Modification Form for Permit BIO-UWO-0245

Permit Holder: Brad Urquhart

Approved Personnel
(Please stroke out any personnel to be removed)

Additional Personnel
(Please list additional personnel here)

	Please stroke out any approved Biohazards to be removed below	Write additional Biohazards for approval below. Give the full name - do not abbreviate.
Approved Microorganisms	E.Coli (TOP10 cells)	
Approved Primary and Established Cells	(Primary) (Rodent): Hepatocytes. (Established) (Human): Caco-2, LS-180, Hela. (dog): MDCKII or MDCK	
Approved Use of Human Source Material		Human plasma
Approved Genetic Modifications (Plasmids/Vectors)	(Plasmid) - pcDNA 3.1, pEF.	
Approved Use of Animals	Sprague-Dawley Rats (2009-058)	
Approved Biological Toxin(s)		

* *PLEASE ATTACH A MATERIAL SAFETY DATA SHEET OR EQUIVALENT FOR NEW BIOHAZARDS.*
** *PLEASE ATTACH A BRIEF DESCRIPTION OF THE WORK THAT EXPLAINS THE BIOHAZARDS USED AND HOW THEY WILL BE STORED, USED AND DISPOSED OF..*

As the principal investigator, I have ensured that all of the personnel named on the form have been trained. 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 of Permit Holder _____

Current Classification: 2 **Containment Level for Added Biohazards:**

Date of Last Biohazardous Agents Registry Form: Nov 11, 2009

Date of Last Modification (if applicable):

BioSafety Officer(s):

Chair, Biohazards Subcommittee: _____ **Date:** _____

Modification for Permit BIO-UWO-0245

Date: May 31, 2011

Permit Holder: Brad Urquhart, PhD

This modification seeks to add the use of human plasma to the existing permit (BIO-UWO-0245). Human plasma will be obtained from patients with kidney disease or from healthy controls by a research nurse or certified phlebotomist. The plasma will be used in incubations with established cell lines such as Caco-2 and or MDCK cells to investigate the effect of kidney disease on drug metabolism and transport. Briefly, the serum in regular cell culture media (fetal bovine serum, FBS) will be replaced by human serum from patients with varying stages of kidney disease. After 24 or 48 hour incubations, the cells will be analyzed for expression and activity of proteins involved in drug disposition, namely drug transporter proteins and drug metabolizing enzymes. In most cases the plasma will be used fresh however in some instances it may be stored at -20 celsius until use (i.e. until cells are ready for an experiment). Upon completion of the experiment, the media containing plasma will be disinfected with bleach and flushed with water into the sewer as detailed in the UWO contaminated biological waste disposal policy.

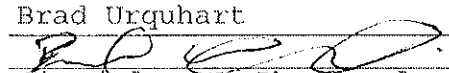
THE UNIVERSITY OF WESTERN ONTARIO
BIOHAZARDOUS AGENTS REGISTRY FORM
Approved Biohazards Subcommittee: June 26, 2009
Biosafety Website: www.uwo.ca/humanresources/biosafety/

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 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	Brad Urquhart
SIGNATURE	
DEPARTMENT	Physiology & Pharmacology
ADDRESS	Medical Sciences Building Room 216
PHONE NUMBER	83756
EMERGENCY PHONE NUMBER(S)	226 663 4389
EMAIL	Brad.Urquhart@schulich.uwo.ca

Location of experimental work to be carried out: Building(s) MSB Room(s) 235

*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: Applied for HSFC, NSERC, KFOC

GRANT TITLE(S): HSFC - The effect of exercise on cardiac drug exposure and toxicity.
NSERC - Nuclear receptor mediated control of drug transporters & DME.
KFOC - Intestinal expression and activity of drug transporters in kidney failure.

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. A GRANT SUMMARY PAGE MAYBE ADEQUATE IF IT PROVIDES SUFFICIENT DETAIL ABOUT EACH BIOHAZARD USED.

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

New Investigator - Personnel to be hired at a later date

1.0 Microorganisms

1.1 Does your work involve the use of biological agents? Ø YES O NO
(including but not limited to 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? O YES Ø 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
TOP10 cells (E. Coli)	Ø Yes O No	Ø Yes O No	Ø Yes O No	1 Litre	Invitrogen	Ø 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? Ø 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:

Cell Type	Is this cell type used in your work?	Source of Primary Cell Culture Tissue	AUS Protocol Number
Human	O Yes O No		Not applicable
Rodent	Ø Yes O No	Hepatocytes	2009-058
Non-human primate	O Yes O No		
Other (specify)	O Yes O 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)*	Supplier / Source
Human	☒ Yes O No	Caco-2, LS-180, Hela,	ATCC
Rodent	O Yes O No		
Non-human primate	O Yes O No		
Other (specify)	☒ Yes O No	MDCKII (dog) or MDCK yl.	ATCC

see email attached yl

*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? ☒ YES O 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	Patients	O Yes ☒ No		O 1 ☒ 2 O 3
Human Blood (fraction) or other Body Fluid	Patients	O Yes ☒ No		O 1 ☒ 2 O 3
Human Organs or Tissues (unpreserved)	Patients	O Yes ☒ No		O 1 ☒ 2 O 3
Human Organs or Tissues (preserved)	Patients	O Yes ☒ No		O 1 ☒ 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? ☒ YES O NO If no, please proceed to Section 5.0

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

Bacteria Used for Cloning *	Plasmid(s) *	Source of Plasmid	Gene Transfected	Describe the change that results
E. Coli	pcDNA3.1, pEF	Invitrogen	Drug transporters (MDR1, ABCG2, ABCC1	Increased efflux transporter expression

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

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

Virus Used for Vector Construction	Vector(s) *	Source of Vector	Gene(s) Transduced	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 _____ Ø NO
- HTLV 1 or 2 or genes from any Level 1 or Level 2 pathogens O YES, specify _____ Ø NO
- SV 40 Large T antigen O YES Ø NO
- E1A oncogene O YES Ø NO
- Known oncogenes O YES, please specify _____ Ø NO
- Other human or animal pathogen and or their toxins O YES, please specify _____ Ø NO

4.5 Will virus be replication defective? O YES Ø NO

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

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

5.0 Human Gene Therapy Trials

5.1 Will human clinical trials be conducted involving a biological agent? O YES Ø 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? O YES O 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? O YES, number: _____ O NO O PENDING

6.0 Animal Experiments

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

6.2 Name of animal species to be used Sprague-Dawley rats

6.3 AUS protocol # 2009-058

6.4 Will any of the agents listed be used in live animals O YES, specify: _____ Ø 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 Ø NO
- Pound source cats O YES Ø NO
- Cattle, sheep or goats O YES Ø NO
- Non-human primates O YES, please specify species _____ Ø NO
- Wild caught animals O YES, please specify species & colony # _____ Ø NO
- Birds O YES Ø NO
- Others (wild or domestic) O YES, please specify _____ Ø NO

8.0 Biological Toxins

8.1 Will toxins of biological origin be used? O YES Ø 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 LD_{50} (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*? _____

*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 Ø 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? O Initiate and maintain colony, give location: _____
O "One-time" 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 Ø 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
If NO, please forward the permit to he Biosafety Officer when available.

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 12.0 Ø 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.

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

SIGNATURE 

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. O 1 Ø 2 O 3

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

✓O YES, permit # if on-campus_____

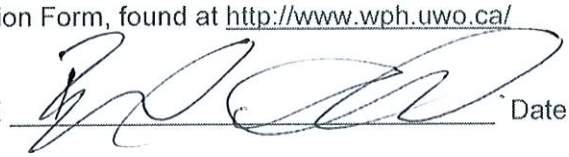
Ø NO, please certify

O NOT REQUIRED for Level 1 containment

⇒ certified Sept. 24/09 gl.

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: September 22, 2009

15.0 Approvals

UWO Biohazard Subcommittee: SIGNATURE: G.M. Kelder
Date: 11 Nov. 2009

Safety Officer for Institution where experiments will take place: SIGNATURE: gl tunley
Date: Nov 6/09

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

Approval Number: BIO-UWO-0245 Expiry Date (3 years from Approval): NOV 10, 2012

Special Conditions of Approval:

Brief Description of Research - Biohazard Subcommittee

PI: Brad Urquhart, PhD
Assistant Professor
Department of Physiology and Pharmacology
Medical Sciences Building room 288 (office)
Tel: 519 661 2111 ext 83756
E-mail: Brad.Urquhart@schulich.uwo.ca

Laboratory: Medical Sciences Building Room 235

The effect of chronic kidney disease on drug disposition

My laboratory will investigate why people with kidney failure respond differently to medications than people with normal kidney function. There are 3 themes of research that my laboratory will conduct which include clinical studies, animal studies and in vitro molecular biology.

Clinical Studies

These studies will involve human subjects and will evaluate tissue expression of proteins and mRNA for genes known to be involved in drug response. Further, these studies will evaluate the concentration of drugs in human blood. We will also isolate DNA from blood samples to evaluate epigenetic changes of target genes in patients with kidney failure. The majority of this work will involve patients with kidney failure who are screened and found negative for conditions such as HIV and Hepatitis. This screening is routinely performed as part of their renal care at LHSC. Although the patients are screened, work carried out in the laboratory will follow strict level 2 containment guidelines under the assumption that every specimen is potentially infectious.

Animal studies

Sprague-Dawley rats will be used in a surgically induced model of kidney failure. Tissues from the animals will be evaluated for protein and mRNA expression of genes known to be involved in drug response. Further, intestinal segments will be used in an everted sac model to evaluate drug transport across the intestine. Hepatocytes will also be isolated for determination of extra-renal drug metabolism and transport in the context of kidney failure.

In Vitro Molecular Biology

Molecular sub-cloning of drug transporter proteins into human (Caco-2, LS-180, HeLa) or dog (MDCKII) immortalized cell lines will be used to assess the specific contribution of individual drug transporters in established in vitro model systems. Sub-cloning will occur using the vectors pcDNA3.1 and pEF6 (both from invitrogen). Plasmids containing drug transporter inserts will be transformed into chemically competent TOP10 E. Coli cells form invitrogen and propagated prior to plasmid extraction and transfection into the above named cell lines.

MSDS FOR ANIMAL CELL CULTURES (Biosafety Level 1 or 2)

ATCC cultures are not hazardous as defined by OSHA 1910.1200. However, as live cells they are potential biohazards.

ATCC Emergency Telephone: (703) 365-2710 (24 hours)
Chemtrec: (800) 424-9300
To be used only in the event of an emergency involving a spill, leak, fire, exposure or accident.

Description

Either frozen or growing cells shipped in liquid cell culture medium (a mixture of components that may include, but is not limited to: inorganic salts, vitamins, amino acids, carbohydrates and other nutrients dissolved in water).

SECTION I
Hazardous Ingredients

Frozen cultures may contain 5 to 10% Dimethyl sulfoxide (DMSO)

SECTION II
Physical data

Pink or red aqueous liquid

SECTION III
Health hazards

For Biosafety Level 1 Cell Lines

This cell line is not known to harbor an agent known to cause disease in healthy adult humans. This cell line has **NOT** been screened for Hepatitis B, human immunodeficiency viruses or other adventitious agents. Handle as a potentially biohazardous material under at least Biosafety Level 1 containment.

For Biosafety Level 2 Cell Lines

This cell line is known to contain an agent that requires handling at Biosafety Level 2 containment [U.S. Government Publication **Biosafety in Microbiological and Biomedical Laboratories** (CDC, 1999)]. These agents have been associated with human disease. This cell line has **NOT** been screened for Hepatitis B, human immunodeficiency viruses or other adventitious agents. Cell lines derived from primate lymphoid tissue may fall under the regulations of 29 CFR 1910.1030 Bloodborne Pathogens.

SECTION IV
Fire and explosion

Not applicable

SECTION V
Reactivity data

Stable. Hazardous polymerization will not occur.

SECTION VI
Method of disposal

Spill: Contain the spill and decontaminate using suitable disinfectants such as chlorine bleach or 70% ethyl or isopropyl alcohol.
Waste disposal: Dispose of cultures and exposed materials by autoclaving at 121°C for 20 minutes. Follow all Federal, State and local regulations.

SECTION VII
Special protection information

For Biosafety Level 1 Cell Lines

Handle as a potentially biohazardous material under at least Biosafety Level 1 containment. Cell lines derived from primate lymphoid tissue may fall under the regulations of 29 CFR 1910.1030 Bloodborne Pathogens.

For Biosafety Level 2 Cell Lines

Handle as a potentially biohazardous material under at least Biosafety Level 2 containment. Cell lines derived from primate lymphoid tissue may fall under the regulations of 29 CFR 1910.1030 Bloodborne Pathogens.

SECTION VIII
Special precautions or comments

ATCC recommends that appropriate safety procedures be used when handling all cell lines, especially those derived from human or other primate material. Detailed discussions of laboratory safety procedures are provided in **Laboratory Safety: Principles and Practice** (Fleming, et al., 1995) the ATCC manual on quality control (Hay, et al., 1992), the Journal of Tissue Culture Methods (Caputo, 1988), and in the U.S. Government Publication, **Biosafety in Microbiological and Biomedical Laboratories** (CDC, 1999). This publication is available in its entirety in the Center for Disease Control Office of Health and Safety's web site at http://www.cdc.gov/od/ohs/biosfty/bmbl4/bmbl4toc.htm.

THE ABOVE INFORMATION IS CORRECT TO THE BEST OF OUR KNOWLEDGE. ALL MATERIALS AND MIXTURES MAY PRESENT UNKNOWN HAZARDS AND SHOULD BE USED WITH CAUTION. THE USER SHOULD MAKE INDEPENDENT DECISIONS REGARDING THE COMPLETENESS OF THE INFORMATION BASED ON ALL SOURCES AVAILABLE. ATCC SHALL NOT BE HELD LIABLE FOR ANY DAMAGE RESULTING FROM HANDLING OR CONTACT WITH THE ABOVE PRODUCT.

February 2002

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The Global Bioresource Center™

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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, 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-37™ Order this Item
Price: $264.00

Designations: Caco-2

Depositors: J Fogh

Biosafety Level: 1

Shipped: frozen

Medium & Serum: See Propagation

Growth Properties: adherent

Organism: *Homo sapiens* (human)

Morphology: epithelial

PHOTO

Source: **Organ:** colon
Disease: colorectal adenocarcinoma

Cellular Products: keratin
retinoic acid binding protein 1
retinol binding protein 2

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.

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

Receptors: heat stable enterotoxin (Sta, E. coli), expressed
epidermal growth factor (EGF), expressed

Virus Susceptibility: Human immunodeficiency virus 1

Tumorigenic: Yes

Reverse Transcript: N

Cytogenetic Analysis: The stemline modal chromosome number is 96, occurring at 16% with polyploidy at 3.2%. Ten common markers were detected i.e., t(1q;?), 10q-, t(11q17q) and 7 others. The t(1q17q) and M11 were found in a portion of cells. The ins(2), 10q-, and t(15q;?) were generally paired, and t(11q;17q) and t(21q;?) were mostly three-copied. Normal N9 was absent, and N21 was lost in some cells. One to 4 small acrocentric chromosomes were detected. No Y chromosome with bright distal q-band was detected by Q-observation.

Related Links
- NCBI Entrez Search
- Cell Micrograph
- Make a Deposit
- Frequently Asked Questions
- Material Transfer Agreement
- Technical Support
- Related Cell Culture Products

Isoenzymes:	AK-1, 1 ES-D, 1 G6PD, B GLO-I, 1 Me-2, 1 PGM1, 1 PGM3, 1
Age:	72 years adult
Gender:	male
Ethnicity:	Caucasian
HeLa Markers:	N
Comments:	Upon reaching confluence, the cells express characteristics of enterocytic differentiation [PubMed ID: 1939345]. Caco-2 cells express retinoic acid binding protein I and retinol binding protein II [PubMed ID: 9040537].
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 20%. **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 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. Add appropriate aliquots of the cell suspension to new culture vessels.The recommended inoculum is 1 X 10(4) viable cells/cm2. Subculture cells when they are about 80% confluent, at a cell concentration between 8 X 10(4) and 1 X 10(5) cell/cm2. 6. Incubate cultures at 37C. **Subcultivation Ratio:** A subcultivation ratio of 1:4 to 1:6 is recommended **Medium Renewal:** 1 to 2 times per week
Preservation:	**Freeze medium:** Complete growth medium, 95%; DMSO, 5% **Storage temperature:** liquid nitrogen vapor temperature
Doubling Time:	about 62 hours
Related Products:	derivative:ATCC CRL-2102 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 recommended serum:ATCC 30-2020 Recommended medium (without the additional supplements or serum described under ATCC Medium):ATCC 30-2003

References:

18385: Didier ES, et al. Characterization of Encephalitozoon (Septata) intestinailis isolates cultured from nasal mucosa and bronchoalveolar lavage fluids of two AIDS patients. J. Eukaryot. Microbiol. 43: 34-43, 1996. PubMed: 8563708
22409: Jumarie C, Malo C. Caco-2 cells cultured in serum-free medium as a model for the study of enterocytic differentiation in vitro. J. Cell. Physiol. 149: 24-33, 1991. PubMed: 1939345
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
22564: Adachi A, et al. Productive, persistent infection of human colorectal cell lines with human immunodeficiency virus. J. Virol. 61: 209-213, 1987. PubMed: 3640832
22861: Trainer DL, et al. Biological characterization and oncogene expression in human colorectal carcinoma cell lines. Int. J. Cancer 41: 287-296, 1988. PubMed: 3338874
23009: Cohen MB, et al. Receptors for Escherichia coli heat stable enterotoxin in human intestine and in a human intestinal cell line (Caco-2). J. Cell. Physiol. 156: 138-144, 1993. PubMed: 8100232
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Cell Biology

ATCC® Number: CL-187™ Order this Item

Price: $323.00

Designations: LS 180

Depositors: Northwestern University

Biosafety Level: 1

Shipped: frozen

Medium & Serum: See Propagation

Growth Properties: adherent

Organism: *Homo sapiens* (human)

Morphology: epithelial

Source:
Organ: colon
Tumor Stage: Dukes' type B
Disease: colorectal adenocarcinoma

Cellular Products:
carcinoembryonic antigen (The production of CEA in the ATCC seed stock was 1916 ng per 10(6) cells in 10 days.)
interleukin 10 [22511]
interleukin 6
mucin

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Tumorigenic: Yes

Reverse Transcript: negative

Antigen Expression: serologically defined colon cancer antigen 3; Homo sapiens, expressed HLA A2, B13, B50; Blood type O

DNA Profile (STR):
Amelogenin: X
CSF1PO: 10,13
D13S317: 10
D16S539: 11,13
D5S818: 10,15
D7S820: 11,9.3
THO1: 6,7
TPOX: 8,9
vWA: 15,18

Cytogenetic Analysis: modal number = 45; range = 42 to 47.
The stemline karyotype was 45,t(X;5) (q12;q31) with tetraploid-like cells occurring at 2.2 percent. Occasional changes in karyotypes were also noted as a result of balanced translocations producing new marker chromosomes. In conjunction with this, decreases in chromosomes involving interchromosomal exchanges were seen. The karyotype was exactly the same as that originally described by the depositor.

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Isoenzymes:	ADA, 1 ES-D, 1 G6PD, B PEP-D, 1 PGD, A PGM1, 1 PGM3, 2
Age:	58 years
Gender:	female
Ethnicity:	Caucasian
Comments:	This line has never been trypsinized; however, the variant LS 174T (ATCC CL-188) has been subcultured by trypsinization.
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:** Remove most medium. Dislodge cells from the floor of the flask by banging flask, washing cells by repeat pipetting and scraping gently. Aspirate and dispense into new flasks. These cells have never been trypsinized. **Subcultivation Ratio:** A subcultivation ratio of 1:2 to 1:5 is recommended **Medium Renewal:** Two to three times per week
Preservation:	**Freeze medium:** Complete growth medium supplemented with 5% (v/v) DMSO **Storage temperature:** liquid nitrogen vapor phase
Related Products:	recommended serum:ATCC 30-2020 derived from same individual:ATCC CL-188 Recommended medium (without the additional supplements or serum described under ATCC Medium):ATCC 30-2003
References:	2148: Tom BH, et al. Human colonic adenocarcinoma cells. I. Establishment and description of a new line. In Vitro 12: 180-191, 1976. PubMed: 1262041 3524: Tom BH, et al. Process of producing carcinoembryonic antigen. US Patent 4,228,236 dated Oct 14 1980 22147: Chen TR, et al. Intercellular karyotypic similarity in near-diploid cell lines of human tumor origins. Cancer Genet. Cytogenet. 10: 351-362, 1983. PubMed: 6652615 22511: Gastl GA, et al. Interleukin-10 production by human carcinoma cell lines and its relationship to interleukin-6 expression. Int. J. Cancer 55: 96-101, 1993. PubMed: 8344757 23079: Lan MS, et al. Polypeptide core of a human pancreatic tumor mucin antigen. Cancer Res. 50: 2997-3001, 1990. PubMed: 2334903 23322: Rodrigues NR, et al. p53 mutations in colorectal cancer. Proc. Natl. Acad. Sci. USA 87: 7555-7559, 1990. PubMed: 1699228 32532: Schuetz EG, et al. P-glycoprotein: a major determinant of rifampicin-inducible expression of cytochrome P4503A in mice and humans. Proc. Natl. Acad. Sci. USA 93: 4001-4005, 1996. PubMed: 8633005

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Cell Bio

ATCC® Number: CCL-2™ Order this Item

Price: $256.00

Designations: HeLa

Depositors: WF Scherer

Biosafety Level: 2 [CELLS CONTAIN PAPOVAVIRUS]

Shipped: frozen

Medium & Serum: See Propagation

Growth Properties: adherent

Organism: *Homo sapiens* (human)

Morphology: epithelial

PHOTO

Source: **Organ:** cervix
Disease: adenocarcinoma
Cell Type: epithelial

Cellular Products: keratin
Lysophosphatidylcholine (lyso-PC) induces AP-1 activity and c-jun N-terminal kinase activity (JNK1) by a protein kinase C-independent pathway [26623]

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Applications: transfection host ([21491] Nucleofection technology from Lonza
Roche FuGENE® Transfection Reagents)
screening for Escherichia coli strains with invasive potential [21447] [21491]

Virus Susceptibility: Human adenovirus 3
Encephalomyocarditis virus
Human poliovirus 1
Human poliovirus 2
Human poliovirus 3

Reverse Transcript: negative

DNA Profile (STR): Amelogenin: X
CSF1PO: 9,10
D13S317: 12,13.3
D16S539: 9,10
D5S818: 11,12
D7S820: 8,12
THO1: 7
TPOX: 8,12
vWA: 16,18

Related Links

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Cytogenetic Analysis:	Modal number = 82; range = 70 to 164. There is a small telocentric chromosome in 98% of the cells. 100% aneuploidy in 1385 cells examined. Four typical HeLa marker chromosomes have been reported in the literature.HeLa Marker Chromosomes: One copy of Ml, one copy of M2, four-five copies of M3, and two copies of M4 as revealed by G-banding patterns. M1 is a rearranged long arm and centromere of chromosome 1 and the long arm of chromosome 3. M2 is a combination of short arm of chromosome 3 and long arm of chromosome 5. M3 is an isochromosome of the short arm of chromosome 5. M4 consists of the long arm of chromosome 11 and an arm of chromosome 19.
Isoenzymes:	G6PD, A
Age:	31 years adult
Gender:	female
Ethnicity:	Black
HeLa Markers:	Y
Comments:	The cells are positive for keratin by immunoperoxidase staining. HeLa cells have been reported to contain human papilloma virus 18 (HPV-18) sequences. P53 expression was reported to be low, and normal levels of pR8 (retinoblastoma suppressor) were found.
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%. **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 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. 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 serum:ATCC 30-2020 derivative:ATCC CCL-2.2 derivative:ATCC CCL-2.3 derivative:ATCC CCL-2.1 Recommended medium (without the additional supplements or serum described under ATCC Medium):ATCC 30-2003
Bioreactive Factors:	**Growth Factors:** T cell growth factor (TCGF)

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Designations:	MDCK (NBL-2)
Depositors:	S Madin, NB Darby
Biosafety Level:	1
Shipped:	frozen
Medium & Serum:	See Propagation
Growth Properties:	adherent
Organism:	*Canis familiaris*
Morphology:	epithelial PHOTO
Source:	**Organ:** kidney **Disease:** normal
Cellular Products:	keratin
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:** September, 1958
Applications:	transfection host (Nucleofection technology from Lonza Roche FuGENE® Transfection Reagents)
Virus Susceptibility:	Human Coxsackievirus B 5 Reovirus type 2 Adeno-associated virus 4 Vaccinia virus Vesicular stomatitis virus Adeno-associated virus 5 Human Coxsackievirus B 3 Human Coxsackievirus B 4 Human poliovirus 2
Reverse Transcript:	negative
Cytogenetic Analysis:	Polyploidy 0.2%. Two large submetacentric chromosomes noted, presumably X chromosomes, and one or two additional chromosomes with median or submedian centromeres.
Age:	adult
Gender:	female
Comments:	The MDCK cell line was derived from a kidney of an apparently normal adult female cocker spaniel, September, 1958, by S.H. Madin and N.B. Darby.The cells are positive for keratin by immunoperoxidase staining.MDCK cells have been used to study processing of beta amyloid precursor protein and sorting of its proteolytic products.

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

Subculturing:

Protocol:

1. Remove and discard culture medium.
2. Rinse the cell layer twice 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: Every 2 to 3 days

Preservation:

Freeze medium: Complete growth medium 95%; DMSO, 5%
Storage temperature: liquid nitrogen vapor phase

Related Products:

recommended serum:ATCC 30-2020
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
parental cell line:ATCC CCL-34.2
Recommended medium (without the additional supplements or serum described under ATCC Medium):ATCC 30-2003

References:

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Subject: Re: Biohazardous Agents Registry Form: Urquhart
From: Brad Urquhart <Brad.Urquhart@schulich.uwo.ca>
Date: Mon, 28 Sep 2009 09:56:30 -0400
To: Jennifer Stanley <jstanle2@uwo.ca>

Hi Jennifer,

I actually just assumed that MDCKII cells were available on ATCC and now see that they aren't. I know that regular MDCK cells are collecting duct cells and MDCKII cells are proximal tubule cells, other than that I think they are pretty similar. I tried to find a source this morning and it seems that everybody who uses them receives them as a gift from another lab. I may have to re-think whether I can change my model system to use the more readily available MDCK cell line.

Brad

Jennifer Stanley <jstanle2@uwo.ca> 28/09/2009 9:27 am >>>

Hi Brad
I noticed on your form that you listed MDCKII cells from ATCC. We were unable to information on this cell line from the ATCC website.
Is it possible that you are getting this from a different source (if so, please give a website)? Do you know what the difference is between the original MDCK cells and the MDCKII version?
Thanks,
Jennifer