271: 8991-8995, 1996. PubMed: 8621545

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