

**THE UNIVERSITY OF WESTERN ONTARIO
BIOLOGICAL AGENTS REGISTRY FORM**
Approved Biohazards Subcommittee: October 14, 2010
Biosafety Website: www.uwo.ca/humanresources/biosafety/

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

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

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

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

PRINCIPAL INVESTIGATOR	<u>Andrew J. Watson</u>
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Location of experimental work to be carried out: Building(s) Victoria Research Labs
Room(s) 5TH Floor

*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: NSERC and CIHR
GRANT TITLE(S): NSERC The Maternal Environment- Influence On Early Mammalian Development; CIHR – Trophoctoderm Development and the Blastocyst

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

<u>Name</u>	<u>UWO E-mail Address</u>	<u>Date of Biosafety Training</u>
<u>Dr Michele Calder</u>	<u>Michele.Calder @Schulich. uwo.ca</u>	<u>Sept 11, 2007</u>
<u>Mr Paul Ricci</u>	<u>pricci@uwo.ca</u>	<u>July 14th 2009</u>
<u>Ms Danyka Belanger</u>	<u>danykabelanger@gmail.ca</u>	<u>August 24th 2011</u>
<u>Mrs Christine Bell</u>	<u>Christinewitchell@gmail.ca</u>	<u>August 24th 2011</u>
<u>Rucha Khanderkar</u>	<u>rkhande@uwo.ca</u>	<u>May 19th 2011</u>
<u>Matthew Nichols</u>	<u>Matthew.nichols@gmail.com</u>	<u>My 19th 2011</u>

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

Overview of Research Directions and Biohazardous Agents –Andrew J. Watson 2010

Our research is directed at defining the cellular and molecular mechanisms that support development to the blastocyst stage. All of our experiments are applied to preimplantation mouse and cow embryos. We have two funded projects, 1 from NSERC and 1 from the CIHR. The NSERC project is exploring the expression and function of a gene family of endoplasmic reticulum stress proteins during preimplantation development. The CIHR projects examine the mechanisms controlling development during the first week and focus on understanding the roles of gene families such as the Na/K-ATPase, tight junction, adherens junction, growth factors and mitogen activated protein kinase families. To conduct these research projects we must have a weekly supply of mouse and cow embryos. To accomplish this we order in mice (MFI, CDI, CFI; 50-100/week) from an outside supplier (Harlan, Charles River or Jackson labs) each week and set up small groups of these mice (ie 5 X 10-30 at a time) for injection with pregnant mares' serum gonadotrophin (PMSG; 5-10 i.u). This hormone stimulates the ovary to produce a larger than normal number of oocytes (10-12 is normal; up to 50 per mouse is possible with the hormone treatment). 46-48 hours after the PMSG injection the animals receive second sub-cutaneous injection of human chorionic gondaotrophin (hCG; 5-10 i.u.) This hormone induces ovulation of the ripening ovarian follicles. The hCG injected mice are place with males overnight for mating and copulation is determined by examining for the presence of a vaginal plug the following morning. Embryos of the appropriate stage (ie 1 cell to the blastocyst stage or day 1 to day 4 of development) are flushed from isolated reproductive tracts following appropriate euthanasia of the mice. This procedure will be repeated on a weekly basis each year to obtain early embryos which will then be used for our experimental needs. The bovine embryo production does not require animal use for we collect oocytes from slaughterhouse ovaries and place the the oocytes into culture for maturation prior to fertilization in culture using thawed bovine semen and then embryo production by culturing bovine zygotes to the appropriate stage for each experiment. We collect bovine ovaries from the slaughterhouse in Guelph, Ontario on a weekly basis for this purpose.

Procedures that we apply in our investigation of early development include; embryo culture; extraction of mRNAs, proteins, application of Rt-PCR and immunofluorescence analysis. We also microinject siRNAs (oligonucleotides) into preimplantation embryos to conduct loss of function experiments. We also employ a number of pharmacological blockers and activators that are routinely purchased from Sigma chemical company to examine the effects of blocking or activaing pathways on early development. These reagents include: MAPK inhibitors (SB203580; SB22025, PD erk blocker); AMPK blockers; growth factors (TGF-beta); TGF beta receptor blockers; ginseng extracts (obtained from Dr Ed Lui, Physiology and Pharmacology) In all cases these agents are used and discarded according to established protocols as outline on Materials Safett data sheets and UWO bioharzards waste disposal protocols.

The only cell lines or primary culture we would use include both mouse and cow primary oviductal epithelial cell cultures that are used for short term experiments to examine paracrine and autocrine regulation of early development. These cells are isolated from mouse and cow oviduct using trypsin and then primary cells are plated in 24 well plates at a density of 1 million cells per well to establish confluent monolayers that are maintained for up to one week. The base medium used is TCM-199 with 10% FBS.

The only established cell lines we might use for our studies are JEG-3 or BeWO choriocarcinoma cell lines. These established cell lines are obtained from our collaborator Dr Kaiping Yang and they are cultured under standard TCM-119 + 10% FBS conditions. The cells used as a model for trophoblast development. We do not modify them by exposing them to any biological agent. Cultures are established for experiments and frozen stocks are maintained.

We envision using trophoblast stem cells derived as primary cultures from mouse blastocysts as an experimental model as well. These cells are isolated from mouse blastocysts established into culture and maintained for periods of up to 2weeks in medium supplemented with FGF and LIF. Once again these culture will not be treated with any biological agents. They will simply be used to investigate gene expression and thus will have loss of function (siRNA) and gain of function (expression of cRNAs) approahes applied to them.

Please include a one page research summary or teaching protocol.

The Maternal Environment: Influence on Early Mammalian Development

Cultured mammalian embryos from all species have a reduced pregnancy rate following embryo transfer, display aberrant patterns and levels of gene expression, and are prone to metabolic and growth disorders. It is important to increase our understanding of the normal relationships that exist between the embryo and its external environment during the first few days of development and then use this information to improve embryo culture environments. **Principal Objective:** Our recent discoveries provide a foundation for our studies over the next 5 years which will focus on understanding the impact of the ER stress pathways on preimplantation development and whether activation of ER stress pathways is a critical component of the adaptive mechanism early embryos employ to offset the deleterious consequence of culture on their development. Our **hypothesis is that ER stress pathway activation is protective and enables cultured preimplantation embryos to proceed through the first week of development in vitro.**

The specific objectives are:

- 1) To define the protein localization patterns for *Bip*, *Atf4*, *Chop*, *Irelα*, *Xbp1*, *Ask1*, *Atf6*, *Gadd34*, *Atf3*, *Perk*, *Grp94* and *eIF2α* during preimplantation development.
- 2) To determine *eIF2α* phosphorylation during preimplantation development.
- 3) To investigate the mechanism of culture medium (glucose levels) induction of ER stress activation during preimplantation development.
- 4) To investigate the consequences of ER stress activation on expression of ER stress pathway constituents, antioxidant enzymes and ER stress pathway targets ie *XBP1*, *eIF2α*, *Atf4*, *Chop*, *Gadd34*
- 5) To determine the developmental consequences of down-regulating *Irelα*, *Atf6* and *Perk* during preimplantation development.
- 6) To determine if treatment with Salubrinal (SOL), and antioxidants (glutathione, catalase and ascorbic acid) improve development of preimplantation embryos in vitro and reduce culture induced ER stress.

Short-and Long Term Goals: The short-term goal is to provide an understanding of the roles that ER stress pathway activation exerts during early mammalian development and to explore its regulation. The long-term goal is to provide an understanding of the influences that the maternal and culture environments can place on the developing embryo and then apply this information to optimize embryo culture environments.

ER stress pathways: The endoplasmic reticulum (ER) controls the production and maturation of secretory and membrane proteins. Nascent, unfolded polypeptide chains enter the ER lumen where they are folded and undergo post-translational modification by ER chaperone proteins. Properly folded proteins are exported to the Golgi apparatus, while malformed or unfolded proteins are exported to the cytoplasm for degradation. The flux of proteins moving into the ER varies in response to a wide range of environmental and physiological conditions. Protein homeostasis is maintained within the ER when the influx of unfolded proteins does not exceed the efflux of properly folded proteins. If the amount of secretory proteins entering the ER exceeds the capacity of the ER machinery, unfolded proteins accumulate in the ER, forming protein aggregates. These aggregates are toxic and this imbalance is referred to as ER stress.

Significance: Research is required to define all of the vital components that support early development. Our experiments will uncover new information regarding the regulation of preimplantation development. This information is required to understand the effects of environmental mediators on embryonic development and to reveal the functional roles of ER stress pathways during preimplantation development.

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
	<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"/> 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"/> 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"/> 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"/> 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:

Cell Type	Is this cell type used in your work?	Source of Primary Cell Culture Tissue	AUS Protocol Number
Human	<input checked="" type="radio"/> Yes <input type="radio"/> No	hTR8 trophoblast cells, obtained from Dr P.K. Lala	Not applicable
Rodent	<input checked="" type="radio"/> Yes <input type="radio"/> No	Oviduct, Trophoblast Stem Cells, embryonic stem cells	2006-012-02
Non-human primate	<input type="radio"/> Yes <input checked="" type="radio"/> No <i>SAW</i>		
Other (specify) cow	<input checked="" type="radio"/> Yes <input type="radio"/> No	Oviduct obtained from the University of Guelph along with cow ovaries. They obtain them from a local abattoir near Guelph.	

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

Cell Type	Is this cell type used in your work?	Specific cell line(s)*	Containment Level of each cell line	Supplier / Source of cell line(s)
Human	<input checked="" type="radio"/> Yes <input type="radio"/> No	JEG3, BeWo, JAR hTR8 human trophoblast cells	1	Dr Kaiping Yang- these are established cells lines that have been maintained in Dr Yang's laboratory for nearly 20 years. They were originally derived from choriocarcinomas and have now been distributed around the world for use in research labs investigating trophoblast biology. Dr Lala will supply the hTR8 cell line
Rodent	<input type="radio"/> Yes <input checked="" type="radio"/> No <i>AW</i>			
Non-human primate	<input type="radio"/> Yes <input checked="" type="radio"/> No <i>AW</i>			
Other (specify) cow	<input checked="" type="radio"/> Yes <input type="radio"/> No	Primary oviduct epithelial cell cultures		Obtained from University of Guelph; they obtain them from a local abattoir

*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 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		<input type="radio"/> Yes <input type="radio"/> Unknown		<input type="radio"/> 1 <input type="radio"/> 2 <input type="radio"/> 2+ <input type="radio"/> 3
Human Blood (fraction) or other Body Fluid		<input type="radio"/> Yes <input type="radio"/> Unknown		<input type="radio"/> 1 <input type="radio"/> 2 <input type="radio"/> 2+ <input type="radio"/> 3
Human Organs or Tissues (unpreserved)		<input type="radio"/> Yes		<input type="radio"/> 1 <input type="radio"/> 2 <input type="radio"/> 2+ <input type="radio"/> 3

		<input type="radio"/> Unknown		
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

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

** Please attach a plasmid map.

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

YES, complete table below NO

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

* Please attach a Material Safety Data Sheet or equivalent.

4.4 Will genetic sequences from the following be involved?

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

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 _____

6.3 AUS protocol # _____

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:

7.0 Use of Animal species with Zoonotic Hazards

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

7.2 Will live animals be used? YES No

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

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

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

8.0 Biological Toxins

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

8.2 If YES, please name the toxin(s) 2 hormones: pregnant mare's serum gonadotrophin; and human chorionic gonadotrophin

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

8.3 What is the LD₅₀ (specify species) of the toxin See info attached

8.4 How much of the toxin is handled at one time*? 75-100 iU

8.5 How much of the toxin is stored*? 5000 iU at a time

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

*For information on biosecurity requirements, please see:

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

9.0 Insects

9.1 Do you use insects? YES NO If no, please proceed to Section 10.0

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

9.3 What is the origin of the insect? _____

9.4 What is the life stage of the insect? _____

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

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

9.7 Do you use insects that require a permit from the CFIA permit? YES NO
If YES, Please attach the CFIA permit & describe any CFIA permit conditions:

10.0 Plants

10.1 Do you use plants? YES NO If no, please proceed to Section 11.0

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

10.3 What is the origin of the plant? _____

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

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

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

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

10.8 Is the CFIA permit attached? YES NO
If YES, Please attach the CFIA permit & describe any CFIA permit conditions:

11.0 Import Requirements

11.1 Will any of the above agents be imported? YES, please give country of origin _____ NO
If no, please proceed to Section 12.0

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

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

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

12.0 Training Requirements for Personnel Named on Form

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

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

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

SIGNATURE 

13.0 Containment Levels

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

13.2 Has the facility been certified by OHS for this level of containment?
 YES, date of most recent biosafety inspection: _____
 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

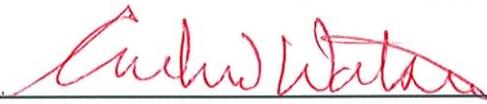
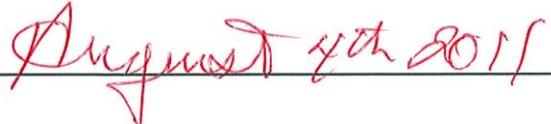
14.1 Please describe additional risk reduction measures will be taken beyond containment level 1, 2, 2+ or 3 measures, that are unique to this agent.

N/A

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:

we will follow established protocols; the stick will be washed thoroughly; bleeding if any stopped by compression; the subject will be advised to seek medical assistance; and an incident report will be filled out and submitted _____

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

SIGNATURE  Date: 

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: _____
Date: _____

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

Special Conditions of Approval:

JEG-3 (Human placental choriocarcinoma cell line) Nuclear Lysate (ab14841) | Abcam

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Products Kits/Lysates/Other >> Lysates >> Nuclear Lysates >> Human >> Other

JEG-3 (Human placental choriocarcinoma cell line) Nuclear Lysate (ab14841) [Preview our new beta datasheet](#)
[Give us your feedback!](#)

Product Name	JEG-3 (Human placental choriocarcinoma cell line) Nuclear Lysate
Product type	Lysates
Description	JEG-3 (Human placental choriocarcinoma cell line) Nuclear Lysate
Tested applications (see key)	EMSA, WB
Application notes (see key)	<p>Recommended dilutions</p> <p>Extracts have been quality control tested by Western blot and the Electrophoretic Mobility Shift Assay (EMSA).</p> <p>Not tested in other applications.</p> <p>Optimal dilutions/concentrations should be determined by the end user.</p>
Research areas	Kits/Lysates/Other >> Lysates >> Nuclear Lysates >> Human >> Other
Relevance	The human placenta is a highly invasive-tumor-like structure in which a subpopulation of placental trophoblast cells known as the "extravillous trophoblast" (EVT) invades the uterine decidua and its vasculature to establish adequate fetal-maternal exchange of molecules. In vitro-propagated non-lived EVT cell lines such as JEG-3 cells are used to study molecular mechanisms responsible for this invasiveness, which are identical to those of cancer cells. However, unlike cancer cells, their proliferation, migration, and invasiveness in situ are stringently controlled by decidua-derived transforming growth factor (TGF)-beta. These epithelial-trophoblast-like cells are therefore used in identifying genetic changes underlying epithelial tumor progression as well as gonadotropin secretion studies.
Storage buffer	Preservative: None Constituents: 20% Glycerol, 20mM HEPES, 100mM Potassium Chloride, 1mM Magnesium Chloride, 0.5mM PMSF, 0.5mM DTT, pH 7.9
Form	Liquid
Concentration	5 000 mg/ml
Storage instructions	Shipped at 4°C. Upon delivery aliquot and store at -20°C or -80°C. Avoid repeated freeze/thaw cycles.
Notes	We recommend aliquoting the extracts into single use fractions and then storing them.

At Abcam, we have one centralized database to hold all of our product information, so that everything we know about this JEG-3 (Human placental choriocarcinoma cell line) Nuclear Lysate is on this datasheet. But please do contact us if you would like any reassurance!

[Customer reviews \(feedback\) regarding JEG-3 \(Human placental choriocarcinoma cell line\) Nuclear Lysate](#)

[Customer FAQs regarding JEG-3 \(Human placental choriocarcinoma cell line\) Nuclear Lysate](#)

[Protocols for JEG-3 \(Human placental choriocarcinoma cell line\) Nuclear Lysate](#)

[Price and availability of products related to JEG-3 \(Human placental choriocarcinoma cell line\) Nuclear Lysate](#)

All products are "FOR RESEARCH USE ONLY AND ARE NOT INTENDED FOR DIAGNOSTIC OR THERAPEUTIC USE"

MERCK ANIMAL HEALTH » Chorulon® (MERCK ANIMAL HEALTH)

MERCK ANIMAL HEALTH

Intervet Canada Corp.

16750 ROUTE TRANSCANADIENNE, KIRKLAND, QC, H9H 4M7

Order Desk: 514-428-7013

Toll-Free: 866-683-7838

Fax: Toll-free 888-498-4444; local 514-428-7014

Website: www.merck-animal-health.ca

**MERCK**
Animal Health

Every effort has been made to ensure the accuracy of the information published. However, it remains the responsibility of the readers to familiarize themselves with the product information contained on the Canada product label or package insert.

CHORULON®*Merck Animal Health***Chorionic Gonadotropin Injection**

Freeze-Dried Cake

DIN 00781851

FOR VETERINARY USE ONLY**DESCRIPTION:**

Chorulon is presented as a white freeze-dried crystalline cake containing either 5 000 or 10 000 USP units

INDICATIONS:

As an aid in the treatment of the following fertility problems in cows and heifers:

Cystic ovary syndrome: anoestrus, prolonged oestrus and/or nymphomania due to cystic ovaries.

Active ingredient:

Chorionic gonadotrophin per vial, together with solvent (5 ml or 10 ml). Concentration after reconstitution is 1 000 USP units per 1 ml.

ACTION

Chorulon (Human chorionic gonadotrophin) is a complex glycoprotein capable of supplementing and being substituted for the luteinizing hormone secreted by the anterior pituitary gland in both the maturation of the follicle, ovulation and the formation of the corpus luteum. In the male Chorulon stimulates the interstitium to the production of testosterone and thus influences the development and maintenance of primary and secondary male sex characteristics. These properties make Chorulon suitable for the treatment of several fertility problems in domestic animals due to deficiencies of natural luteinizing hormone (LH) or interstitial cell stimulating hormone (ICSH).

RECONSTITUTION:

To prepare for use, add 5 ml of diluent into the vial containing 5 000 units USP or 10 ml of diluent into the vial containing 10 000 units USP. Shake thoroughly until the suspension is uniform after which the injection may be given. After reconstitution, the solution should be used immediately or within a few hours. Refrigeration after reconstitution is recommended.

DOSAGE AND ADMINISTRATION:

Animal species	Indications	
Cow, Heifer	Cystic ovary syndrome: anoestrus, prolonged oestrus and/or nymphomania due to cystic ovaries	2 500 USP units I.V. (2.5 mL) 10 000 USP units I.M. (10 mL)

DURATION:

Dosage may be repeated in 21 days if necessary.

WARNINGS:

Keep out of reach of children. Wash hands immediately and thoroughly after applying Chorulon or wear gloves. If accidental skin contact occurs, wash the affected area immediately with soap and water. In case of accidental self-injection, seek medical attention.

Toxin Info

CAUTIONS:

In rare cases as with all protein preparations incidents may occur shortly after injection. Epinephrine HCL given intravenously or intramuscularly when symptoms appear is the standard treatment.

STORAGE INSTRUCTIONS:

Do not store above 25°C. Protect from light. Keep from freezing. After reconstitution the product may be stored at 2-8°C for no longer than 24 hours.

PRESENTATION: 5 X 5 mL 5 000 USP and 1 x 10 mL 10 000 USP

INTERVET CANADA CORP., Kirkland, QC H9H 4M7

1 888 306-0069

Rev. 23NOV2009

NAC No.: 12081343

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MERCK ANIMAL HEALTH » Folligon® (MERCK ANIMAL HEALTH)

MERCK ANIMAL HEALTH

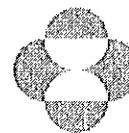
Intervet Canada Corp.

16750 ROUTE TRANSCANADIENNE, KIRKLAND, QC, H9H 4M7

Order Desk: 514-428-7013

Toll-Free: 866-683-7838

Fax: Toll-free 888-498-4444; local 514-428-7014

Website: www.merck-animal-health.ca**MERCK**
Animal Health

Every effort has been made to ensure the accuracy of the information published. However, it remains the responsibility of the readers to familiarize themselves with the product information contained on the Canada product label or package insert.

FOLLIGON®**Merck Animal Health****Pregnant Mare Serum Gonadotropin 5000 Injection IU**

Freeze-Dried Cake

FOR VETERINARY USE ONLY

DIN 00806285

DESCRIPTION:

Each vial contains Pregnant Mare serum gonadotrophin 5,000 I.U.

Pregnant mare serum gonadotrophin (PMSG) is predominantly follicle stimulating. Due to its FSH-activity, serum gonadotrophin stimulates the growth of the interstitial cells of the ovary as well as the growth and maturation of the follicles.

ACTIVE INGREDIENT:

Each vial contains Pregnant Mare serum gonadotrophin 5,000 I.U.

INDICATIONS:

As an aid to stimulate follicle development in functional ovaries in Cattle, Pig and Sheep.

DIRECTIONS FOR RECONSTITUTION:

Use a sterile syringe. Add 25 ml of the sterile diluent for injection U.S.P. to the vial containing the dry powder to obtain a clear solution containing 200 IU/ml of PMSG. After reconstitution, the solution should be used immediately or within a few hours.

DOSAGE AND ADMINISTRATION:

Cattle 1000 to 1250 iu.

Pigs 750 to 1250 iu.

Sheep 300 to 1000 iu.

Administer intramuscularly, intravenously, or subcutaneously.

WARNINGS:

Treated animals must not be slaughtered for use in food for at least 7 days after the latest treatment.

Keep out of reach of children.

If accidental skin contact occurs, wash the affected area immediately with soap and water. In case of accidental self-injection, seek medical attention.

Wash hands immediately and thoroughly after applying Folligon or wear gloves. If accidental skin contact occurs, wash the affected area immediately with soap and water. In case of accidental self-injection, seek medical attention.

CAUTION:

In case of anaphylactic reaction administer epinephrine.

STORAGE INSTRUCTIONS:

Do not store above 25°C. Protect from light. Protect from freezing. After reconstitution the product may be stored at 2-8°C for no longer than 24 hours.

PACKAGING:

Vial containing 5,000 IU of PMSG.

25 ml vial of diluent containing 2 mg methylparaoxybenzoate/ml (0.2%).

INTERVET CANADA CORP., Kirkland, QC H9H 4M7

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TOXIN USE RISK ASSESSMENT

Name of Toxin:	Human chorionic gonadotropin (hCG)
Proposed Use Dose:	10 µg
Proposed Storage Dose:	500 µg
LD₅₀ (species):	36000 µg

Calculation:	
36000 µg/kg	x 50 kg/person
Dose per person based on LD ₅₀ in µg = 1800000	
LD₅₀ per person with safety factor of 10 based on LD₅₀ in µg = 180000	

Comments/Recommendations:

Chronic exposure, reproductive hazard
Dose: 36mg/kg
Effects on fertility.
Species: Woman
Source: Sigma MSDS



TOXIN USE RISK ASSESSMENT

Name of Toxin:	Pregnant mare's serum gonadotropin (PMSG)
Proposed Use Dose:	10 µg
Proposed Storage Dose:	500 µg
LD ₅₀ (species):	1200000 µg

Calculation:	
1200000 µg/kg	x 50 kg/person
Dose per person based on LD ₅₀ in µg = 60000000	
LD₅₀ per person with safety factor of 10 based on LD₅₀ in µg =	6000000

Comments/Recommendations: