

**THE UNIVERSITY OF WESTERN ONTARIO  
BIOLOGICAL AGENTS REGISTRY FORM**  
Approved Biohazards Subcommittee: October 14, 2010  
Biosafety Website: [www.uwo.ca/humanresources/biosafety/](http://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, 1<sup>st</sup> 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](mailto: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/](http://www.uwo.ca/humanresources/biosafety/)

PRINCIPAL INVESTIGATOR	<u>Gabriel DiMattia</u>
DEPARTMENT	<u>Oncology</u>
ADDRESS	<u>790 Commissioners Rd., London, ON</u>
PHONE NUMBER	<u>53625</u>
EMERGENCY PHONE NUMBER(S)	<u>519-649-4445</u>
EMAIL	<u>dimattia@uwo.ca</u>

Location of experimental work to be carried out: Building(s) LRCP Room(s) A4-921; A4-908

\*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: CCSRI; LRCP Small Grant

GRANT TITLE(S): CCSRI: Implications of activated BMP signalling and Id1/Id3 function in ovarian cancer pathogenesis. LRCP Small Grant: PI3K/Akt/mTOR signalling & autophagy in epithelial ovarian cancer (EOC) spheroids

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

Name	UWO E-mail Address	Date of Biosafety Training
Yudith Ramos-Valdes	yudithramos@yahoo.es	June 2011
Rohann Correa	rcorrea4@uwo.ca	October 2008

**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.**

**Established human cell lines:**

The cell lines are cancer derived lines and include human breast cancer lines T47-D, MCF-7, MCF-10A, ZR-75-1 and human ovarian cancer cell lines OVCAR3, Caov3, SkOV3, Hey, Ric2, and OW-1. When these cells are cultured they are maintained in a 37C humidified chamber with 5% CO<sub>2</sub> for defined periods of time after which they are harvested for extraction of protein and/or RNA. Any unused cultures are treated with bleach before disposal as biological waste using the LHSC system of waste management using biohazardous waste containers kept in the cell culture room (A4-908), which are then sealed to be autoclaved/incinerated. This includes the plastic cell culture vessels that were used to culture the cells. These lines are stored in 1.8 ml cryovials at -150C (room A4-918) when not in use.

**Mouse primary cell culture:**

Mouse embryonic fibroblasts generated from different mouse strains including wildtype mice and transgenic mice overexpressing STC1 or STC2 are currently stored in 1.8 ml cryovials at -150C (room A4-918). These fibroblasts have a limited lifespan in culture (~1 month) after which they are eliminated as described above for the established human cell lines. Primary cultures of mouse granulosa cells from the above strains are generated for short term experiments (e.g., 10 days) and are discarded as described above. Primary ovarian granulosa cells cannot be cryopreserved because they do not survive the freezing process.

**Primary human cell cultures:**

We also generate cultures of cells from the ascites fluid of ovarian cancer patients as well as primary tumour samples. These are short term cultures relative to the established cell lines; the primary human ovarian cancer cells can be kept in culture for ~1 month before they expire. These cultures are maintained in a humidified cell culture chamber as described above for defined periods of time after which they are harvested for extraction of protein and/or RNA. Any unused cultures are treated with bleach before disposal as biological waste using the LHSC system of waste management. We cryopreserve the primary human ovarian cancer cells and store aliquots of these cells in 1.8 ml cryovials at -150C (room A4-918).

**Bacterial cell cultures:**

The E. Coli DH5 $\alpha$  bacterial cultures are harvested for the extraction of plasmid DNAs (pcDNA3.0 expression vector, pBKSII DNA cloning vector, pGL3-basic gene promoter analysis vector) and any unused portions of these cultures are bleached prior to disposal. The bacterial stocks are kept at -80C as 50% glycerol solutions. Agar culture plates containing the bacteria are kept at 4C sealed in parafilm and are discarded as biological waste after 2 weeks at 4C again, following the LHSC waste management system as described above. All purified plasmid DNAs are stored at -20C or -80C as aqueous solutions.

**Please include a one page research summary or teaching protocol.**

### **Implications of activated BMP signalling and ID1/ID3 function in ovarian cancer pathogenesis**

#### **Project summary:**

Ovarian cancer has one of the highest death rates of all cancers in women due primarily to unreliable early detection and ineffective drugs to treat the disease after it has spread. Essential to developing better detection methods and drugs is the discovery of critical factors (i.e., proteins) that drive the development of ovarian cancer. Dr. Shepherd's research focusses on a group of proteins (bone morphogenetic proteins - BMPs) that allow cells to communicate with each other and regulates the behaviour of ovarian cancer cells isolated from patients.

#### **Previous research:**

Dr. Shepherd has shown that the BMPs are produced by ovarian cancer cells and that they feedback on these cells to control their shape, movement and proliferation; characteristics that determine the aggressiveness of cancer cells. As a Translational Oncology Scientist of the LRCP, he is developing novel experimental models utilizing ovarian cancer patient tumour cells to determine how the BMPs, and two genes controlled by BMPs, specifically ID1 and ID3, function to regulate the initiation and progression of ovarian cancer. Dr. Shepherd, with co-investigator Dr. DiMattia, form the basic scientist component of the Translational Ovarian Cancer Research Program at the London Health Sciences Centre and work with gynecologic oncology surgeons to procure and maintain ovarian cancer patient cells to use in laboratory studies.

#### **Project description:**

How BMP signals and ID1 and ID3 genes regulate the behaviour of normal cells of the ovary and ovarian cancer cells will be studied using three new "model systems" being developed in the Translational Ovarian Cancer Research Program:

Research Aim #1: Collect ovarian cancer cells from patients and grow them in the lab under conditions that imitate how they grow in ovarian cancer patients as 3D aggregates or spheroids—direct analysis of ovarian cancer cells from patients using this model system will provide more accurate results and thereby generate clinically-relevant insights into the disease;

Research Aim #2: Establish ovarian tumours on the surface of shell-less chick embryos as a model of ovarian cancer growth—this will be a unique opportunity to develop an innovative "bioassay" to assess how BMPs regulate tumour growth and blood supply and for future testing of new drugs that can be used to treat ovarian cancer patients;

Research Aim #3: Develop genetically-altered mice with higher than normal levels of ID1, as Dr. Shepherd has reported for ovarian tumours. We expect these mice will mimic early stages of ovarian cancer—understanding the initial molecular changes that can cause normal cells of the ovary to become cancer cells is indispensable to develop new strategies for prevention and early detection of ovarian cancer.

#### **Impact and relevance:**

These three Research Aims will define the role of BMP signals and ID1 and ID3 genes in both early and late stages of ovarian cancer. Most importantly, these studies serve to lay the foundation of the newly-established Translational Ovarian Cancer Research Program in London. To that end, Dr. Shepherd's studies will develop innovative experimental models for future pre-clinical research endeavours for early detection and to identify and test important drug targets for treating women diagnosed with ovarian cancer.

### **PI3K/Akt/mTOR signalling & autophagy in epithelial ovarian cancer (EOC) spheroids**

This proposal is designed to test the hypothesis that the transcriptional repressor protein, TRPS1 plays an important role in mammary gland development and when present at higher than normal levels results in cellular changes that predispose the gland to neoplastic transformation. This is based on the fact that TRPS1 is overexpressed across all stages of breast cancer and that it can inhibit the activity of key regulators of mammary epithelial cell differentiated function, namely the GATA family of transcription factors and specifically, GATA3. If elevated levels of TRPS1 antagonize the role GATA3 plays in maintaining differentiated function, then it is possible that mammary epithelial cells may lose proliferative control predisposing this compartment to hyperplasia or dysplasia. To test this hypothesis we have generated transgenic mice with elevated levels of TRPS1 in the mammary gland and propose to characterize the phenotypic effects of the transgene with an emphasis on studying changes in mammary gland structure that may predict a loss of proliferative control. We will compliment these studies with human breast cancer cell

line studies where the level of TRPS1 is genetically modified and determine whether this alters the ability of these cells to grow, recover from stress, and migrate. To date there is no functional data on the role of TRPS1 in human breast cells or murine mammary gland and this proposal seeks to add significant new knowledge that will indicate whether TRPS1 is a key molecular switch governing mammary epithelial cell proliferation.

## 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 DH5 $\alpha$	Yes <input checked="" type="checkbox"/> No	Yes <input checked="" type="checkbox"/> No	Yes <input checked="" type="checkbox"/> No	1 litre	Stratagene	<input checked="" type="checkbox"/> 1 2 2+ 3
	Yes No	Yes No	Yes No			1 2 2+ 3
	Yes No	Yes No	Yes No			1 2 2+ 3
	Yes No	Yes No	Yes No			1 2 2+ 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:

Cell Type	Is this cell type used in your work?	Source of Primary Cell Culture Tissue	AUS Protocol Number
Human	<input checked="" type="checkbox"/> Yes <input type="checkbox"/> No	Ovarian cancer patient ascites & solid tumour	Not applicable

		cells	
Rodent	<input checked="" type="checkbox"/> Yes    No	Mouse embryonic fibroblasts and mouse granulosa cells	2009-023
Non-human primate	Yes <input checked="" type="checkbox"/> No		
Other (specify)	Yes <input checked="" type="checkbox"/> 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="checkbox"/> Yes <input type="checkbox"/> No	T47-D, MCF-7, MCF-10A, ZR-75-1, vOSE-14, OVCAR3, Caov3, SkOV3, Hey, Ric2, and OW-1	Containment Level 1 for all cell lines	ATCC, Dr. Hal Hirte (McMaster University), Dr. Cheryl Conover (Mayo Clinic in Rochester, MN)
Rodent	<input checked="" type="checkbox"/> Yes <input type="checkbox"/> No			
Non-human primate	<input type="checkbox"/> Yes <input checked="" type="checkbox"/> No			
Other (specify)	<input type="checkbox"/> Yes <input checked="" type="checkbox"/> No			

\*Please attach a Material Safety Data Sheet or equivalent from the supplier. (For more information, see [www.atcc.org](http://www.atcc.org)) The MSDS sheets are not available for the Hey, Ric2, OW-1 (obtained from Dr. Hirte), and vOSE-14 (obtained from Dr. Conover) cell lines.

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/UNKNOWN	Name of Infectious Agent (If applicable)	PHAC or CFIA Containment Level (Select one)
Human Blood (whole) or other Body Fluid	LHSC	<input type="checkbox"/> Yes <input checked="" type="checkbox"/> Unknown		<input type="checkbox"/> 1 <input checked="" type="checkbox"/> 2 <input type="checkbox"/> 2+ <input type="checkbox"/> 3
Human Blood (fraction) or other Body Fluid		<input type="checkbox"/> Yes <input type="checkbox"/> Unknown		<input type="checkbox"/> 1 <input type="checkbox"/> 2 <input type="checkbox"/> 2+ <input type="checkbox"/> 3
Human Organs or Tissues (unpreserved)	LHSC	<input type="checkbox"/> Yes <input checked="" type="checkbox"/> Unknown		<input type="checkbox"/> 1 <input checked="" type="checkbox"/> 2 <input type="checkbox"/> 2+ <input type="checkbox"/> 3
Human Organs or Tissues (preserved)		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
E. Coli DH5 $\alpha$	<i>pcDNA3.0</i> , <i>pBKSII</i> , <i>pGL3-Basic</i>	Promega, Invitrogen, Stratagene	<i>STC1</i> , <i>STC2</i>	Changes in gene expression and/or reduction in cell proliferation

- \* 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? **NO**

- ◆ 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 \_\_\_\_\_ 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 #   2009-023  

6.4 Will any of the agents listed in section 4.0 be used in live animals    YES, specify: \_\_\_\_\_     NO

6.5 Will the agent(s) be shed by the animal:      YES       NO, please justify:

\_\_\_\_\_



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:

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**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 O "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.

*Handwritten signature*

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: Dec. 10, 2010 *Janice*  
 NO, please certify  
 NOT REQUIRED for Level 1 containment

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

**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.  
Not applicable  
\_\_\_\_\_  
\_\_\_\_\_

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:  
OH&S at LHSC (52286) would be contacted immediately, the injured area would be flushed with water and a disinfectant, then the injured person would be escorted to OH&SS (E1-505A) for immediate treatment (if necessary, otherwise monitored) and the appropriate accident forms completed and submitted to OH&SS at LHSC.

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/>

*Handwritten signature*

SIGNATURE \_\_\_\_\_ Date: 21 June 2011

**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: M. Ryden  
Date: July 25, 2011

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

Special Conditions of Approval:

----- Original Message -----

**Subject:**Re: Biological Agents Registry Form: DiMattia

**Date:**Tue, 06 Dec 2011 11:07:45 -0500

**From:**Gabriel DiMattia <dimattia@uwo.ca>

**To:**Jennifer Stanley <jstanle2@uwo.ca>

New Info

Jennifer:

E-Mail issue: Yudith doesn't have a UWO e-mail address because she's not a UWO employee (she's an LHSC technician and uses her yahoo account for everything).

Chicks: We had intended to use chick Cams in collaboration with John Lewis' lab but since he is moving to the University of Alberta, that aspect of the proposed research is no longer feasible. I have been unable to find any indication or documents showing that chick embryos could be a source of zoonotic organisms.

With regard to the cell lines: I've changed them to level 1

I hope these revision are OK.

Thank you and I apologize for my late attention to this matter.

Gabe



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: ImportZoopath@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: ImportZoopath@inspection.gc.ca

October 20<sup>th</sup>, 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

## 1. IDENTIFICATION OF THE SUBSTANCE/PREPARATION AND THE COMPANY/UNDERTAKING

**Product code** 18258012  
**Product name** SUPERScript PLAMSID SYS WITH GATEWAY TECH (MAX EFF DH5)

**Contact manufacturer**  
 INVITROGEN CORPORATON  
 1600 FARADAY AVENUE  
 PO BOX 6482  
 CARLSBAD, CA 92008  
 760-603-7200

INVITROGEN CORPORATION  
 2270 INDUSTRIAL STREET  
 BURLINGTON, ONT  
 CANADA L7P 1A1  
 800-263-6236

GIBCO PRODUCTS  
 INVITROGEN CORPORATION  
 3175 STALEY ROAD P.O. BOX 68  
 GRAND ISLAND, NY 14072  
 716-774-6700

## 2. COMPOSITION/INFORMATION ON INGREDIENTS

### Hazardous/Non-hazardous Components

Chemical Name	CAS-No	Weight %
dimethylsulfoxide	67-68-5	3-7

## 3. HAZARDS IDENTIFICATION

### Emergency Overview

Irritating to eyes. Irritating to skin.

### Form

Liquid

### Principle Routes of Exposure/ Potential Health effects

<b>Eyes</b>	May cause eye irritation with susceptible persons.
<b>Skin</b>	May cause skin irritation in susceptible persons.
<b>Inhalation</b>	No information available
<b>Ingestion</b>	No information available

### Specific effects

<b>Carcinogenic effects</b>	No information available
<b>Mutagenic effects</b>	No information available
<b>Reproductive toxicity</b>	No information available
<b>Sensitization</b>	No information available

**Target Organ Effects** Eyes. Skin.

<b>Skin contact</b>	Wash off immediately with plenty of water
<b>Eye contact</b>	Rinse immediately with plenty of water, also under the eyelids, for at least 15 minutes
<b>Ingestion</b>	Never give anything by mouth to an unconscious person
<b>Inhalation</b>	Move to fresh air
<b>Notes to physician</b>	Treat symptomatically

<b>Suitable extinguishing media</b>	Dry chemical
<b>Special protective equipment for firefighters</b>	Wear self-contained breathing apparatus and protective suit

<b>Personal precautions</b>	Use personal protective equipment
<b>Methods for cleaning up</b>	Soak up with inert absorbent material

<b>Handling</b>	No special handling advice required
<b>Storage</b>	Keep in properly labelled containers

**Occupational exposure controls**

**Exposure limits**

dimethylsulfoxide	-	-	-	-
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<b>Engineering measures</b>	Ensure adequate ventilation, especially in confined areas
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**Personal protective equipment**

<b>Respiratory protection</b>	In case of insufficient ventilation wear suitable respiratory equipment
<b>Hand protection</b>	Protective gloves
<b>Eye protection</b>	Safety glasses with side-shields
<b>Skin and body protection</b>	Lightweight protective clothing
<b>Hygiene measures</b>	Handle in accordance with good industrial hygiene and safety practice
<b>Environmental exposure controls</b>	Prevent product from entering drains

**General Information**

<b>Form</b>	Liquid
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**Important Health Safety and Environmental Information**

<b>Boiling point/range</b>	°C No data available	°F No data available
<b>Melting point/range</b>	°C No data available	°F No data available
<b>Flash point</b>	°C No data available	°F No data available
<b>Autoignition temperature</b>	°C No data available	°F No data available
<b>Oxidizing properties</b>	No information available	
<b>Water solubility</b>	No data available	



<b>Stability</b>	Stable under normal conditions.
<b>Materials to avoid</b>	No information available
<b>Hazardous decomposition products</b>	No information available
<b>Polymerization</b>	Hazardous polymerisation does not occur



**Acute toxicity**

dimethylsulfoxide	14500 mg/kg (Rat)	No data available	No data available
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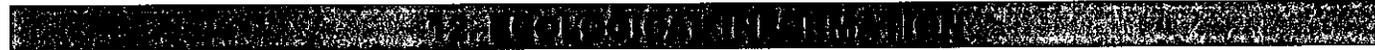
**Principle Routes of Exposure/  
Potential Health effects**

<b>Eyes</b>	May cause eye irritation with susceptible persons.
<b>Skin</b>	May cause skin irritation in susceptible persons.
<b>Inhalation</b>	No information available
<b>Ingestion</b>	No information available

**Specific effects**

<b>Carcinogenic effects</b>	No information available
<b>Mutagenic effects</b>	No information available
<b>Reproductive toxicity</b>	No information available
<b>Sensitization</b>	No information available

**Target Organ Effects** Eyes. Skin.



<b>Ecotoxicity effects</b>	No information available.
<b>Mobility</b>	No information available.
<b>Biodegradation</b>	No information available.
<b>Bioaccumulation</b>	No information available.



Dispose of in accordance with local regulations



**IATA**

<b>Proper shipping name</b>	Not classified as dangerous in the meaning of transport regulations
<b>Hazard Class</b>	No information available
<b>Subsidiary Class</b>	No information available
<b>Packing group</b>	No information available
<b>UN-No</b>	No information available

## 15. REGULATORY INFORMATION

### International Inventories

Chemical Name	TSCA	PICCS	ENCS	DSL	NDSL	AICS
dimethylsulfoxide	Listed	Listed	Listed	Listed	-	Listed

### U.S. Federal Regulations

**SARA 313**  
Not regulated

#### **Clean Air Act, Section 112 Hazardous Air Pollutants (HAPs) (see 40 CFR 61)**

This product contains the following HAPs:

### U.S. State Regulations

Chemical Name	Massachusetts - RTK	New Jersey - RTK	Pennsylvania - RTK	Illinois - RTK	Rhode Island - RTK
dimethylsulfoxide	-	-	-	-	-

#### **California Proposition 65**

This product contains the following Proposition 65 chemicals:

#### **WHMIS hazard class:**

D2B Toxic materials

This product has been classified according to the hazard criteria of the CPR and the MSDS contains all of the information required by the CPR

## 16. OTHER INFORMATION

This material is sold for research and development purposes only. It is not for any human or animal therapeutic or clinical diagnostic use. It is not intended for food, drug, household, agricultural, or cosmetic use. An individual technically qualified to handle potentially hazardous chemicals must supervise the use of this material.

The above information was acquired by diligent search and/or investigation and the recommendations are based on prudent application of professional judgment. The information shall not be taken as being all inclusive and is to be used only as a guide. All materials and mixtures may be present unknown hazards and should be used with caution. Since Invitrogen Corporation cannot control the actual methods, volumes, or conditions of use, the Company shall not be held liable for any damages or losses resulting from the handling or from contact with the product as described herein. THE INFORMATION IN THIS MSDS DOES NOT CONSTITUTE A WARRANTY, EXPRESS OR IMPLIED, INCLUDING ANY IMPLIED WARRANTY OF MERCHANTABILITY OR FITNESS FOR ANY PARTICULAR PURPOSE.

**End of Safety Data Sheet**

## 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, Japan, Hong Kong, Korea, 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.

### Cell Lines

<b>ATCC® Number:</b> HTB-133™ <a href="#">Order this item</a>		<b>Price:</b> \$185.00
<b>Designations:</b> T-47D		<b>Depositors:</b> I Keydar
<b>Biosafety Level:</b> 1		<b>Shipped:</b> frozen
<b>Medium &amp; Serum:</b> <a href="#">See Propagation</a>		<b>Growth Properties:</b> adherent
<b>Organism:</b> <i>Homo sapiens</i> (human)		<b>Morphology:</b> epithelial 
<b>Source:</b> <b>Organ:</b> mammary gland; breast <b>Tissue:</b> duct <b>Disease:</b> ductal carcinoma <b>Derived from metastatic site:</b> pleural effusion		
<b>Permits/Forms:</b> In addition to the <a href="#">MTA</a> 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 <a href="#">click here</a> for information regarding the specific requirements for shipment to your location.		
<a href="#">Related Cell Culture Products</a>		
<b>Receptors:</b>	calcitonin; androgen receptor, positive; progesterone receptor, positive; glucocorticoid; prolactin; estrogen receptor, positive calcitonin, expressed androgen receptor, positive, expressed estrogen receptor, positive, expressed progesterone receptor, positive, expressed glucocorticoid receptor, positive, expressed prolactin, expressed	
<b>Tumorigenic:</b>	Yes, forms colonies in soft agar	
<b>DNA Profile (STR):</b>	Amelogenin: X CSF1PO: 11,13 D13S317: 12 D16S539: 10 D5S818: 12 D7S820: 11 TH01: 6 TPOX: 11 vWA: 14	
<b>Cytogenetic Analysis:</b>	This is a hypotriploid human cell line. The modal chromosome number is 65 occurring at 50% and polyploidy at 0.8%. 18 marker chromosomes are common to most cells, of which 7 are paired and 11 are single-copied. The t(8q14q), t(9q17q), t(10q17p) are among 7 paired markers common to most cells. N7, N9, and N10 are absent and N11 is generally present in 4 copies. DM's occurred, but infrequently. Q-band examination did not show the presence of a Y chromosome.	
<b>Isoenzymes:</b>	AK-1, 1; ES-D, 2; G6PD, B; GLO-I, 1-2; PGM1, 1; PGM3, 1	
<b>Age:</b>	54 years adult	
<b>Gender:</b>	female	
<b>Comments:</b>	The cells express the WNT7B oncogene [PubMed: 8168088]. The T-47 line was isolated by I. Keydar from a pleural effusion obtained from a 54 year old female patient with an infiltrating ductal carcinoma of the breast. This differentiated epithelial substrain (T-47D) was found to contain cytoplasmic junctions and receptors to 17 beta estradiol, other steroids and calcitonin.	
<b>Propagation:</b>	<b>ATCC complete growth medium:</b> RPMI 1640 medium with 2 mM L-glutamine adjusted to contain 1.5 g/L sodium bicarbonate, 4.5 g/L glucose, 10 mM HEPES, and 1.0 mM sodium pyruvate and supplemented with 0.2 Units/ml bovine insulin, 90%; fetal bovine serum, 10% <b>Temperature:</b> 37.0C <b>Atmosphere:</b> air, 95%; carbon dioxide (CO2), 5%	
<b>Subculturing:</b>	<b>Protocol:</b> <ol style="list-style-type: none"> <li>1. Remove and discard culture medium.</li> <li>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.</li> <li>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.</li> <li>4. Add 6.0 to 8.0 ml of complete growth medium and aspirate cells by gently pipetting.</li> <li>5. Add appropriate aliquots of the cell suspension to new culture vessels.</li> <li>6. Incubate cultures at 37°C.</li> </ol> <b>Subcultivation ratio:</b> A subcultivation ratio of 1:3 to 1:5 is recommended <b>Medium renewal:</b> 2 to 3 times per week	
<b>Preservation:</b>	<b>Freeze medium:</b> Complete growth medium supplemented with 5% (v/v) DMSO <b>Storage temperature:</b> liquid nitrogen vapor phase	
<b>Doubling Time:</b>	32 hrs	
<b>Related Products:</b>	Recommended medium (without the additional supplements or serum described under ATCC Medium): ATCC <a href="#">30-2001</a> recommended serum: ATCC <a href="#">30-2020</a> purified DNA: ATCC <a href="#">HTB-133D</a> purified RNA: ATCC <a href="#">HTB-133R</a>	
<b>References:</b>	<a href="#">1120</a> : Judge SM, Chatterton RT Jr. Progesterone-specific stimulation of triglyceride biosynthesis in a breast cancer cell line (T-47D). Cancer Res. 43: 4407-4412, 1983. PubMed: <a href="#">6871874</a>	

## Cell Biology

ATCC® Number:	HTB-22™ <a href="#">Order this item</a>	Price
Designations:	MCF7	
Depositors:	CM McGrath	
Biosafety Level:	1	
Shipped:	frozen	
Medium & Serum:	<a href="#">See Propagation</a>	
Growth Properties:	adherent	
Organism:	<i>Homo sapiens</i> (human)	
Morphology:	epithelial	
		
Source:	<b>Organ:</b> mammary gland; breast <b>Disease:</b> adenocarcinoma <b>Derived from metastatic site:</b> pleural effusion <b>Cell Type:</b> epithelial	
Cellular Products:	insulin-like growth factor binding proteins (IGFBP) BP-2; BP-4; BP-5	
Permits/Forms:	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.	
Applications:	transfection host ( <a href="#">Nucleofection technology from Lonza</a> <a href="#">Roche FuGENE® Transfection Reagents</a> )	
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-1, 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:	<b>ATCC complete growth medium:</b> 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% . <b>Atmosphere:</b> air, 95%; carbon dioxide (CO2), 5% <b>Temperature:</b> 37.0°C	
Subculturing:	<b>Protocol:</b> 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. <b>Note:</b> 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.  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). <b>Note:</b> 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.	

<b>ATCC® Number:</b>	<b>CRL-10317™</b> <small>Order this Item</small>	<b>Price:</b>
<b>Designations:</b>	MCF 10A	
<b>Depositors:</b>	Michigan Cancer Foundation	
<b>Biosafety Level:</b>	1	
<b>Shipped:</b>	frozen	
<b>Medium &amp; Serum:</b>	<a href="#">See Propagation</a>	
<b>Growth Properties:</b>	adherent	
<b>Organism:</b>	<i>Homo sapiens</i> (human)	
<b>Morphology:</b>	epithelial	
<b>Source:</b>	<b>Organ:</b> mammary gland; breast <b>Disease:</b> fibrocystic disease <b>Cell Type:</b> epithelial	
<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>Isolation:</b>	<b>Isolation date:</b> August 22, 1984	
<b>Applications:</b>	transfection host ( <a href="#">Roche FuGENE® Transfection Reagents</a> )	
<b>Tumorigenic:</b>	No	
<b>DNA Profile (STR):</b>	Amelogenin: X CSF1PO: 10,12 D13S317: 8,9 D16S539: 11,12 D5S818: 10,13 D7S820: 10,11 TH01: 8,9,3 TPOX: 9,11 vWA: 15,17	
<b>Isoenzymes:</b>	AK-1, 1 <a href="#">[23084]</a> ES-D, 1 <a href="#">[23084]</a> G6PD, B <a href="#">[23084]</a> GLO-I, 1-2 <a href="#">[23084]</a> PGM1, 1-2 <a href="#">[23084]</a> PGM3, 1 <a href="#">[23084]</a>	
<b>Age:</b>	36 years	
<b>Gender:</b>	female	
<b>Ethnicity:</b>	Caucasian	
<b>Comments:</b>	The MCF 10A cell line is a non-tumorigenic epithelial cell line. <a href="#">[21968]</a> The line was produced by long term culture in serum free medium with low Ca++ concentration. <a href="#">[21968]</a> MCF 10A was derived from adherent cells in the population. <a href="#">[21968]</a> Cells derived from a floating population are available (see MCF 10F, ATCC CRL-10318). <a href="#">[21968]</a> The cells are positive for epithelial sialomucins, cytokeratins and milk fat globule antigen. <a href="#">[21968]</a> They exhibit three dimensional growth in collagen, and form domes in confluent cultures. <a href="#">[21968]</a> Thus far, the cells have shown no signs of terminal differentiation or senescence. The line is responsive to insulin, glucocorticoids, cholera enterotoxin, and epidermal growth factor (EGF). <a href="#">[21968]</a> By electron microscopy the cells display characteristics of luminal ductal cells but not of myoepithelial cells. <a href="#">[23085]</a> They also express breast specific antigens as detected by positive reaction with MFA-Breast and MC-5 monoclonal antibodies. <a href="#">[23085]</a> The calcium content of the medium exerts a strong effect on the morphology of the cells. <a href="#">[22248]</a>	
<b>Propagation:</b>	<b>ATCC complete growth medium:</b> The base medium for this cell line (MEBM) along with the additives can be obtained from Lonza/Clonetics Corporation as a kit: MEGM, Kit Catalog No. CC-3150. ATCC does not use the GA-1000 (gentamycin-amphotericin B mix) provided with kit. To make the complete growth medium, you will need to add the following components to the kit (sold separately):  <ul style="list-style-type: none"> <li>• 100 ng/ml cholera toxin</li> </ul> <p>Note: Do not filter complete medium <b>Temperature:</b> 37.0°C <b>Atmosphere:</b> air, 95%; carbon dioxide (CO2), 5%</p>	
<b>Subculturing:</b>	<b>Protocol:</b> Remove medium and rinse monolayer with PBS (ATCC Cat# 30-2200). Add 3.0 ml 0.05% trypsin, 0.53 mM EDTA and incubate at 37C for 15 minutes. To neutralize trypsin, add 3 ml solution of 0.1% soybean trypsin inhibitor. Centrifuge cell suspension at 125 xg for 5 to 10 minutes. Resuspend cell pellet in complete culture medium. Add appropriate aliquots of cell suspension to new culture vessels. <b>Subcultivation Ratio:</b> A subcultivation ratio of 1:3 to 1:4 is recommended <b>Medium Renewal:</b> Every 2 to 3 days	

# LISTED CELL LINE INFORMATION

Cell Biology:	CRL-1500™ <a href="#">Order this item</a>	Price:	\$
ATCC® Number:	ZR-75-1		F
Designations:	LW Engel		M
Depositors:	1		C
Biosafety Level:	frozen		M
Shipped:	<a href="#">See Propagation</a>		F
Medium & Serum:	adherent		M
Growth Properties:	<i>Homo sapiens</i> (human)		I
Organism:	epithelial		F
Morphology:			L
Source:	Organ: mammary gland; breast Tissue: duct Disease: ductal carcinoma Derived from metastatic site: ascites Cell Type: epithelial		
Cellular Products:	mucin (apomucin, MUC-1, MUC-2)		
Permits/Forms:	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.		
Receptors:	estrogen receptor, expressed		
Tumorigenic:	Yes		
DNA Profile (STR):	Amelogenin: X CSF1PO: 10,11 D13S317: 9 D16S539: 11 D5S818: 13 D7S820: 10,11 TH01: 7,9,3 TPOX: 8 vWA: 16,18		
Cytogenetic Analysis:	This human cell line has a hypertriploid chromosome number. The modal chromosome number was 72 occurring in 26% of the cells and the rate of higher ploidies was at 1.2%. Eighteen markers were common. They are: t(1q,?), M2,M3, del(3) (p21), der(5)t(5;?) (q35;?), del(6) (q21), t(11q14q), t(11;11) (p15;q11), der(14)t(2;14) (q21;q32), t(17q,?), M13, M14, M15, M16, M17, der(8)t(8;?) (t(9p,?) and 19pt. Of these M13 was paired. Normal N14 was not found. N6 was single copied, X had 3 copies and N18 had 4 copies in each cell.		
Isoenzymes:	G6PD, B		
Age:	63 years adult		
Gender:	female		
Ethnicity:	Caucasian		
Comments:	The cells produce high levels of MUC-1 mucin mRNA, low levels of MUC-2 mRNA but do not express the MUC-3 gene. [23102]		
Propagation:	<b>ATCC complete growth medium:</b> 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: fetal bovine serum to a final concentration of 10%. <b>Atmosphere:</b> air, 95%; carbon dioxide (CO2), 5% <b>Temperature:</b> 37.0°C		
Subculturing:	<b>Protocol:</b> <ol style="list-style-type: none"><li>1. Remove and discard culture medium.</li><li>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.</li><li>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.</li><li>4. Add 6.0 to 8.0 ml of complete growth medium and aspirate cells by gently pipetting.</li><li>5. Add appropriate aliquots of the cell suspension to new culture vessels.</li><li>6. Incubate cultures at 37°C.</li></ol> <b>Subcultivation Ratio:</b> A subcultivation ratio of 1:4 to 1:6 is recommended <b>Medium Renewal:</b> 2 to 3 times per week		
Preservation:	<b>Freeze medium:</b> Complete growth medium supplemented with 5% (v/v) DMSO <b>Storage temperature:</b> liquid nitrogen vapor phase		
Doubling Time:	80 hrs		

<b>ATCC<sup>®</sup> Number:</b>	<b>ATCC<sup>®</sup> Accession Number:</b>	<b>Price:</b>
<b>Designations:</b>	NIH:OVCAR-3	
<b>Depositors:</b>	R Ozols, TC Hamilton	
<b>Biosafety Level:</b>	1	
<b>Shipped:</b>	frozen	
<b>Medium &amp; Serum:</b>	<a href="#">See Propagation</a>	
<b>Growth Properties:</b>	adherent	
<b>Organism:</b>	<i>Homo sapiens</i> (human)	
<b>Morphology:</b>	epithelial	
		
<b>Source:</b>	<b>Organ:</b> ovary <b>Disease:</b> adenocarcinoma <b>Cell Type:</b> epithelial	
<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>Isolation:</b>	<b>Isolation date:</b> 1982	
<b>Applications:</b>	transfection host ( <a href="#">Roche FuGENE<sup>®</sup> Transfection Reagents</a> )	
<b>Receptors:</b>	androgen receptor, positive; estrogen receptor, positive; progesterone receptor, positive	
<b>Tumorigenic:</b>	Yes	
<b>DNA Profile (STR):</b>	Amelogenin: X CSF1PO: 11,12 D13S317: 12 D16S539: 12 D5S818: 11,12 D7S820: 10 TH01: 9,9,3 TPOX: 8 vWA: 17	
<b>Cytogenetic Analysis:</b>	The cell line is aneuploid human female, with chromosome counts in the sub to near-tetraploid 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.	
<b>Isoenzymes:</b>	AK-1, 1 ES-D, 1 G6PD, B GLO-I, 1 PGM1, 1 PGM3, 1	
<b>Age:</b>	60 years	
<b>Gender:</b>	female	
<b>Ethnicity:</b>	Caucasian	
<b>Comments:</b>	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.	
<b>Propagation:</b>	<b>ATCC complete growth medium:</b> 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%. <b>Temperature:</b> 37.0°C <b>Atmosphere:</b> air, 95%; carbon dioxide (CO <sub>2</sub> ), 5%	

<b>ATCC<sup>®</sup> Number:</b>	<b>C1279</b> <small>(click this item)</small>	<b>Price:</b>	
<b>Designations:</b>	Caov-3		
<b>Depositors:</b>	J Fogh		
<b>Biosafety Level:</b>	1		
<b>Shipped:</b>	frozen		
<b>Medium &amp; Serum:</b>	<a href="#">See Propagation</a>		
<b>Growth Properties:</b>	adherent		
<b>Organism:</b>	<i>Homo sapiens</i> (human)		
<b>Morphology:</b>	epithelial		
			
<b>Source:</b>	<b>Organ:</b> ovary <b>Disease:</b> adenocarcinoma		
<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>Restrictions:</b>	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.		
<b>Isolation:</b>	<b>Isolation date:</b> 1976		
<b>DNA Profile (STR):</b>	Amelogenin: X CSF1PO: 10,13 D13S317: 12 D16S539: 9 D5S818: 12 D7S820: 10 TH01: 7 TPOX: 8,10 vWA: 16,18		
<b>Isoenzymes:</b>	AK-1, 1 ES-D, 1 G6PD, B GLO-I, 1-2 Me-2, 2 PGM1, 1 PGM3, 1		
<b>Age:</b>	54 years		
<b>Gender:</b>	female		
<b>Ethnicity:</b>	Caucasian		
<b>Propagation:</b>	<b>ATCC complete growth medium:</b> 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%. <b>Atmosphere:</b> air, 95%; carbon dioxide (CO <sub>2</sub> ), 5% <b>Temperature:</b> 37.0°C		
<b>Subculturing:</b>	<b>Protocol:</b> <ol style="list-style-type: none"><li>1. Remove and discard culture medium.</li><li>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.</li><li>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.</li><li>4. Add 6.0 to 8.0 ml of complete growth medium and aspirate cells by gently pipetting.</li><li>5. Add appropriate aliquots of the cell suspension to new culture vessels.</li><li>6. Incubate cultures at 37°C.</li></ol> <b>Subcultivation Ratio:</b> A subcultivation ratio of 1:3 to 1:8 is recommended <b>Medium Renewal:</b> 2 to 3 times per week		

**Cell Biology****ATCC® Number:****HTB-77™** (Order this item)**Price:****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  
 TH01: 9,9,3  
 TPOX: 8,11  
 vWA: 17,18

**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;7;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.

**Propagation:**

**ATCC complete growth medium:** The base medium for this cell line is ATCC-formulated McCoy's 5a Medium Modified, Catalog No. 30-2007. To make the complete growth medium, add the following components to the base medium: fetal bovine serum to a final concentration of 10%.  
**Atmosphere:** air, 95%; carbon dioxide (CO<sub>2</sub>), 5%  
**Temperature:** 37.0°C





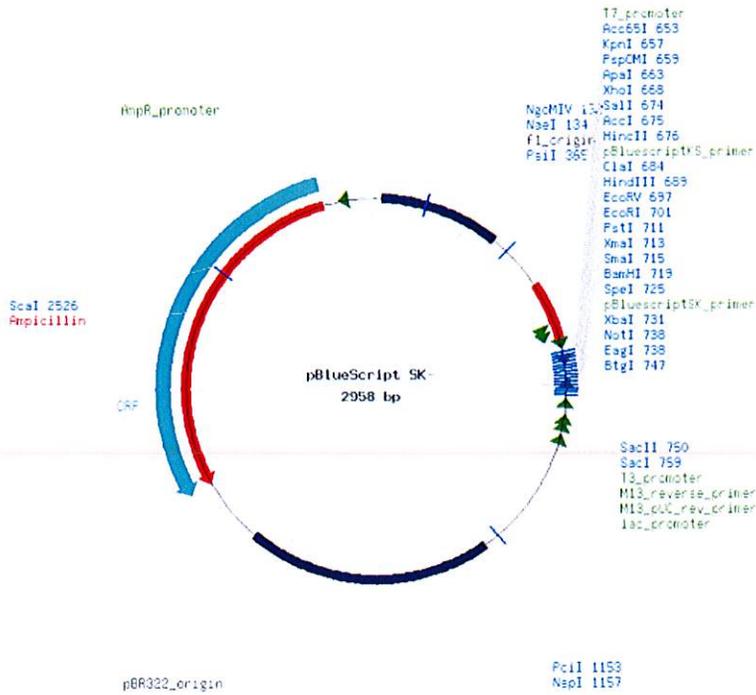
Community

Vector Database > pBlueScript SK-



Vector Database is a list of plasmid backbones from publications and several companies, including cloning, mammalian expression, bacterial expression, and lentiviral and retroviral plasmids. The database is compiled by [Addgene](#), and hosted on LabLife. LabLife does not sell or distribute any of the plasmids listed in this catalog.

pBlueScript SK-  
pBSK- , pBSSK  
Stratagene  
Bacterial  
3000  
M13/T7/T3  
Amp  
Discontinued  
[View Sequence](#)





Community

Vector Database > pcDNA3



Vector Database is a list of plasmid backbones from publications and several companies, including cloning, mammalian expression, bacterial expression, and lentiviral and retroviral plasmids. The database is compiled by [Addgene](#), and hosted on LabLife. LabLife does not sell or distribute any of the plasmids listed in this catalog.

pcDNA3  
Invitrogen  
Mammalian expression  
CMV  
5446  
T7  
Ampicillin  
Neomycin  
[View Sequence](#)

