

# Modification Form for Permit BIO-UWO-0141

Permit Holder: Sashko Damjanovski

## Approved Personnel

(Please stroke out any personnel to be removed)

Michelle Niewansteeg

Logan Walsh

## Additional Personnel

(Please list additional personnel here)

MAX STAFER (SEPT 09  
START)

Please stroke out any approved  
Biohazards to be removed below

Write additional Biohazards for  
approval below. \*

### Approved Microorganisms

E. coli DHS - alpha

### Approved Cells

human (established) Hs 578 BST

human (established)  
MDA MB 231  
MDA MB 435  
frog (established) A6

### Approved Use of Human Source Material

### Approved GMO

### Approved use of Animals

Xenopus laevis

\* 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 USED.

Classification: 1

Date of last Biohazardous Agents Registry Form: Mar 28, 2008

Signature of Permit Holder:



BioSafety Officer(s):

Chair, Biohazards Subcommittee:

**Modification Form for Permit BIO-UWO-0141**

**Permit Holder: Sashko Damjanovski**

Approved 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 USED.

Classification: 1

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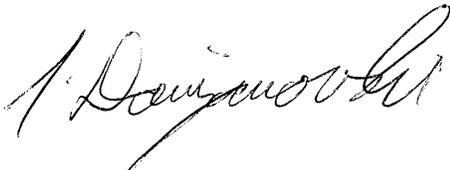
June 23, 2009

We wish to begin to use 3 new cell lines in our research.

The MDA MB 231 cell line, and its derivative MDA MB 435 line, are established human epithelial breast adenocarcinoma cell lines that are available from ATCC. Both cell types proliferate relatively quickly and have migratory capabilities, however only the 231 line, and not the 435 has invasive capabilities. Thus when plated on a 3D extracellular matrix substrate *in vitro* (such as collagen, or commercially available matrigel) the 231 cells can invade the matrix, while the 435 cannot. In addition the *Xenopus laevis* (frog) established epithelial cell line A6, (also from ATCC) will also be used in our migration studies. As our lab studies the functions of matrix metalloproteinases, secreted enzymes that are in part responsible for the cleavage and remodelling of extracellular matrices, we will use these like to look more specifically at the function of our specific matrix metalloproteinase through the transient transfection of our genes into these cells and examine their subsequent invasive capabilities. Work with these cells will all be *in vitro* and performed in the certified tissue culture room in the molecular genetics core in the WSC building (not on out lab in BGS).

Thus in addition to the cell line we are using HS 578 BST, we wish to add  
human MDA MB 231 and  
human MDA MB 435  
frog A6

Thanks

A handwritten signature in black ink, appearing to read 'Sashko Damjanovski', written in a cursive style.

Sashko Damjanovski



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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.

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## Cell Biology

**ATCC® Number:** HTB-26™

**Price:** \$256.00

**Designations:** MDA-MB-231

**Depositors:** R Cailleau

**Biosafety Level:** 1

**Shipped:** frozen

**Medium & Serum:** [See Propagation](#)

**Growth Properties:** adherent

**Organism:** *Homo sapiens* (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.

[Related Cell Culture Products](#)

**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, B  
GLO-I, 2  
Me-2, 1-2  
PGM1, 1-2  
PGM3, 1

**Age:** 51 years adult



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## Cell Biology

<b>ATCC® Number:</b>	<b>HTB-129™</b> <input type="button" value="Order this Item"/>	<b>Price:</b>	<b>\$264.00</b>
<b>Designations:</b>	MDA-MB-435S	<b>Shipped:</b>	frozen
<b><u>Biosafety Level:</u></b>	1	<b>Growth Properties:</b>	adherent
<b>Medium &amp; Serum:</b>	<a href="#">See Propagation</a>	<b>Morphology:</b>	spindle shaped
<b>Organism:</b>	<i>Homo sapiens</i> (human)		



**Source:** **Organ:** previously described as: mammary gland; breast  
**Disease:** previously described as ductal carcinoma  
**Derived from metastatic site:** pleural effusion

**Cellular Products:** tubulin; actin

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

[Related Cell Culture Products](#)

**Isolation:** **Isolation date:** 1976

**Tumorigenic:** No

**DNA Profile (STR):** Amelogenin: X  
 CSF1PO: 11  
 D13S317: 12  
 D16S539: 13  
 D5S818: 12  
 D7S820: 8,10  
 THO1: 6,7  
 TPOX: 8,11  
 vWA: 16,18

**Cytogenetic Analysis:** modal number = 56; range = 55 to 62  
 The cell line is aneuploid human female (XX), with most chromosome counts in the 55 to 60 range. Normal chromosomes N6, N11, and N22 were absent, while chromosomes N7, N13, N18 and N21 were single. Most of the remainder of normal chromosomes were usually paired, but chromosome N2 was triple. Nineteen marker chromosomes were identified, with most of them formed from structural alterations of the missing copies of the normal chromosomes. Six of these markers involve regions of chromosome N7, while three are recognized as derivatives of chromosome N6. Regions of a third copy of the normal and paired chromosomes N3, N15, N17, N20 are noted in markers M1, M2, M15, and M5, respectively.

**Isoenzymes:** AK-1, 1  
 ES-D, 1  
 G6PD, B  
 GLO-I, 2  
 PGM1, 2  
 PGM3, 1

**Age:** 31 years adult



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## Product Description

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## Cell Biology

<b>ATCC® Number:</b>	<b>CCL-102™</b>	<a href="#">Order this Item</a>	<b>Price:</b>	<b>\$323.00</b>
<b>Designations:</b>	A6		<b>Depositors:</b>	KA Rafferty
<b><u>Biosafety Level:</u></b>	1		<b>Shipped:</b>	frozen
<b>Medium &amp; Serum:</b>	<a href="#">See Propagation</a>		<b>Growth Properties:</b>	adherent
<b>Organism:</b>	Xenopus laevis (frog, South African clawed)		<b>Morphology:</b>	epithelial
<b>Source:</b>	<b>Organ:</b> kidney <b>Disease:</b> normal			
<b>Permits/Forms:</b>	In addition to the <a href="#">MTA</a> mentioned above, other <a href="#">ATCC and/or regulatory permits</a> may be required for the transfer of this ATCC material. Anyone purchasing ATCC material is ultimately responsible for obtaining the permits. Please <a href="#">click here</a> for information regarding the specific requirements for shipment to your location.			
			<b><u><a href="#">Related Cell Culture Products</a></u></b>	
<b>Virus Susceptibility:</b>	FV-4 (Frog virus)			
<b>Virus Resistance:</b>	poliovirus 1; vesicular stomatitis (Indiana); herpes simplex; vaccinia; pseudorabies			
<b>Reverse Transcript:</b>	negative			
<b>Age:</b>	adult			
<b>Gender:</b>	male			
<b>Propagation:</b>	<b>ATCC complete growth medium:</b> NCTC 109 medium, 75%; distilled water, 15%; fetal bovine serum, 10% <b>Temperature:</b> 26.0°C			
<b>Subculturing:</b>	<b>Subcultivation Ratio:</b> A subcultivation ratio of 1:2 to 1:3 is recommended <b>Medium Renewal:</b> Twice per week Remove medium, and rinse with 0.25% trypsin, 0.03% EDTA solution. Remove the solution and add an additional 1 to 2 ml of trypsin-EDTA solution. Allow the flask to sit at room temperature (or at 37C) until the cells detach. Add fresh culture medium, aspirate and dispense into new culture flasks.			
<b>Preservation:</b>	culture medium 95%; DMSO, 5%			
<b>Related Products:</b>	recommended serum:ATCC <a href="#">30-2020</a>			
<b>References:</b>	21414: . Biology of amphibian tumors. New York: Springer-Verlag; 1969. 33005: Rokaw MD, et al. Regulation of a sodium channel-associated G-protein by aldosterone. J. Biol. Chem. 271: 4491-4496, 1996. PubMed: <a href="#">8626803</a>			

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## Notices and Disclaimers

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While ATCC uses reasonable efforts to include accurate and up-to-date information on this site, ATCC makes no warranties or representations as to its accuracy. Citations from scientific literature and patents are provided for informational purposes only. ATCC does

THE UNIVERSITY OF WESTERN ONTARIO  
BIOHAZARDOUS AGENTS REGISTRY FORM  
Revised Biohazards Subcommittee: September, 2007

This form must be completed by each Principal Investigator holding a grant administered by the University of Western Ontario where the use of biohazardous infectious agents are described in the experimental work proposed. The form must also be completed if animal work is proposed involving the use of biohazardous agents or animal carrying zoonotic agents infectious to humans. Containment Levels will be required in accordance with Laboratory Biosafety Guidelines, 3rd edition, 2004, Health Canada (HC) or Containment Standards for Veterinary Facilities, 1<sup>st</sup> edition 1996, Canadian Food Inspection Agency (CFIA).

Completed forms are to be returned to Occupational Health and Safety (Stevenson-Lawson Building, Room 60) for forward to the Biohazard Subcommittee. For questions regarding this form, please contact the Biosafety Coordinator at extension 81135. If there are changes to the information on this form (excluding grant title and funding agencies) modifications must be completed and sent to Occupational Health and Safety. See website: www.uwo.ca/humanresources

PRINCIPAL INVESTIGATOR SASHKO DAMJANOVSKI  
SIGNATURE \_\_\_\_\_  
DEPARTMENT BIOLOGY  
ADDRESS 1151 RICHMOND ST LONDON ON  
PHONE NUMBER (519) 661-2111 x 84704  
EMAIL sdamjano@uwo.ca

Location of experimental work to be carried out: Building(s) BGS Room(s) 318 (Being Renovated)  
WSC 328

\*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 it being sent to Occupational Health and Safety (See Section 12.0, Approvals). For research being done at Lawson Health Research Institute, London Regional Cancer Centre, Child and Parent Research Institute or Robarts Research Institute, University Biosafety Committee members can also sign as the Safety Officer.

GRANT TITLE(S): MMP ACTIVATION DURING XENDPMS DEVELOPMENT

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

FUNDING AGENCY/AGENCIES NSERC

Names of all personnel working under Principal Investigators super vision in this location:  
LOGAN WALSH  
MICHELLE Niewensteege

**1.0 Microorganisms**

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

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?	Source/Supplier	Health Canada or CFIA Containment Level
Bacteria (DH5 $\alpha$ )	<input checked="" type="radio"/> Yes <input checked="" type="radio"/> No	<input checked="" type="radio"/> Yes <input checked="" type="radio"/> No	<input type="radio"/> Yes <input checked="" type="radio"/> No	50 mL	Invitrogen	<input checked="" type="radio"/> 1 <input type="radio"/> 2 <input type="radio"/> 3
	<input type="radio"/> Yes <input type="radio"/> No	<input type="radio"/> Yes <input type="radio"/> No	<input type="radio"/> Yes <input type="radio"/> No			<input type="radio"/> 1 <input type="radio"/> 2 <input type="radio"/> 3
	<input type="radio"/> Yes <input type="radio"/> No	<input type="radio"/> Yes <input type="radio"/> No	<input type="radio"/> Yes <input type="radio"/> No			<input type="radio"/> 1 <input type="radio"/> 2 <input type="radio"/> 3
	<input type="radio"/> Yes <input type="radio"/> No	<input type="radio"/> Yes <input type="radio"/> No	<input type="radio"/> Yes <input type="radio"/> No			<input type="radio"/> 1 <input type="radio"/> 2 <input type="radio"/> 3

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

**2.0 Cell Culture**

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

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

Cell Type	Is this cell type used in your work?	Source of Primary Cell Culture Tissue
Human	<input type="radio"/> Yes <input type="radio"/> No	
Rodent	<input type="radio"/> Yes <input type="radio"/> No	
Non-human primate	<input type="radio"/> Yes <input type="radio"/> No	
Other (specify)		

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

Cell Type	Is this cell type used in your work?	Specific cell line(s)	Supplier / Source
Human	<input checked="" type="radio"/> Yes <input type="radio"/> No	Hs 57805+	ATCC # HTB 125
Rodent	<input type="radio"/> Yes <input type="radio"/> No		
Non-human primate	<input type="radio"/> Yes <input type="radio"/> No		
Other (specify)	<input type="radio"/> Yes <input type="radio"/> No		

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

Please attach a Material Safety Data Sheet or equivalent from the supplier. (For more information, see [www.atcc.org](http://www.atcc.org))

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

**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 Known to Be Infected With An Infectious Agent? YES/NO	Name of Infectious Agent (If applicable)	HC or CFIA Containment Level (select one)
Human Blood (whole) or other Body Fluid		<input type="radio"/> Yes <input type="radio"/> No		<input type="radio"/> 1 <input type="radio"/> 2 <input type="radio"/> 3
Human Blood (fraction) or other Body Fluid		<input type="radio"/> Yes <input type="radio"/> No		<input type="radio"/> 1 <input type="radio"/> 2 <input type="radio"/> 3
Human Organs (unpreserved)		<input type="radio"/> Yes <input type="radio"/> No		<input type="radio"/> 1 <input type="radio"/> 2 <input type="radio"/> 3
Human Tissues (unpreserved)		<input type="radio"/> Yes <input type="radio"/> No		<input type="radio"/> 1 <input type="radio"/> 2 <input type="radio"/> 3
Human Organs (preserved)		<input type="radio"/> Yes <input type="radio"/> No		<input type="radio"/> 1 <input type="radio"/> 2 <input type="radio"/> 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  NO  
 If no, please proceed to Section 5.0

4.2 Will genetic sequences from the following be involved:  
 ♦ HIV  YES  NO  
 if YES specify \_\_\_\_\_  
 ♦ HTLV 1 or 2 or genes from any CDC class 1 pathogens  YES  NO  
 if YES specify \_\_\_\_\_  
 ♦ Other human or animal pathogen and or their toxins  YES  NO  
 if YES specify \_\_\_\_\_

4.3 Will intact genetic sequences be used from  
 ♦ SV 40 Large T antigen  YES  NO If YES specify \_\_\_\_\_  
 ♦ Known oncogenes  YES  NO If YES specify \_\_\_\_\_

4.4 Will a live viral vector(s) or bacterial plasmid be used for gene transduction  YES  NO  
 If YES name \_\_\_\_\_  
 Please attach a Material Safety Data Sheet or equivalent.

4.5 List specific vector(s) to be used: \_\_\_\_\_

4.6 Will virus be replication defective  YES  NO  
 4.7 Will virus be infectious to humans or animals  YES  NO  
 4.8 Will this be expected to increase the Containment Level required  YES  NO

## 5.0 Human Gene Therapy Trials

5.1 Will human clinical trials using the viral vector in 4.0 be conducted?  YES  NO

If no, please proceed to Section 6.0

If YES attach a full description of the make-up of the virus.

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

5.3 How will the virus be administered? \_\_\_\_\_

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

5.5 Has human ethics approval been obtained?  YES  NO  N/A  PENDING

## 6.0 Animal Experiments

6.1 Will any of the agents listed be used in live animals?  YES  NO

If no, please proceed to section 7.0

6.2 Name of animal species to be used Xenopus laevis

6.3 AUS protocol # 2005-027-05

6.4 If using murine cell lines, have they been tested for murine pathogens?  YES  NO N/A

## 7.0 Use of Animal species with Zoonotic Hazards

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

- ◆ Pound source dogs  YES  NO
- ◆ Pound source cats  YES  NO
- ◆ Cattle, sheep or goats  YES  NO
- ◆ Non- Human Primates  YES  NO If YES specify species \_\_\_\_\_
- ◆ Wild caught animals  YES  NO If YES specify species \_\_\_\_\_  
colony # \_\_\_\_\_
- ◆ Birds  YES  NO
- ◆ Others (wild or domestic)  YES  NO

## 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 \_\_\_\_\_

8.3 What is the LD<sub>50</sub> (specify species) of the toxin \_\_\_\_\_

8.4 Please attach information, such as a Material Safety Data Sheet, for the toxin(s) used.

9.0 Import Requirements

9.1 Will the agent be imported?  YES  NO  
If no, please proceed to Section 10.0  
If yes, country of origin \_\_\_\_\_

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

9.3 Has an import permit been obtained from CFIA for animal pathogens?  YES  NO

9.4 Has the import permit been sent to OHS?  YES  NO  
If yes, Permit # \_\_\_\_\_

10.0 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
- ◆ Employee Health and Safety Orientation

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

SIGNATURE *A. Danvers*

11.0 Containment Levels

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

1  2  3

11.2 Has the facility been certified by OHS for this level of containment?  YES  NO

11.3 If yes, please give the date and permit number: \_\_\_\_\_

*BG 5318 closed for renovations until 2009. Sharing WSC 323 with Dr. Kohalmi under her level 1 permit.*

12.0 Approvals

UWO Biohazard Subcommittee.

Signature *G.M. Kilder* Date *28 Mar '08*

Safety Officer for Institution where experiments will take place

Signature *J. Stanley, UWO* Date *March 28, 2008*

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

Signature \_\_\_\_\_ Date \_\_\_\_\_

Expiry Date (3 years from Approval): \_\_\_\_\_