

**THE UNIVERSITY OF WESTERN ONTARIO
BIOLOGICAL AGENTS REGISTRY FORM**
Approved Biohazards Subcommittee: October 14, 2010
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 (UWO) or in charge of a laboratory/facility where the use of Level 1, 2 or 3 biological 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 be updated at least every 3 years or when there are changes to the biological agents 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 Biohazards 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	<u>Dr. Eva Turley</u>
DEPARTMENT	<u>Oncology, Cancer Research lab, LRCP</u>
ADDRESS	<u>790 Commissioners Rd. E. Room A4-931</u>
PHONE NUMBER	<u>519-685-8500 Ex. 53677 Lab Ex. 53280</u>
EMERGENCY PHONE NUMBER(S)	<u>519-685-8500 Ex. 53280</u>
EMAIL	<u>Eva.Turley@lhsc.on.ca</u>

Location of experimental work to be carried out: Building(s) LRCP building A Room(s)_A4-931, A4928, A4-824

*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 15.0, Approvals).

FUNDING AGENCY/AGENCIES: CBCF, NSERC, MITACS (Octoderma)

GRANT TITLE(S):

1. RHAMM / HMMR in Breast and Prostate Cancer progression
2. The role of RHAMM in wound repair.
3. Unconventional export and trafficking of RHAMM

List all personnel working under Principal Investigators supervision in this location:

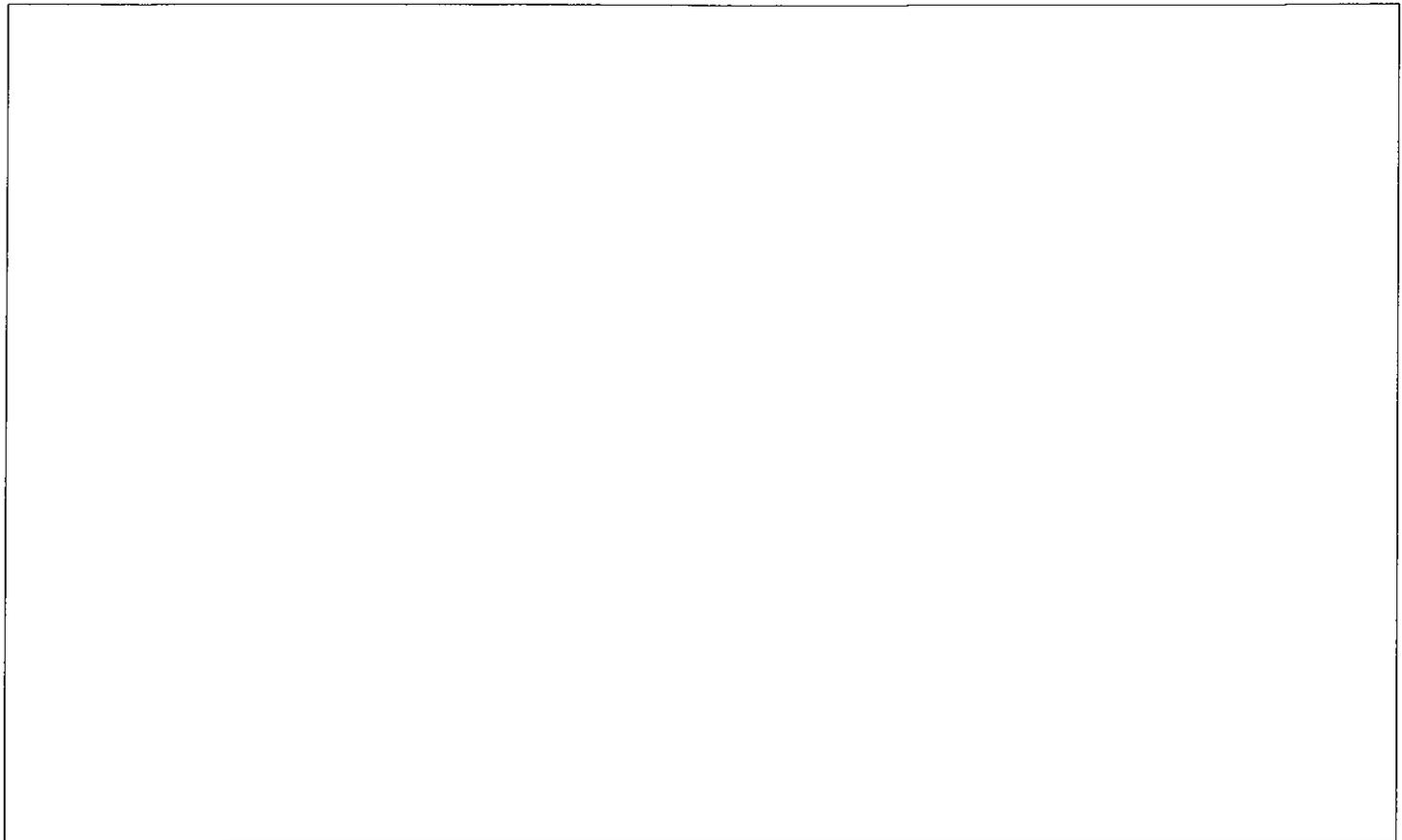
<u>Name</u>	<u>UWO E-mail Address</u>	<u>Date of Biosafety Training</u>
<u>Jenny Ma</u>	<u>Jenny.Ma@lhsc.on.ca</u>	<u>Jun 23, 2011</u>
<u>Conny Toelg</u>	<u>Conny.Toelg@lhsc.on.ca</u>	<u>Jun 24, 2011</u>
<u>Natalia Akentieva</u>	<u>Natalia.Akentieva@lhsc.on.ca</u>	<u>Jun 23, 2011</u>
<u>Pat Telmer</u>	<u>Patrick.Telmer@lhsc.on.ca</u>	<u>Jun 24, 2011</u>
<u>Caitlin Ward</u>	<u>Caitlin.Ward@lhsc.on.ca</u>	<u>Jun 23, 2011</u>
<u>Colleen Biggs</u>	<u>cbiggs4@uwo.ca</u>	<u>June 23/2011</u>
<u>Siddika Pardhan</u>	<u>spardha@uwo.ca</u>	<u>Jun 23, 2011</u>

Please explain the biological agents and/or biohazardous substances used and how they will be stored, used and disposed of. Projects without this description will not be reviewed.

1. We use *E.coli* strain dH5 α bacteria for producing recombinant RHAMM proteins. We use these proteins as reagents for producing polyclonal antibodies, as function blockers of cell invasion/migration and proliferation in culture and *in vivo*, for characterizing binding characteristics *in vitro* and for screening peptide mimetic/small chemical libraries to identify Rhamm function blocking reagents.
2. We use retroviral constructs containing Rhamm to infect murine and human fibroblast cell lines.
3. We use human cell lines including fibroblast, breast cancer, prostate cancer, and melanoma obtained from ATCC for in culture and *in vivo* (immune compromised mice) experiments.
4. We have several varieties of rodent cells which we use to assess the function and localization of Rhamm and to characterize the effects of blocking Rhamm function on cell migration/proliferation. We use murine fibroblast lines obtained from ATCC along with primary cultures which are prepared by our lab. The primary cultures are murine embryonic fibroblasts, dermal fibroblasts, wound site fibroblasts, and bone marrow fibroblasts (stem cells). These are grown from explanted tissue and maintained in culture for approximately 8 passages. We obtain these primary cells from wildtype BL6 mice, Rhamm^{-/-} BL6 mice, CD44^{-/-} BL6 mice, and Rhamm^{-/-};CD44^{-/-} BL6 mice.

Please include a one page research summary or teaching protocol.

My research program currently focuses upon defining the mechanisms by which the polysaccharide hyaluronan controls wound repair and the related process of tumour progression. Hyaluronan is a large negatively charged glycosaminoglycan produced by hyaluronan synthases. It functions both to organize the pericellular matrix and to activate signaling cascades that control cell migration and cell division. My laboratory identified, characterized and cloned the first cellular hyaluronan receptor, termed Rhamm (gene name HMMR). We are investigating the mechanisms by which this intracellular and extracellular protein controls wound repair, mesenchymal stem cell trafficking and breast/prostate cancer progression.



1.0 Microorganisms

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

Do you use microorganisms that require a permit from the CFIA? YES 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)* (Be specific)	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
E. Coli Parental Strain =K12*	<input type="radio"/> Yes <input checked="" type="radio"/> No	<input type="radio"/> Yes <input checked="" type="radio"/> No	<input type="radio"/> Yes <input checked="" type="radio"/> No	4L	Invitrogen	<input checked="" type="radio"/> 1 <input type="radio"/> 2 <input type="radio"/> 2+ <input type="radio"/> 3

* The strain *E. coli* K-12 is a debilitated strain which does not colonize the human intestine. It has also been shown to survive poorly in the environment, has a history of safe commercial use, and is not known to have adverse effects on microorganisms or plants.

2.0 Cell Culture

2.1 Does your work involve the use of cell cultures? YES 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:

Cell Type	Is this cell type used in your work?	Source of Primary Cell Culture Tissue	AUS Protocol Number
Human	<input type="radio"/> Yes <input checked="" type="radio"/> No		Not applicable
Rodent	<input checked="" type="radio"/> Yes <input type="radio"/> No	mouse fibroblastic cells	2009-060, 2009-051
Non-human primate	<input type="radio"/> Yes <input checked="" type="radio"/> No		
Other (specify)	<input type="radio"/> Yes <input type="radio"/> No		

↑ NO

2.3 Please indicate the type of established cells that will be grown in culture in:

Cell Type	Is this cell type used in your work?	Specific cell line(s)*	Containment Level of each cell line	Supplier / Source of cell line(s)
Human	<input checked="" type="radio"/> Yes <input type="radio"/> No	prostate stem cells, MDA-mb-231, <i>-level 1</i> MCF7, <i>-level 1</i> Melanoma <i>-level 1</i> WM1552C, Ovarian cancer SKOV3, <i>-level 1</i> OVCAR <i>-level 1</i>	2	Dr. Jim McCarthy's lab Minnesota, ATCC
Rodent	<input checked="" type="radio"/> Yes <input type="radio"/> No	Rhamm-/-MEF, CD44-/-MEF, Rhamm-/-CD44-/-MEF, 10T1/2, C3 <i>-level 1</i> 10T1/2, MEF Rhamm+MEK, rat dermal fibroblasts, rat mesenchymal stem cells, Raw 264.7, <i>-level 2</i>	2	Our mice, ATCC
Non-human primate	<input type="radio"/> Yes <input checked="" type="radio"/> No			
Other (specify)	<input type="radio"/> Yes <input checked="" type="radio"/> No			

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

**level 1 per info from ATCC.*

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

3.0 Use of Human Source Materials

3.1 Does your work involve the use of human source materials? YES NO

If no, please proceed to Section 4.0

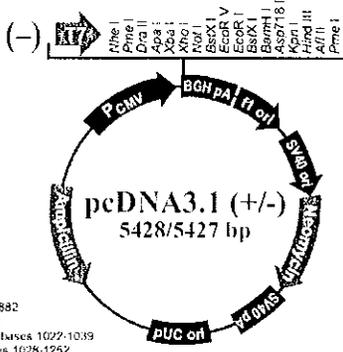
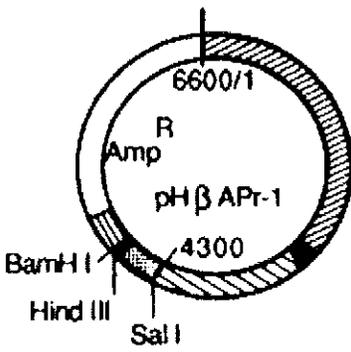
3.2 Indicate in the table below the Human Source Material to be used.

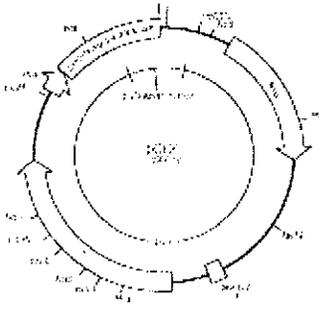
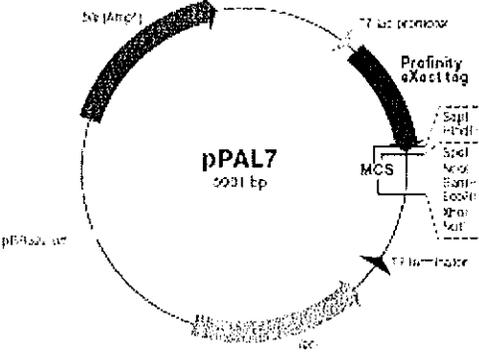
Human Source Material	Source/Supplier /Company Name	Is Human Source Material Infected With An Infectious Agent? YES/UNKNO	Name of Infectious Agent (If applicable)	PHAC or CFIA Containment Level (Select one)
Human Blood (whole) or other Body Fluid		<input type="radio"/> Yes <input type="radio"/> Unknown	Containment Level?	
Human Blood (fraction) or other Body Fluid		<input type="radio"/> Yes <input type="radio"/> Unknown		
Human Organs or Tissues (unpreserved)	LRCP patients, Dr. Trevor Shepherd	<input type="radio"/> Yes <input checked="" type="radio"/> Unknown		<input checked="" type="radio"/> 1 <input type="radio"/> 2 <input type="radio"/> 2+ <input type="radio"/> 3
Human Organs or Tissues (preserved)	Isu Abxis Accumax tissue arrays	Not Applicable		Not Applicable

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? YES NO If no, please proceed to Section 5.0

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

Bacteria Used for Cloning *	Plasmid(s) **	Source of Plasmid	Gene Transfected	Describe the change that results from transformation or tranfection
<p><i>E. coli</i> K12 Strains: DH5 alpha XL-1 Blue BI21 HB101 TOP10</p> <p>All plasmids are propagated in one of these strains</p>	<p>pcDNA3.1</p> <p>pH-Apr-1-Neo</p> <p>pH-Apr-1-Hygro</p> <p>REFERENCE: Gunning, P., J. Leavitt, G. Muscat, S. Y. Ng, and L. Kedes. 1987. A human beta-actin expression vector system directs high-level accumulation of antisense transcripts. Proc. Natl. Acad. Sci. USA 84:4831-4835.</p>	<p>Parental Vector from INVITROGEN. Inserts were generated in laboratory or obtained from ATCC/NCBI</p>  <p>3-882 10 © Invitrogen 1022-1039 ISBN 1028-1252</p> <p>Plasmid map attached. Plasmid generated by colleague referenced. Neo denotes Neomycin selection cassette and Hygro indicates Hygromycin selection cassette</p> 	<p>containing the following inserts: -RHAMM cDNAs from mouse and human. -RHAMM fusion protein vector containing RHAMM fused to ZsGreen coding sequence. -Mutant – Active MEK1 coding sequence from mouse. -H-RAS from Mouse</p> <p>-RHAMM cDNAs from mouse and Human</p>	<p>-Alterations in cell motility -Transformation of fibroblasts -Fluorescent proteins for visualization of protein trafficking</p> <p>-Alterations in cell motility -Transformation of fibroblasts</p>

	<p>pGEX-2TK</p>	<p>GE-HEALTHCARE</p> 	<p>-RHAMM cDNAs from mouse and Human for recombinant protein production in <i>E.coli</i> strains listed</p>	<p>None. Protein production</p>
	<p>pPAL7</p>	<p>BIORAD</p>  <p>Fig. 1. Profinity exact pPAL7 vector.</p>	<p>-RHAMM cDNAs from mouse for recombinant protein production in <i>E.coli</i> strains listed</p>	<p>None. Protein production</p>

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

** Please attach a plasmid map.

4.3 Will genetic modification(s) of bacteria and/or cells involving viral vectors be made?

YES, complete table below NO

Virus Used for Vector Construction	Vector(s) *	Source of Vector	Gene(s) Transduced	Describe the change that results from transduction

* Please attach a Material Safety Data Sheet or equivalent.

4.4 Will genetic sequences from the following be involved?

- ◆ HIV YES, please specify _____ NO
- ◆ HTLV 1 or 2 or genes from any Level 1 or Level 2 pathogens YES, specify _____ NO
- ◆ SV 40 Large T antigen YES NO
- ◆ E1A oncogene YES NO
- ◆ Known oncogenes YES, please specify: RHAMM/HMMR, RAS, MEK1
_____ NO
- ◆ Other human or animal pathogen and or their toxins YES, please specify _____ NO

4.5 Will virus be replication defective? YES NO

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

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

5.0 Human Gene Therapy Trials

5.1 Will human clinical trials be conducted involving a biological agent? YES NO
(including but not limited to microorganisms, viruses, prions, parasites or pathogens of plant or animal origin)
If no, please proceed to Section 6.0

5.2 If YES, please specify which biological agent will be used: _____
Please attach a full description of the biological agent.

5.2 Will the biological agent be able to replicate in the host? YES NO

5.3 How will the biological agent 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? YES, number: _____ NO PENDING

6.0 Animal Experiments

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

6.2 Name of animal species to be used Mus Musculus (Mouse)

6.3 AUS protocol # _2009-060, 2009-051_____

6.4 Will any of the agents listed in section 4.0 be used in live animals YES, specify: Fibroblasts stably transfected with RHAMM cDNA will be used in Xenograft studies NO

6.5 Will the agent(s) be shed by the animal: YES NO, please justify: Cells used in study may form tumours in mammary fat pads, but these are not shed by the animal.

7.0 Use of Animal species with Zoonotic Hazards

7.1 Will any animals with zoonotic hazards or their organs, tissues, lavages or other body fluids including blood be used (see list below)? YES No If no, please proceed to section 8.0

7.2 Will live animals be used? YES No

7.3 If yes, please specify the animal(s) used:

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

7.4 If no live animals are used, please specify the source of the specimens:

8.0 Biological Toxins

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

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

8.3 What is the LD₅₀ (specify species) of the toxin _____

8.4 How much of the toxin is handled at one time*? _____

8.5 How much of the toxin is stored*? _____

8.6 Will any biological toxins be used in live animals? YES, Please provide details: _____ NO

*For information on biosecurity requirements, please see:

http://www.uwo.ca/humanresources/docandform/docs/healthandsafety/biosafety/Biosecurity_Requirements.pdf

9.0 Insects

9.1 Do you use insects? YES 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 life stage of the insect? _____

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

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

9.7 Do you use insects that require a permit from the CFIA permit? YES NO

If YES, Please attach the CFIA permit & describe any CFIA permit conditions:

10.0 Plants

10.1 Do you use plants? 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? Grow and maintain a crop "One-time" use

10.6 Do you do any modifications to the plant? YES 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? YES NO

If YES, Please attach the CFIA permit & describe any CFIA permit conditions:

11.0 Import Requirements

11.1 Will any of the above agents be imported? YES, please give country of origin _____ NO

If no, please proceed to Section 12.0

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

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

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

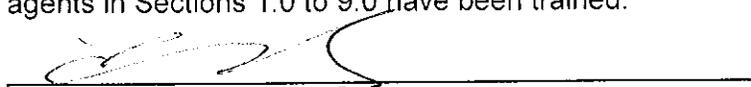
12.0 Training Requirements for Personnel Named on Form

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 biological agents in Sections 1.0 to 9.0 have been trained.

SIGNATURE _____



13.0 Containment Levels

13.1 For the work described in sections 1.0 to 9.0, please indicate the highest HC or CFIA Containment Level required.

1 2 2+ 3

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

YES, date of most recent biosafety inspection: Bio-LRCC-006 _____

NO, please certify

NOT REQUIRED for Level 1 containment

*Level 2 Biosafety Inspection
Dec. 10, 2010
[Signature]*

13.3 Please indicate permit number (not applicable for first time applicants):

Bio-LRCC-0006 _____

14.0 Procedures to be Followed

14.1 Please describe additional risk reduction measures will be taken beyond containment level 1, 2, 2+ or 3 measures, that are unique to this agent.

No specific measures beyond standard biosafety associated with levels 1 and 2

14.2 Please outline what will be done if there is an exposure to the biological agents listed, such as a needlestick injury or an accidental splash:

- Seek first aid or emergent intervention if required (refer to MSDS for Infectious Substances, or Blood-Borne Pathogen exposure protocol:

http://intra.sjhc.london.on.ca/policy/search_res.php?polid=STF008&live=1

- Immediately report injury, exposure to supervisor
- Notify Occupational Health and Safety Services (OHSS) ASAP.

Complete Workplace Occurrence Report:

http://appserver.lhsc.on.ca/policy/search_res.php?polid=OHS011&live=1

14.3 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 [Signature] Date: June 29 / 11

15.0 Approvals

1) UWO Biohazards Subcommittee: SIGNATURE: _____
Date: _____

2) Safety Officer for the University of Western Ontario
SIGNATURE: _____
Date: _____

3) Safety Officer for Institution where experiments will take place (if not UWO):
SIGNATURE: [Signature]
Date: July 7, 2011

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

Special Conditions of Approval:



Office of Biohazard Containment and Safety
Science Branch, CFIA
59 Camelot Drive, Ottawa, Ontario K1A 0Y9
Tel: (613) 221-7068 Fax: (613) 228-6129
Email: lmpoortzoopath@inspection.gc.ca

Bureau du confinement des biorisques et sécurité
Direction générale des sciences, ACIA
59 promenade Camelot, Ottawa, Ontario K1A 0Y9
Tél: (613) 221-7068 Téléc: (613) 228-6129
Courriel: lmpoortzoopath@inspection.gc.ca

October 20th, 2009

Ms. Shamila Survery / Mr. Michael Decosimo
Cedarlane Laboratories Ltd
4410 Paletta Court
Burlington, Ontario L7L 5R2

By Facsimile: (289) 288-0020

SUBJECT: Importation of *Escherichia coli* strains

Dear Ms. Survery / Mr. Decosimo:

Our office received your query about the importation of *Escherichia coli* from the American Type Culture Collection (ATCC) located in Manassas, Virginia, United States. The following *Escherichia coli* strains are considered to be level 1 animal pathogens:

- | | | | | |
|---------------|--------------------|--------------|-------------------|-------------------|
| • 5K | • CIE85 | • J52 | • MC4100 (MuLac) | • U5/41 |
| • 58 | • DH1 | • J53 | • MG1655 | • W208 |
| • 58-161 | • DH10 GOLD | • JC3272 | • MM294 | • W945 |
| • 679 | • DH10B | • JC7661 | • MS101 | • W1485 |
| • 1532 | • DH5 | • JC9387 | • NC-7 | • W3104 |
| • AB284 | • DH5-alpha | • JF1504 | • Nissle 1917 | • W3110 |
| • AB311 | • DP50 | • JF1508 | • One Shot STBL3 | • WA704 |
| • AB1157 | • DY145 | • JF1509 | • OP50 | • WP2 |
| • AB1206 | • DY380 | • JJ055 | • P678 | • X1854 |
| • AG1 | • E11 | • JM83 | • PA309 | • X2160T |
| • B | • EJ183 | • JM101 | • PK-5 | • X2541 |
| • BB4 | • EL250 | • JM109 | • PMC103 | • X2547T |
| • BD792 | • EMG2 | • K12 | • PR13 | • XL1-BLUE |
| • BL21 | • EPI 300 | • KC8 | • Rri | • XL1-BLUE-MRF |
| • BL21 (DE3) | • EZ10 | • KA802 | • RV308 | • XL0LR |
| • BM25.8 | • FDA Seattle 1946 | • KAM32 | • S17-1λ -PIR | • Y10 |
| • C | • Fusion-Blue | • KAM33 | • SCS1 | • Y1090 (1090) |
| • C-1a | • H1443 | • KAM43 | • SMR10 | • YN2980 |
| • C-3000 | • HF4714 | • LE450 | • SOLR | • W3110 |
| • C25 | • HB101 | • LE451 | • SuperchargeEZ10 | • WG1 |
| • C41 (DE3) | • HS(PFAMP)R | • LE452 | • SURE | • WG439 |
| • C43 (DE3) | • Hfr3000 | • MB408 | • TOP10 | • WG443 |
| • C600 | • Hfr3000 X74 | • MBX1928 | • TG1 | • WG445 |
| • Cavalli Hfr | • HMS174 | • MC1061 | | |

The Office of Biohazard Containment and Safety (BCS) of the Canadian Food Inspection Agency (CFIA) only issues import permits for microorganisms that are pathogenic to animals, or parts of microorganisms that are pathogenic to animals. As the products listed above are not considered pathogenic to animals, the Office of BCS does not have any regulatory requirements for their importation.

Please note that other legislation may apply. You may wish to contact the Public Health Agency of Canada's (PHAC) Office of Laboratory Security at (613) 957-1779.

Note: Microorganisms pathogenic to animals and veterinary biologics require an import permit from the CFIA.

Sincerely,

Cinthia Labrie
Head, Animal Pathogen Importation Program
Office of Biohazard Containment & Safety



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 GET QUALITY DELIVERED™

Info on Cell Line(s)

[ATCC Advanced Catalog Search](#) » **Product Details**

Product Description

Before submitting an order you will be asked to read and accept the terms and conditions of ATCC's [Material Transfer Agreement](#) or, in certain cases, an MTA specified by the depositing institution.

Customers in Europe, Australia, Canada, China, Hong Kong, India, Israel, Japan, Korea, Macau, Mexico, New Zealand, Singapore, and Taiwan, R.O.C. must contact a [local distributor](#) for pricing information and to place an order for ATCC cultures and products.

[Print this Page](#)

Cell Biology

ATCC® Number:	HTB-26™	Order this Item	Price:	\$279.00
Designations:	MDA-MB-231			Related Links ▶ NCBI Entrez Search Cell Micrograph Make a Deposit Frequently Asked Questions Material Transfer Agreement Technical Support Related Cell Culture Products Login Required ▶ Product Information Sheet
Depositors:	R Cailleau			
Biosafety Level:	1			
Shipped:	frozen			
Medium & Serum:	See Propagation			
Growth Properties:	adherent			
Organism:	<i>Homo sapiens</i> (human)			
Morphology:	epithelial			



Source:	Organ: mammary gland; breast Disease: adenocarcinoma Derived from metastatic site: pleural effusion Cell Type: epithelial
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.
Applications:	transfection host (Nucleofection technology from Lonza Roche FuGENE® Transfection Reagents)
Receptors:	epidermal growth factor (EGF), expressed transforming growth factor alpha (TGF alpha), expressed
Tumorigenic:	Yes
DNA Profile (STR):	Amelogenin: X CSF1PO: 12,13 D13S317: 13 D16S539: 12 D5S818: 12 D7S820: 8,9 THO1: 7,9,3 TPOX: 8,9 vWA: 15,18
Cytogenetic Analysis:	The cell line is aneuploid female (modal number = 64, range = 52 to 68), with chromosome counts in the near-triploid range. Normal chromosomes N8 and N15 were absent. Eleven stable rearranged marker chromosomes are noted as well as unassignable chromosomes in addition to the majority of autosomes that are trisomic. Many of the marker chromosomes are identical to those shown in the karyotype reported by K.L. Satya-Prakash, et al.

Isoenzymes:	AK-1, 1 ES-D, 1 G6PD, 8 GLO-I, 2 Me-2, 1-2 PGM1, 1-2 PGM3, 1
Age:	51 years adult
Gender:	female
Ethnicity:	Caucasian
Comments:	The cells express the WNT7B oncogene [PubMed: 8168088].
Propagation:	ATCC complete growth medium: The base medium for this cell line is ATCC-formulated Leibovitz's L-15 Medium, Catalog No. 30-2008. To make the complete growth medium, add the following components to the base medium: fetal bovine serum to a final concentration of 10%. Atmosphere: air, 100% Temperature: 37.0°C
Subculturing:	Protocol: <ol style="list-style-type: none">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 that 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.6. Incubate cultures at 37°C without CO₂. Subcultivation Ratio: A subcultivation ratio of 1:2 to 1:4 is recommended Medium Renewal: 2 to 3 times per week
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 30-2008 recommended serum:ATCC 30-2020 purified DNA:ATCC 45518 purified DNA:ATCC 45519 purified DNA:ATCC HTB-26D purified RNA:ATCC HTB-26R

References:

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Cell Biology

ATCC® Number:	HTB-22™	Order this Item	Price:	\$279.00
Designations:	MCF7			Related Links ▶ NCBI Entrez Search Cell Micrograph Make a Deposit Frequently Asked Questions Material Transfer Agreement Technical Support Related Cell Culture Products Login Required ▶ Product Information Sheet
Depositors:	CM McGrath			
Biosafety Level:	1			
Shipped:	frozen			
Medium & Serum:	See Propagation			
Growth Properties:	adherent			
Organism:	<i>Homo sapiens</i> (human)			
Morphology:	epithelial			



Source:	Organ: mammary gland; breast Disease: adenocarcinoma Derived from metastatic site: pleural effusion Cell Type: epithelial
Cellular Products:	insulin-like growth factor binding proteins (IGFBP) BP-2; BP-4; BP-5
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.
Applications:	transfection host (Nucleofection technology from Lonza Roche FuGENE® Transfection Reagents)
Receptors:	estrogen receptor, expressed
Antigen Expression:	Blood Type O; Rh+
DNA Profile (STR):	Amelogenin: X CSF1PO: 10 D13S317: 11 D16S539: 11,12 D5S818: 11,12 D7S820: 8,9 THO1: 6 TPOX: 9,12 vWA: 14,15
Cytogenetic Analysis:	modal number = 82; range = 66 to 87. The stemline chromosome numbers ranged from hypertriploidy to hypotetraploidy, with the 2S component occurring at 1%. There were 29 to 34 marker chromosomes per S metaphase; 24 to 28 markers occurred in at least 30% of cells, and generally one large submetacentric (M1) and 3 large subtelocentric (M2, M3, and M4) markers were recognizable in over 80% of metaphases. No DM were detected. Chromosome 20 was nullisomic and X was disomic.

Isoenzymes:	AK-1, 1 ES-D, 1-2 G6PD, B GLO-I, 1-2 PGM1, 1-2 PGM3, 1
Age:	69 years adult
Gender:	female
Ethnicity:	Caucasian
Comments:	The MCF7 line retains several characteristics of differentiated mammary epithelium including ability to process estradiol via cytoplasmic estrogen receptors and the capability of forming domes. The cells express the WNT7B oncogene [PubMed: 8168088]. Growth of MCF7 cells is inhibited by tumor necrosis factor alpha (TNF alpha). Secretion of IGFBP's can be modulated by treatment with anti-estrogens.
Propagation:	ATCC complete growth medium: The base medium for this cell line is ATCC-formulated Eagle's Minimum Essential Medium, Catalog No. 30-2003. To make the complete growth medium, add the following components to the base medium: 0.01 mg/ml bovine insulin; fetal bovine serum to a final concentration of 10% . Atmosphere: air, 95%; carbon dioxide (CO ₂), 5% Temperature: 37.0°C
Subculturing:	Protocol: Volumes used in this protocol are for 75 sq cm flasks; proportionally reduce or increase amount of dissociation medium for culture vessels of other sizes. Note: if floating cells are present, it is recommended that they be transferred at the first two (2) subcultures as described below. It is not necessary to transfer floating cells for subsequent subcultures. <ol style="list-style-type: none"> 1. Remove culture medium to a centrifuge tube. 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 37C to facilitate dispersal. 4. Add 6.0 to 8.0 ml of complete growth medium and aspirate cells by gently pipetting. 5. Transfer the cell suspension to the centrifuge tube with the medium and cells from step 1, and centrifuge at approximately 125 xg for 5 to 10 minutes. Discard the supernatant. 6. Resuspend the cell pellet in fresh growth medium. Add appropriate aliquots of the cell suspension to new culture vessels. 7. Incubate cultures at 37C. Subcultivation Ratio: A subcultivation ratio of 1:3 to 1:6 is recommended Medium Renewal: 2 to 3 times per week
Preservation:	Freeze medium: Complete growth medium supplemented with 5% (v/v) DMSO Storage temperature: liquid nitrogen vapor phase
Doubling Time:	29 hrs
Related Products:	Recommended medium (without the additional supplements or serum described under ATCC Medium): ATCC 30-2003 recommended serum: ATCC 30-2020 purified DNA: ATCC HTB-22D purified RNA: ATCC HTB-22R 0.25% (w/v) Trypsin - 0.53 mM EDTA in Hank' BSS (w/o Ca ⁺⁺ , Mg ⁺⁺): ATCC 30-2101 Cell culture tested DMSO: ATCC 4-X

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Cell Biology

ATCC® Number: HTB-77™ [Order this Item](#)

Price: \$279.00

Designations: SK-OV-3 [SKOV-3]

Depositors: G Trempe, LJ Old

Biosafety Level: 1

Shipped: frozen

Medium & Serum: [See Propagation](#)

Growth Properties: adherent

Organism: *Homo sapiens* (human)

Morphology: epithelial

Source: **Organ:** ovary
Disease: adenocarcinoma
Derived from metastatic site: ascites

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.

Restrictions: The cells are distributed for research purposes only. The Memorial Sloan-Kettering Cancer Center releases the line subject to the following: 1.) The cells or their products must not be distributed to third parties. Commercial interests are the exclusive property of Memorial Sloan-Kettering Cancer Center. 2.) Any proposed commercial use of these cells must first be negotiated with The Director, Office of Industrial Affairs, Memorial Sloan-Kettering Cancer Center, 1275 York Avenue, New York, NY 10021; phone (212) 639-6181; FAX (212) 717-3439.

Isolation: Isolation date: 1973

Applications: transfection host ([Nucleofection technology from Lonza Roche FuGENE® Transfection Reagents](#))

Tumorigenic: Yes

Antigen Expression: Blood Type B; Rh+

DNA Profile (STR): Amelogenin: X
CSF1PO: 11
D13S317: 8,11
D16S539: 12
D5S818: 11
D7S820: 13,14
THO1: 9,9.3
TPOX: 8,11
vWA: 17,18

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Cytogenetic Analysis:	This is a hypodiploid human cell line. The modal chromosome number was 43, occurring in 63.3% of cells. The range was 42 to 45. The rate of higher ploidies was 32%. The del(1)(q21), der(13)t(1;?:13)(q11:?:q34), der(11)t(11;?) (q12), del(10)(q22) and 3 other marker chromosomes were common to most cells, and 3 others were found only in some cells. One N11 had the HSR segment from p11 to the distal end. The normal N10, N12, N15, N17 and N19 were absent. Others were either single or paired. There were from 1 to 6 rearranged and unassignable chromosomes. The X chromosome was either single or paired.
Isoenzymes:	AK-1, 1 ES-D, 1 G6PD, B GLO-I, 1-2 Me-2, 1 PGM1, 1-2 PGM3, 1
Age:	64 years
Gender:	female
Ethnicity:	Caucasian
Comments:	SK-OV-3 cells are resistant to tumor necrosis factor and to several cytotoxic drugs including diphtheria toxin, cis-platinum and adriamycin.
<u>Propagation:</u>	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%. Atmosphere: air, 95%; carbon dioxide (CO ₂), 5% Temperature: 37.0°C
Subculturing:	Protocol: <ol style="list-style-type: none">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 that 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.6. Incubate cultures at 37°C. Subcultivation Ratio: A subcultivation ratio of 1:2 to 1:6 is recommended Medium Renewal: 2 to 3 times per week
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 30-2007 recommended serum: ATCC 30-2020

References:

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Cell Biology

ATCC® Number: HTB-161™ [Order this Item](#) Price: \$279.00

Designations: NIH:OVCAR-3
 Depositors: R Ozols, TC Hamilton
 Biosafety Level: 1
 Shipped: frozen
 Medium & Serum: [See Propagation](#)
 Growth Properties: adherent
 Organism: *Homo sapiens* (human)
 Morphology: epithelial



Source: Organ: ovary
 Disease: adenocarcinoma
 Cell Type: epithelial

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.

Isolation: Isolation date: 1982

Applications: transfection host ([Roche FuGENE® Transfection Reagents](#))

Receptors: androgen receptor, positive; estrogen receptor, positive; progesterone receptor, positive

Tumorigenic: Yes

DNA Profile (STR): Amelogenin: X
 CSF1PO: 11,12
 D13S317: 12
 D16S539: 12
 D5S818: 11,12
 D7S820: 10
 THO1: 9,9.3
 TPOX: 8
 vWA: 17

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Cytogenetic Analysis:	The cell line is aneuploid human female, with chromosome counts in the sub to near-triploid range. Several normal chromosomes (N11, N13, N14, N15, N16, N17, and N22) are clearly under-represented. Many of these missing chromosomes are represented in the large number of cytogenetically altered chromosomes identified as marker chromosomes. In addition to the marker chromosomes, there are a large number of other structurally abnormal and unassignable chromosomes that are not recognized as markers. Random loss and gain of chromosomes from cell to cell are noted in the exact chromosome counts and in the analysis of the karyotypes.
Isoenzymes:	AK-1, 1 ES-D, 1 G6PD, 8 GLO-I, 1 PGM1, 1 PGM3, 1
Age:	60 years
Gender:	female
Ethnicity:	Caucasian
Comments:	The NIH:OVCAR-3 line was established in 1982 by T.C. Hamilton, et al. from the malignant ascites of a patient with progressive adenocarcinoma of the ovary. Forms colonies in soft agar and has an abnormal karyotype. Resistant to clinically relevant concentrations of adriamycin, melphalan and cisplatin. Both cultured cells and xenografts exhibit androgen and estrogen receptors. Xenograft models have been used to show that treatment with 17 beta estradiol can induce progesterone receptors in this human ovarian carcinoma. NIH:OVCAR-3 is an appropriate model system in which to study drug resistance in ovarian cancer, and the presence of hormone receptors should be useful for the evaluation of hormonal therapy.
Propagation:	ATCC complete growth medium: The base medium for this cell line is ATCC-formulated RPMI-1640 Medium, Catalog No. 30-2001. To make the complete growth medium, add the following components to the base medium: 0.01 mg/ml bovine insulin; fetal bovine serum to a final concentration of 20%. Temperature: 37.0°C Atmosphere: air, 95%; carbon dioxide (CO ₂), 5%
Subculturing:	Protocol: Volumes used in this protocol are for 75 sq cm flasks; proportionally reduce or increase amount of dissociation medium for culture vessels of other sizes. <ul style="list-style-type: none">• Remove and discard culture medium.• Briefly rinse the cell layer with Ca⁺⁺/Mg⁺⁺ free Dulbecco's phosphate-buffered saline (D-PBS) or 0.25% (w/v) Trypsin - 0.53 mM EDTA solution to remove all traces of serum which contains trypsin inhibitor.• 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 37C to facilitate dispersal.• Add 2.0 to 3.0 ml of complete growth medium and aspirate cells by gently pipetting• Resuspend the cell pellet in fresh growth medium. Add appropriate aliquots of the cell suspension to new culture vessels.• Incubate cultures at 37C. Subcultivation Ratio: A subcultivation ratio of 1:2 to 1:4 is recommended Medium Renewal: Every 2 to 3 days
Preservation:	Freeze medium: Complete growth medium, 95%; DMSO, 5% Storage temperature: liquid nitrogen vapor temperature
Related Products:	Recommended medium (without the additional supplements or serum described under ATCC Medium): ATCC 30-2001 recommended serum: ATCC 30-2020

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Cell Biology

ATCC® Number: CRL-1213™ [Order this Item](#) Price: \$429.00

Designations: FR (Rat Dermal Fibroblastic)

Depositors: B Smith

Biosafety Level: 1

Shipped: frozen

Medium & Serum: [See Propagation](#)

Growth Properties: adherent

Organism: Rattus norvegicus (rat)

Morphology: fibroblast



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Source: Organ: skin
Strain: Sprague-Dawley
Disease: normal

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.

Age: 18 days gestation

Propagation: **ATCC complete growth medium:** The base medium for this cell line is ATCC-formulated Eagle's Minimum Essential Medium, Catalog No. 30-2003. 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: Volumes used in this protocol are for 75 sq cm flasks; proportionally reduce or increase amount of dissociation medium for culture vessels of other sizes.

1. Remove and discard culture medium.
2. Briefly rinse the cell layer with Ca⁺⁺/Mg⁺⁺ free Dulbecco's phosphate-buffered saline (D-PBS) or 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 10 to 20 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 37C 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.
6. Incubate cultures at 37°C.

Subcultivation Ratio: A subcultivation ratio of 1:3 is recommended

Medium Renewal: 2 to 3 times per week

Preservation:

Freeze medium: Complete growth medium, 95%; DMSO, 5%

Storage temperature: liquid nitrogen vapor phase

Related Products:

Recommended medium (without the additional supplements or serum described under ATCC Medium): [ATCC 30-2003](#)

recommended serum: [ATCC 30-2020](#)

0.25% (w/v) Trypsin - 0.53mM EDTA in Hank's BSS (w/o Ca⁺⁺, Mg⁺⁺): [ATCC 30-2101](#)

Phosphate-buffered saline: [ATCC 30-2200](#)

Cell culture tested DMSO: [ATCC 4-X](#)

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Cell Biology

ATCC® Number: **CCL-226™** [Order this Item](#) Price: **\$279.00**

Designations: **C3H/10T1/2, Clone 8**

Depositors: C Heidelberg

Biosafety Level: 1

Shipped: frozen

Medium & Serum: [See Propagation](#)

Growth Properties: adherent

Organism: **Mus musculus (mouse)**

Morphology: **fibroblast**



Source: **Strain: C3H**
Organ: embryo

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.

Applications: transfection host ([Roche FuGENE® Transfection Reagents](#))

Tumorigenic: No

Antigen Expression: H-2k

Cytogenetic Analysis: Mouse karyotype with a modal number of 80 chromosomes.

Age: embryo

Related Links



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- Comments:** C3H/10T1/2, Clone 8 was isolated by C. Reznikoff, D. Brankow and C. Heidelberger in 1972 from a line of C3H mouse embryo cells. [23019]
The cells are very sensitive to post confluence inhibition of cell division, do not produce tumors in syngeneic mice, have no background of spontaneous transformation, nor do they contain overt endogenous transforming murine leukemia or sarcoma viruses. [22697]
The cells are contact sensitive.
There is no detectable background spontaneous transformation.
They are highly susceptible to transformation by chemical agents. [1208]
Tested and found negative for ectromelia virus (mousepox).
NOTE: THE INOCULATION DENSITY, FEEDING AND HARVESTING SCHEDULES MUST BE FOLLOWED RIGIDLY IF THE LINE IS TO RETAIN ITS ESSENTIAL CHARACTERISTICS.
THE BATCH OF SERUM USED FOR GROWTH AND FOR TRANSFORMATION ASSAYS MAY AFFECT BOTH THE MORPHOLOGY OF THIS LINE AND THE RESULTS OBTAINED.
Monolayers established and maintained for the standard transformation assay should be free of all foci after 6 weeks. [1208]
The donor recommends that the line be used between the 5th and 15th passages only.
- Propagation:** **ATCC complete growth medium:** The base medium for this cell line is Eagle's Basal medium with 2 mM L-glutamine , 1.5 g/L sodium bicarbonate and Earle's BSS. To make the complete growth medium, add the following components to the base medium: heat-inactivated fetal bovine serum to a final concentration of 10%.
Temperature: 37.0°C
Atmosphere: air, 95%; carbon dioxide (CO2), 5%
- Subculturing:** **Protocol:** Remove medium, and rinse with 0.25% trypsin, 0.53 mM 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. SUBCULTURE MUST BE DONE BEFORE THE CULTURE REACHES CONFLUENCE.
Subcultivation Ratio: Seed new flasks at 2000 viable cells/sq cm.
Medium Renewal: Once between subcultures if necessary
- Preservation:** **Freeze medium:** Complete growth medium 95%; DMSO, 5%
Storage temperature: liquid nitrogen vapor temperature
- References:** 1208: Reznikoff CA, et al. Quantitative and qualitative studies of chemical transformation of cloned C3H mouse embryo cells sensitive to postconfluence inhibition of cell division. *Cancer Res.* 33: 3239-3249, 1973. PubMed: [4796800](#)
1209: Terzaghi M, Little JB. Repair of potentially lethal radiation damage in mammalian cells is associated with enhancement of malignant transformation. *Nature* 253: 548-549, 1975. PubMed: [1167940](#)
1210: Mondal S, Heidelberger C. Transformation of C3H/10T1/2 CL8 mouse embryo fibroblasts by ultraviolet irradiation and a phorbol ester. *Nature* 260: 710-711, 1976. PubMed: [1264242](#)
22440: Smith GJ, et al. Clonal analysis of the expression of multiple transformation phenotypes and tumorigenicity by morphologically transformed 10T1/2 cells. *Cancer Res.* 53: 500-508, 1993. PubMed: [8425183](#)
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33039: Jain MK, et al. Molecular cloning and characterization of SmlIM, a developmentally regulated LIM protein preferentially expressed in aortic smooth muscle cells. *J. Biol. Chem.* 271: 10194-10199, 1996. PubMed: [8626582](#)

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Cell Biology
ATCC® Number: TIB-71™ [Order this Item](#) **Price:** \$279.00

Designations: RAW 264.7

Depositors: WC Raschke

Biosafety Level: 2

Shipped: frozen

Medium & Serum: [See Propagation](#)
Growth Properties: adherent

Organism: *Mus musculus* (mouse)

Morphology: monocyte/macrophage

Source: **Tissue:** ascites
Strain: BALB/c
Disease: Abelson murine leukemia virus-induced tumor
Cell Type: macrophage; Abelson murine leukemia virus transformed

Cellular Products: lysozyme [1207]

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.

Applications: Biological response [92560]
 transfection host ([Roche FuGENE® Transfection Reagents](#))

Receptors: complement (C3) [1207]

Antigen Expression: H-2d

Age: adult

Gender: male

Comments: This line was established from a tumor induced by Abelson murine leukemia virus. They are negative for surface immunoglobulin (slg-), Ia (Ia-) and Thy-1.2 (Thy-1.2) This line does not secrete detectable virus particles and is negative in the XC plaque formation assay. The cells will pinocytose neutral red and will phagocytose latex beads and zymosan. They are capable of antibody dependent lysis of sheep erythrocytes and tumor cell targets. LPS or PPD treatment for 2 days stimulates lysis of erythrocytes but not tumor cell targets. Data communicated in Feb. 2007 by Dr Janet W. Hartley, indicates the expression of infectious ecotropic MuLV closely related, if not identical, to the Moloney MuLV helper virus used in the original virus inoculum. The cells also express polytropic MuLV, unsurprisingly based on the mouse passage history of the virus stocks [PubMed 18177500].

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<u>Propagation:</u>	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%. Atmosphere: air, 95%; carbon dioxide (CO ₂), 5% Temperature: 37.0°C
<u>Subculturing:</u>	Protocol: Subcultures are prepared by scraping. For a 75 cm ² flask, remove all but 10 ml culture medium (adjust amount accordingly for other culture vessels). Dislodge cells from the flask substrate with a cell scraper; aspirate and add appropriate aliquots of the cell suspension into new culture vessels. Subcultivation Ratio: A subcultivation ratio of 1:3 to 1:6 is recommended Medium Renewal: Replace or add medium every 2 to 3 days.
<u>Preservation:</u>	Freeze medium: Complete growth medium supplemented with 5% (v/v) DMSO Storage temperature: liquid nitrogen vapor phase
<u>Related Products:</u>	Recommended medium (without the additional supplements or serum described under ATCC Medium): ATCC 30-2002 recommended serum: ATCC 30-2020
<u>References:</u>	1135: Ralph P, Nakoinz I. Antibody-dependent killing of erythrocyte and tumor targets by macrophage-related cell lines: enhancement by PPD and LPS. <i>J. Immunol.</i> 119: 950-954, 1977. PubMed: 894031 1207: Raschke WC, et al. Functional macrophage cell lines transformed by Abelson leukemia virus. <i>Cell</i> 15: 261-267, 1978. PubMed: 212198 32443: Denlinger LC, et al. Regulation of inducible nitric oxide synthase expression by macrophage purinoreceptors and calcium. <i>J. Biol. Chem.</i> 271: 337-342, 1996. PubMed: 8550583 32466: Hambleton J, et al. Activation of c-Jun N-terminal kinase in bacterial lipopolysaccharide-stimulated macrophages. <i>Proc. Natl. Acad. Sci. USA</i> 93: 2774-2778, 1996. PubMed: 8610116 32553: Taylor GA, et al. Identification of a novel GTPase, the inducibly expressed GTPase, that accumulates in response to interferon gamma. <i>J. Biol. Chem.</i> 271: 20399-20405, 1996. PubMed: 8702776 32901: Li YM, et al. Molecular identity and cellular distribution of advanced glycation endproduct receptors: relationship of p60 to OST-48 and p90 to 80K-H membrane proteins. <i>Proc. Natl. Acad. Sci. USA</i> 93: 11047-11052, 1996. PubMed: 8855306 33046: Panneerselvam K, Freeze HH. Mannose enters mammalian cells using a specific transporter that is insensitive to glucose. <i>J. Biol. Chem.</i> 271: 9417-9421, 1996. PubMed: 8621609 33076: Lokuta MA, et al. Mechanisms of murine RANTES chemokine gene induction by Newcastle disease virus. <i>J. Biol. Chem.</i> 271: 13731-13738, 1996. PubMed: 8662857 33162: Taylor MF, et al. In vitro efficacy of morpholino-modified antisense oligomers directed against tumor necrosis factor-alpha mRNA. <i>J. Biol. Chem.</i> 271: 17445-17452, 1996. PubMed: 8663413 92560: Standard Practice for Testing for Biological Responses to Particles in Vitro. West Conshohocken, PA:ASTM International;ASTM Standard Test Method F 1903-98R03. 16173094: Hartley JW, et al. Expression of infectious murine leukemia viruses by RAW264.7 cells, a potential complication for studies with a widely used mouse macrophage cell line. <i>Retrovirology.</i> 4: 5:1, 2008. PubMed 18177500.

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Cell Biology

ATCC® Number:	CRL-2192™	Order this Item	Price:	\$279.00
Designations:	NR8383 [AgC11x3A, NR8383.1]			Related Links ▶ NCBI Entrez Search Cell Micrograph Make a Deposit Frequently Asked Questions Material Transfer Agreement Technical Support Related Cell Culture Products Login Required ▶ Product Information Sheet
Depositors:	RJ Helmke			
Biosafety Level:	1			
Shipped:	frozen			
Medium & Serum:	See Propagation			
Growth Properties:	mixed, adherent and suspension			
Organism:	Rattus norvegicus (rat)			
Morphology:	macrophage			



Source:	Strain: Sprague-Dawley Organ: lung Disease: normal Cell Type: macrophage (alveolar);
Cellular Products:	transforming growth factor beta (TGF beta); interleukin 1 (IL-1); interleukin 6 (IL-6)
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.
Isolation:	Isolation date: August 3, 1983
Receptors:	Fc

Comments:	<p>NR8383 (normal rat, August 3, 1983) was established from normal rat alveolar macrophage cells obtained by lung lavage. The cells were cultured in the presence of gerbil lung cell conditioned medium for approximately 8 to 9 months. Subsequently the requirement for exogenous growth factors was lost. NR8383 cells were cloned and subcloned from single cells by limiting dilution, and then subcloned from soft agar three times. The cells exhibit characteristics of macrophage cells: Phagocytosis of zymosan and <i>Pseudomonas aeruginosa</i>, nonspecific esterase activity, Fc receptors, oxidative burst, IL-1, TNF beta and IL-6 secretion, and replicative response to exogenous growth factors. The cells respond to appropriate microbial, particulate or soluble stimuli with phagocytosis and killing. NR8383 cells respond to bleomycin by secreting latent transforming growth factor (TGF beta). Stimulation with bleomycin also increases TGF beta mRNA expression. These cells are sensitive to endotoxin. LPS levels of 1 to 10 ng/ml inhibit replication by 50%. LPS inhibition is nontoxic and reversible even after levels up to 0.001 mg/ml for extended periods. The NR8383 cell line provides a homogenous source of highly responsive alveolar macrophages which can be used in vitro to study macrophage related activities.</p>
Propagation:	<p>ATCC complete growth medium: Ham's F12K medium with 2 mM L-glutamine adjusted to contain 1.5 g/L sodium bicarbonate, 85%; heat inactivated fetal bovine serum, 15% Temperature: 37.0°C</p>
Subculturing:	<p>Protocol: Cultures can be maintained by transferring floating cells to additional flasks. Adherent cells may be harvested by scraping. Upon reseeded, about one half of the cells will re-attach. Cultures are most successful when set up at a floating cell concentration of 1 to 4 X 10⁵ viable cells/ml. Medium Renewal: Two to three times weekly</p>
Preservation:	<p>Freeze medium: Complete growth medium, 95%; DMSO, 5% Storage temperature: liquid nitrogen vapor phase</p>
Related Products:	<p>Recommended medium (without the additional supplements or serum described under ATCC Medium): ATCC 30-2004 purified RNA: ATCC CRL-2192R</p>
References:	<p>22160: Hidalgo HA, et al. Pneumocystis carinii induces an oxidative burst in alveolar macrophages. <i>Infect. Immun.</i> 60: 1-7, 1992. PubMed: 1729174 22316: Helmke RJ, et al. A continuous alveolar macrophage cell line: comparisons with freshly derived alveolar macrophages. <i>In Vitro Cell. Dev. Biol.</i> 25: 44-48, 1989. PubMed: 2914814 22674: Helmke RJ, et al. From growth factor dependence to growth factor responsiveness: the genesis of an alveolar macrophage cell line. <i>In Vitro Cell. Dev. Biol.</i> 23: 567-574, 1987. PubMed: 3497918 22848: Limper AH, Standing JE. Vitronectin interacts with <i>Candida albicans</i> and augments organism attachment to the NR8383 macrophage cell line. <i>Immunol. Lett.</i> 42: 139-144, 1994. PubMed: 7534269 22970: Hidalgo HA, et al. The effects of cyclosporine and dexamethasone on an alveolar macrophage cell line (NR8383). <i>Transplantation</i> 53: 620-623, 1992. PubMed: 1549855 23173: Denholm EM, Rollins SM. Expression and secretion of transforming growth factor-beta by bleomycin-stimulated rat alveolar macrophages. <i>Am. J. Physiol.</i> 264: L36-L42, 1993. PubMed: 7679254 23190: Krieg DP, et al. Resistance of mucoid <i>Pseudomonas aeruginosa</i> to nonopsonic phagocytosis by alveolar macrophages in vitro. <i>Infect. Immun.</i> 56: 3173-3169, 1988. PubMed: 3141284 23369: Sherman MP, et al. Pyrrolidine dithiocarbamate inhibits induction of nitric oxide synthase activity in rat alveolar macrophages. <i>Biochem. Biophys. Res. Commun.</i> 191: 1301-1308, 1993. PubMed: 7682068 23484: Griscavage JM, et al. Inducible nitric oxide synthase from a rat alveolar macrophage cell line is inhibited by nitric oxide. <i>J. Immunol.</i> 151: 6329-6337, 1993. PubMed: 7504017 23566: Henderson SA, et al. Nitric oxide reduces early growth response-1 gene expression in rat lung macrophages treated with interferon-gamma and lipopolysaccharide. <i>J. Biol. Chem.</i> 269: 25239-25242, 1994. PubMed: 7523382 36466: Huang S, et al. Rat KC cDNA cloning and mRNA expression in lung macrophages and fibroblasts. <i>Biochem. Biophys. Res. Commun.</i> 184: 922-929, 1992. PubMed: 1374243</p>

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Cell Biology

ATCC® Number: CRL-2808™ [Order this Item](#) Price: **\$338.00**

Designations: WM1552C [Part of the Wistar Special Collection]

Depositors: M Herlyn

Biosafety Level: 1

Shipped: frozen

Medium & Serum: [See Propagation](#)

Growth Properties: adherent

Organism: *Homo sapiens* (human)

Morphology: Spindle-shaped



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Source: Organ: skin
Tumor Stage: stage 3?
Disease: primary superficial spreading melanoma (SSM) (radial growth phase (RGP)/vertical growth phase (VGP))

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.

Restrictions: Distribution of this cell line is governed by the Wistar Special Collection [Material Transfer Agreement](#).

Isolation: Isolation date: February 8, 1988

Tumorigenic: Yes

Age: 72 years

Gender: male

Comments: The WM1552C line was established from a primary superficial spreading melanoma (SSM) in radial growth phase (RGP)/vertical growth phase (VGP) from the buttocks of a patient on 02/08/88.

Propagation: ATCC complete growth medium: 2% Tumor Medium (Tu2%) containing a 4:1 mixture of MCDB 153 medium with 1.5 g/L sodium bicarbonate and Leibovitz's L-15 medium with 2 mM L-glutamine supplemented with 0.005 mg/ml bovine insulin, 1.68 mM CaCl₂, and 2% fetal bovine serum.

Atmosphere: air, 95%; carbon dioxide (CO₂), 5%
Temperature: 37.0°C

- Subculturing:**
- Protocol:**
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 that 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.
An inoculum of 5×10^3 to 7×10^3 viable cells/cm² is recommended.
 6. Incubate cultures at 37°C.

Interval: Maintain cultures at a cell concentration between 1×10^4 and 7×10^4 cells/cm²

Subcultivation Ratio: A subcultivation of 1:3 to 1:4 is recommended

Medium Renewal: Two to three times weekly

- Preservation:**
- Freeze medium:** Complete growth medium supplemented with an additional 8% fetal bovine serum and 5% (v/v) DMSO
- Storage temperature:** liquid nitrogen vapor phase

Doubling Time: 42 hours

Related Products: recommended serum:ATCC [30-2020](#)

- References:**
- 89402: Hsu MY, et alMelanoma: The Wistar (WM) melanoma cell lines/n: Hsu MY, et alHuman Cell Culture1Great BritainKluwer Academic Publishers259-274, 1999
- 89403: Satyamoorthy K, et al. Melanoma cell lines from different stages of progression and their biological and molecular analyses. Melanoma Res. 7: S35-S42, 1997. PubMed: [9578415](#)

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