Modification Form for Permit BIO-UWO-0062

Permit Holder: Carole Crewchet

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

Najwa Zebian

Jeffrey Lester

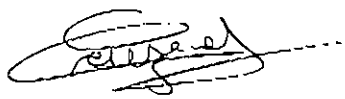
Matthew McCallum

Anthony Wong

Rachel Ford

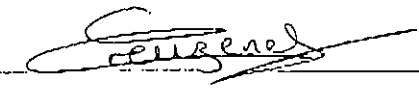
Additional Personnel
(Please list additional personnel here)

	Please stroke out any approved Biohazards to be removed below	Write additional Biohazards for approval below. Give the full name - do not abbreviate.
Approved Microorganisms	Helicobacter pylori, Campylobacter jejuni, Yersinia pseudotuberculosis, Escherichia coli JM 109, DH5alpha	
Approved Primary and Established Cells	Human [established]:stomach Rodent [established]: blood	
Approved Use of Human Source Material		
Approved Genetic Modifications (Plasmids/Vectors)	[plasmids]: pET23, pBluescriptSK	Plasmid: pRK793 From Addgene Order # 63655
Approved Use of Animals		
Approved Biological Toxin(s)		



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

As the principal investigator, I have ensured that all of the personnel named on the form have been trained. I will ensure that this project will follow the Western Biosafety Guidelines and Procedures Manual for Containment Level 1 2 Laboratories (and the Level 3 Facilities Manual for Level 3 projects). I will ensure that UWO faculty, staff and students working in my laboratory have an up-to-date Hazard Communication Form, found at http://www.wph.uwo.ca.

Signature of Permit Holder: 

Current Classification: 2 **Containment Level for Added Biohazards:** 1

Date of Last Biohazardous Agents Registry Form: Dec 23, 2010

Date of Last Modification (if applicable):

BioSafety Officer(s):

Chair, Biohazards Subcommittee: **Date:**

We are ordering plasmid pRK793 from Addgene so that we can over-express and purify the TEV protease from it. This protease will be used to clip off tags that are used at the N or C-termini of other proteins that we express, and facilitate their purification by affinity chromatography. However, the tags often prevent the activity of the proteins, so we engineered a TEV cleavage site between the tags and the proteins of interest so that the tags can be removed enzymatically using the TEV protease.
The plasmid will be preserved as a pure preparation at -20oC as well as within a standard E. coli cloning strain (DH5a) for propagation purposes (stored at -80oC).

This is only a plasmid that does not encode a toxin. As such, there is no MSDS sheet or equivalent available.

addgene

Find this plasmid at: www.addgene.org
Enter "8827" in the search box

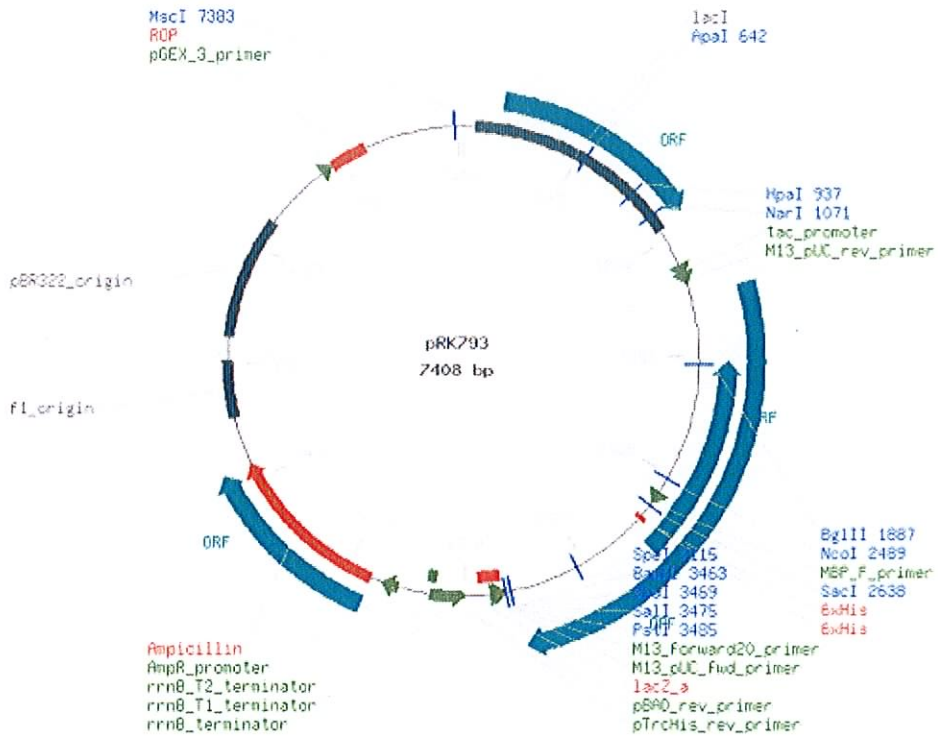
Plasmid 8827: pRK793

Gene/insert name:	TEV protease, S219V mutant
Alternative names:	tobacco etch virus protease
Insert size (bp):	750
Species of gene(s):	Other
Relevant mutations/deletions:	S219V mutation
Fusion proteins or tags:	His
Terminal:	N terminal on insert
Fusion proteins or tags:	polyarginine
Terminal:	C terminal on insert
Fusion proteins or tags:	MBP (with TEV)
Terminal:	N terminal on insert
Vector backbone:	pMal-C2 (Search Vector Database)
Backbone manufacturer:	New England Biolabs
Type of vector:	Bacterial expression
Backbone size (bp):	6700
Cloning site 5':	SacI
Site destroyed during cloning:	No
Cloning site 3':	BamHI
Site destroyed during cloning:	No
5' Sequencing primer:	N/A (List of Sequencing Primers)
3' Sequencing primer:	M13-F20
Bacteria resistance:	Ampicillin for p793; Chloramphenicol for pRIL.
High or low copy:	Low Copy
Grow in standard E. coli @ 37C:	No
Please specify bacterial strain for growth and growth condition:	Shifting the temperature from 37C to 30C during induction maximizes the yield of soluble TEV protease.
If you **did not originally clone** this gene, from whom and where did you receive the plasmid used to derive this plasmid:	ATCC
Sequence:	Visit www.addgene.org/8827
Plasmid Provided In:	BL21(DE3)-RIL
Principal Investigator:	David S. Waugh

Please check www.addgene.org/8827 for updated plasmid information and related links.

addgene

Find this plasmid at: www.addgene.org
Enter "8827" in the search box



Please check www.addgene.org/8827 for updated plasmid information and related links.

addgene

Find this plasmid at: www.addgene.org
Enter "8827" in the search box

Plasmid 8827: pRK793

Comments: Bacterial strain: E.coli BL21(DE3)-RIL (Stratagene).

pRK793 overproduces the catalytic domain of tobbaco etch virus (TEV) protease in the form of an MBP fusion protein that cleaves itself in vivo to yield a TEV protease catalytic domain with an N-terminal His-tag and a C-terminal polyarginine tag.

Article: Tobacco etch virus protease: mechanism of autolysis and rational design of stable mutants with wild-type catalytic proficiency. Kapust RB et al. (Protein Eng. 2001 Dec . 14(12):993-1000. Pubmed)

Please acknowledge the principal investigator and cite this article if you use this plasmid in a publication.

Also, please include the text "Addgene plasmid 8827" in your Materials and Methods section. This information allows Addgene to create a link from the plasmid page to your publication.

Please check www.addgene.org/8827 for updated plasmid information and related links.

THE UNIVERSITY OF WESTERN ONTARIO
BIOLOGICAL AGENTS REGISTRY FORM
Approved Biohazards Subcommittee: July 9, 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	Dr Carole CREUZENET
DEPARTMENT	Microbiology and Immunology
ADDRESS	DSB 3031
PHONE NUMBER	661 3204
EMERGENCY PHONE NUMBER(S)	519 672 8888
EMAIL	ccreuzen@uwo.ca

Location of experimental work to be carried out: Building(s) _____DSB_____ Room(s)_3031

*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:__CIHR, NSERC, Canadian Association of gastroenterology_____
GRANT TITLE(S):
CIHR: Protein glycosylation and virulence factors in *Helicobacter pylori*
CAG: Function and role in virulence of novel cysteine-rich proteins and associated chaperones from *Helicobacter pylori*
NSERC: Biosynthesis and function of novel modified heptoses present in bacterial surface polysaccharides.

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

Name	UWO E-mail Address	Date of Biosafety Training
Rachel Ford	rford5@uwo.ca	23 July 2009
Anthony Wong	awong392@uwo.ca	12 Sept 2009
Matthew McCallum	mmccal22@uwo.ca	16 June 2009
Jeffrey Lester	jlester3@uwo.ca	26 Oct 2010
Najwa Zebian	nzebian2@uwo.ca	2 Oct 2013

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.

In our search for genes determining virulence properties of Helicobacter pylori, including the protein glycosylation genes, we disrupt candidate genes and analyze phenotypic differences between the resulting mutants and the original wild-type strain in vitro.

In our search of genes responsible for deoxyheptose biosynthesis in Campylobacter jejuni and Yersinia pseudotuberculosis, we have disrupted several candidate genes and assessed the phenotypic differences in vitro. The information gathered helps us reconstruct a biosynthesis pathway for these unusual sugars. Not present in mammals, these sugars and the enzymes responsible for their synthesis represent novel antibiotic targets.

We also produce large amount of recombinant proteins and enzymes. For this, we use a variety of E. coli non-pathogenic lab strains to over-express the protein.

All the bacteria that we use are level 2 biosafety agents. To dispose of samples, all waste (liquid, or plasticware that was in contact with level 2 pathogens) is autoclaved by the dedicated departmental agent using a certified autoclave. All our glass pipette soak in decontaminating solution immediately after use and are further soaked in bleach before going to the wash.

Please include a one page research summary or teaching protocol.

CIHR: *Helicobacter pylori* is a highly prevalent human gastric pathogen that causes a considerable burden on the health care system. Like the closely related *Campylobacter jejuni*, *H. pylori* produces glycoproteins (GPs). My research focuses on the molecular basis for the production of GPs which contribute to bacterial virulence. The sugars present on GPs are not found in humans. Hence, our long term goal is to find inhibitors of the biosynthesis of these sugars which might serve as therapeutics against *H. pylori* and related bacteria. Our work will also allow assessing the enzymes involved as glyco-engineering tools, to produce carbohydrate epitopes with medical applications.

Based on our published and preliminary data, our hypothesis is that glycosylation of proteins other than flagellins occurs in *H. pylori* and affects bacterial virulence. Our immediate goals are to: 1/ Inventory the glycoproteins and associated sugars in *H. pylori*. 2/ Assess the role of a novel glycosylation pathway in virulence. 3/ Decipher how the currently known glycosylation pathway affects the production of multiple virulence factors in *H. pylori*.

Our long-term goal is to identify novel approaches to inhibit the production of virulence factors for therapeutic development. Targeting protein glycosylation for this purpose is novel and promising since (i) bacterial protein glycosylation uses machineries not present in humans that can be targeted by specific inhibitors, and (ii) the *H. pylori* protein glycosylation machinery affects multiple GPs and also controls the production of other (non GP) virulence factors. Therefore, inhibiting protein glycosylation should be very efficient at decreasing bacterial virulence while concomitantly limiting the appearance of resistance. This research will yield highly needed therapeutic targets, and will also provide fundamental information about bacterial protein glycosylation and the molecular basis for virulence in *H. pylori*.

CAG: The "Helicobacter Cysteine-rich Protein" (Hcp) family is a family of 7 proteins of unknown function specific to Helicobacters and Campylobacters. Several of them have been shown to elicit strong immune responses and to be secreted. Also, they have a signal peptide for secretion through the periplasm. All this suggests that these proteins may interact with periplasmic proteins of the Dsb (DiSulfide Bond) family prior to final secretion. This would allow them to acquire their proper structure via formation of appropriate disulfide bonds between their multiple cysteines.

Based on the preliminary data that are described in the research proposal, we hypothesize that specific Dsb proteins are involved in the folding and secretion of Hcp proteins, and that the Hcps and the associated Dsb proteins may play a role in the virulence of *H. pylori*.

Our long term goal is to exploit Hcp and Dsb proteins as novel therapeutic targets against *H. pylori*. This requires a better understanding of the mechanisms that supports the folding / secretion of Hcps and of their role for host / bacteria interactions.

This research is significant as it targets two types of proteins (Hcp and Dsb) that may be involved in bacterial virulence, and it will determine if they are good therapeutic targets. We will follow up with translational research to turn our discoveries into practical applications to control *H. pylori* infections, and help decrease the incidence of *H. pylori*–induced gastric cancer.

NSERC: Some bacteria cover their surface with long chains of sugars that affect their interactions with host cells and their ability to survive to adverse growth conditions encountered in the environment or during the colonization of a new host. These sugars can sometimes be very unusual. Their exact function and the mechanism whereby they affect interactions with host cells are not known. To better understand their function, it is necessary to identify the genes that are necessary for their synthesis, as well as to characterize the enzymes that are encoded by the genes. We are focusing on unusual heptoses that are found on the surface of several bacteria (Yersiniae, Campylobacters) and have made great progress in elucidating their biosynthesis and function using Yersiniae as a model system. We propose to pursue our studies using Campylobacters as the unusual heptoses are part of a different type of surface macromolecule in Campylobacters. The fundamental knowledge gathered in our studies will help us understand how a specific sugar can affect the function of a macromolecule, a question that is at the heart of the field of glycobiology and that applies not only to bacteria but also to plants and animals. We also plan to perform extensive structure/function studies of the enzymes that we have identified as this will provide novel tools to engineer novel sugar derivatives that could have technological applications

1.0 Microorganisms

1.1 Does your work involve the use of biological agents? x YES O 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? x YES (import permit) O NO
If YES, please give the name of the species. Helicobacter pylori, Campylobacter jejuni, Yersinia was from dr Skurnik (Finland)
What is the origin of the microorganism(s)? _CJ was ATCC, HP was other Canadian researchers, YP___
Please describe the risk (if any) of escape and how this will be mitigated: __No risk of escape. All cultures are in sealed tubes/flasks because of the necessary microaerophilic atmosphere. When we process samples, the left overs are always autoclaved before disposal.

Please attach the CFIA permit. A2001-02030-4; A2002-00996; A2002-01088-4.
Please describe any CFIA permit conditions:
_none_____
_____
_____

1.2 Please complete the table below:

Name of Biological agent(s)*	Is it known to be a human pathogen? YES/NO	Is it known to be an animal pathogen? YES/NO	Is it known to be a zoonotic agent? YES/NO	Maximum quantity to be cultured at one time? (in Litres)	Source/ Supplier	PHAC or CFIA Containment Level
Helicobacter pylori	x Yes O No	O Yes x No	O Yes x No	1	Dr Susan Logan (NRC, Ottawa). Dr Diane Taylor (Formerly U of Alberta, now retired)	O 1 x 2 O 2+ O 3
Campylobacter jejuni	x Yes O No	O Yes x No	x Yes O No	1	ATCC	O 1 x 2 O 2+ O 3
Yersinia pseudotuberculosis	x Yes O No	O Yes x No	O Yes x No	1	Dr Skurnik, Finland	O 1 2 2 O 2+ O 3
Escherichia coli JM109, DH5alpha	O Yes x No	O Yes x No	O Yes x No	10	Dr Lam (U of Guelph).	x 1 O 2 O 2+ O 3

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

2.0 Cell Culture

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

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

Cell Type	Is this cell type used in your work?	Source of Primary Cell Culture Tissue	AUS Protocol Number

Human	O Yes x No		Not applicable
Rodent	O Yes x No		
Non-human primate	O Yes x No		
Other (specify)	O Yes x No		

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

Cell Type	Is this cell type used in your work?	Specific cell line(s)*	Supplier / Source
Human	x Yes O No	Stomach (we get an immortalized cell line, AGS)	Dr Sherman, UofToronto
Rodent	x Yes O No	Blood (macrophages)	Dr Valvano UWO
Non-human primate	O Yes x No		
Other (specify)	O Yes x No		

*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 x 1 O 2 O 2+ O 3

3.0 Use of Human Source Materials

3.1 Does your work involve the use of human source materials? O YES x 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/NO**	**Name of Infectious Agent (If applicable)**	**PHAC or CFIA Containment Level (Select one)**
Human Blood (whole) or other Body Fluid		O Yes O Unknown		O 1 O 2 O 2+ O 3
Human Blood (fraction) or other Body Fluid		O Yes O Unknown		O 1 O 2 O 2+ O 3
Human Organs or Tissues (unpreserved)		O Yes O Unknown		O 1 O 2 O 2+ O 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? x YES O NO If no, please proceed to Section 5.0

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

Bacteria Used for Cloning *	Plasmid(s) **	Source of Plasmid	Gene Transfected	Describe the change that results from transformation or tranfection

[illegible]	[illegible]	[illegible]	Enzymes that modify sugars: CJ1427, CJ1428, CJ1429, Cj1430, HP0366, HP0840, WcbK, WcaG, DmhA, DmhB, Cj1121c, Cj1123c, CJ1293, CJ1293, WbpM. Proteins involved in secretion: HP0235, HP0231, HP0253, HP0879.	[illegible]

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

** Please attach a plasmid map.

4.3 Will genetic modification(s) involving viral vectors be made? O YES, complete table below x 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 O YES, please specify _____ x NO
- HTLV 1 or 2 or genes from any Level 1 or Level 2 pathogens O YES, specify _____ x NO
- SV 40 Large T antigen O YES x NO
- E1A oncogene O YES x NO
- Known oncogenes O YES, please specify _____ x NO
- Other human or animal pathogen and or their toxins O YES, please specify _____ x NO

4.5 Will virus be replication defective? O YES O NO

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

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

5.0 Human Gene Therapy Trials

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

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

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

5.3 How will the biological agent be administered? _____

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

5.5 Has human ethics approval been obtained? O YES, number: _____ O NO O PENDING

6.0 Animal Experiments

6.1 Will live animals be used? O YES x 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 O YES, specify: _____ O NO

6.5 Will the agent(s) be shed by the animal: O YES O 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)? O YES x No If no, please proceed to section 8.0

7.2 Please specify the animal(s) used:

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

8.0 Biological Toxins

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

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

8.3 What is the LD_{50} (specify species) of the toxin_____

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

8.5 How much of the toxin is stored*? _____

8.6 Will any biological toxins be used in live animals? O YES, Please provide details:_____ O 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? O YES x NO If no, please proceed to Section 10.0

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

9.3 What is the origin of the insect? _____

9.4 What is the life stage of the insect? _____

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

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

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

10.0 Plants

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

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

10.3 What is the origin of the plant? _____

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

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

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

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

10.8 Is the CFIA permit attached? O YES O NO
If 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? O YES, please give country of origin_____ x NO
If no, please proceed to Section 12.0

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

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

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

12.0 Training Requirements for Personnel Named on Form

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

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

As the Principal Investigator, I have ensured that all of the personnel named on the form who will be using any of the 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. O 1 x 2 O 2+ O 3

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

x YES, permit # if on-campus___BioUWO0062_____

O NO, please certify

O NOT REQUIRED for Level 1 containment

14.0 Procedures to be Followed

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

SIGNATURE _____ Date: Nov 30, 2010

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

__Staff/trainees wear lab coats, do not eat/drink in lab, took all training sessions, follow proper procedures and autoclave all waste before disposal._____

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

_Our bacteria are serum sensitive, so no worries about septicemia. Just disinfect site of wounds to prevent entry of other pathogens._____

15.0 Approvals

1) UWO Biohazards Subcommittee: SIGNATURE: _____
Date: 23 Dec 2010

2) Safety Officer for the University of Western Ontario
SIGNATURE: J Stanley
Date: Dec 23/10

3) Safety Officer for Institution where experiments will take place (if not UWO):
SIGNATURE:_____
Date:_____

Approval Number: BIO-UWO-0062 Expiry Date (3 years from Approval): December 22, 2013

Special Conditions of Approval:

Risk Group Classification for Infectious Agents

Bacteria Search Results

Genus: Helicobacter	Species: pylori	
	Risk Group Level	**Notes**
Australia/New Zealand 2002:	2	
Belgium 2004:	2	
Switzerland 2003:	2	(Campylobacter pylori; Campylobacter pylori subsp. pylori)
United Kingdom 2004:	2	
Germany 2001:	2	AR
NIH 2002	2	
European Community 2000:	2	
Singapore 2004:		Singapore Schedule:
Japan:	2	
Human Pathogen: Yes **Animal Pathogen:** No **Plant Pathogen:** No		**Select Agent CDC:** No **Select Agent USDA:** No
MSDS:		

American Biological Safety Association, 1200 Allanson Road, Mundelein, IL 60060-3808
Phone: 1-866-425-1385 (toll free), 847-949-1517
Fax: 847-566-4580 **E-mail: info@absa.org**

You may need to download Acrobat Reader to view PDF documents appearing on this site.

Campylobacter fetus ssp. fetus - Material Safety Data Sheets (MSDS)

MATERIAL SAFETY DATA SHEET - INFECTIOUS SUBSTANCES

SECTION I - INFECTIOUS AGENT

NAME: ***Campylobacter jejuni, C. coli, C. fetus* subsp. *jejuni***

SYNONYM OR CROSS REFERENCE: *Campylobacter* enteritis, Vibrionic enteritis, Traveller's diarrhea

CHARACTERISTICS: Gram negative spiral and S-shaped bacteria; darting motility; microaerophilic; will grow only under reduced oxygen tension; optimal growth temperature is at 42C

SECTION II - HEALTH HAZARD

PATHOGENICITY: Acute enteric disease of variable severity; diarrhea, abdominal pain, malaise, fever, nausea and vomiting; prolonged illness in up to 20% of patients; blood in association with mucus and WCBs present in liquid of foul smelling stools; typhoidal-like syndrome, reactive arthritis may occur ; rare cases of febrile convulsions, Guillain-Barré syndrome and meningitis

EPIDEMIOLOGY: Important cause of diarrheal illness worldwide in all age groups (5-14% of diarrhea in world); common source outbreaks most often associated with foods, unpasteurized milk and unchlorinated water; largest number of sporadic cases in temperate climates occur in warmer months

HOST RANGE: Humans, animals and birds

INFECTIOUS DOSE: 500 organisms or less (by ingestion)

MODE OF TRANSMISSION: By ingestion of organisms in undercooked food or in unpasteurized milk or water; from contact with infected pets (puppies and kittens), farm animals or infected infants; possibly from cross-contamination from these sources to foods eaten uncooked or poorly refrigerated

INCUBATION PERIOD: 2-5 days, with a range of 1-10 days; dose-dependent

COMMUNICABILITY: Communicable throughout course of infection; individuals not treated with antibiotics excrete organisms for as long as 2-7 weeks; chronic carrier state is unusual

SECTION III - DISSEMINATION

RESERVOIR: Animals - swine, cattle, sheep, cats, dogs, other pets and rodents; birds, including poultry

ZOONOSIS: Yes - chronic carrier state established and animals constitute primary source of infection

VECTORS: None

SECTION IV - VIABILITY

DRUG SUSCEPTIBILITY: Sensitive to erythromycin, tetracyclines, fluoroquinolones and aminoglycosides

DRUG RESISTANCE: Single- and multiple-drug resistant strains have been reported

SUSCEPTIBILITY TO DISINFECTANTS: Susceptible to many disinfectants - 1% sodium hypochlorite, 70% ethanol or isopropyl alcohol, 2% glutaraldehyde, iodines, phenolics, formaldehyde; commonly used disinfectants for drinking water treatment (0.1 mg/l of free chlorine, and 1 mg/l of monochloramine) are sufficient to kill *C. jejuni*

PHYSICAL INACTIVATION: Sensitive to moist heat (121°C for at least 15 min) and dry heat (160-170°C for at least 1 hour); highly sensitive to gamma irradiation and UV radiation

SURVIVAL OUTSIDE HOST: Will survive in moist environments (including droplets), especially at lower temperatures, but cannot tolerate drying; Feces - up to 9 days; milk - 3 days; glass slides - 24 hours; water - 2 to 5 days

SECTION V - MEDICAL

SURVEILLANCE: Monitor for symptoms; confirmation by isolation from stool

FIRST AID/TREATMENT: Rehydration and electrolyte replacement; short antibiotic course for severe or prolonged illness

IMMUNIZATION: None

PROPHYLAXIS: Not usually administered

SECTION VI - LABORATORY HAZARDS

LABORATORY-ACQUIRED INFECTIONS: 2 reported cases of laboratory-acquired infection

SOURCES/SPECIMENS: Feces, blood

PRIMARY HAZARDS: Ingestion, parenteral inoculation

SPECIAL HAZARDS: Infected laboratory animals

SECTION VII - RECOMMENDED PRECAUTIONS

CONTAINMENT REQUIREMENTS: Biosafety level 2 practices, containment equipment and facilities for activities with clinical materials known or potentially infected and cultures; animals biosafety level 2 facilities and practices

PROTECTIVE CLOTHING: Laboratory coat; gloves when contact with infected materials is unavoidable

OTHER PRECAUTIONS: Good personal hygiene and frequent handwashing

SECTION VIII - HANDLING INFORMATION

SPILLS: Allow aerosols to settle; wearing protective clothing, gently cover spill with paper towels and apply 1% sodium hypochlorite, starting at perimeter and working towards the centre; allow sufficient contact time (30 min) before clean up

DISPOSAL: Decontaminate before disposal; steam sterilization, chemical disinfection, incineration

STORAGE: In sealed containers that are appropriately labelled

SECTION IX - MISCELLANEOUS INFORMATION

Date prepared: November 1999

Prepared by: Office of Laboratory Security, PHAC

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Date Modified: 2001-01-23

Bacteria

ATCC® Number: **29428™** Order this Item Price: **$40.00**

Preceptrol® Culture

Organism:	*Campylobacter jejuni* subsp. *jejuni* (Jones et al.) Veron and Chatelain deposited as *Campylobacter fetus* subsp. *jejuni* Smibert
Designations:	VPI H840 [CIP 103778]
Isolation:	feces, human (diarrheic stool of child)
Depositor:	NR Krieg
History:	ATCC<<--NR Krieg<<--P. Dekeyser
Biosafety Level:	2
Shipped:	freeze-dried
Growth Conditions:	ATCC medium1116: Brucella broth with 0.16% agar Alternate medium 177: Fluid thioglycollate medium **Temperature:** 37.0°C **Atmosphere:** Microaerophilic
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.
Cross References:	Nucleotide (GenBank) : X83374 C.jejuni omp18 gene.
Applications:	media testing [92390] [92845]
References:	92390: Microbiology of food and animal feeding stuffs--Guidelines on preparation and production of culture media--Part 2: Practical guidelines on performance testing of culture media.. Geneva (Switzerland):International Organization for Standardization/ANSI;ISO ISO 11133-2:2003. 92845: Microbiology of food and animal feeding stuffs ---Guidelines on preparation and production of culture media ---- Part 2: Practical guidelines on performance testing of culture media - Annex B: Recommended test microorganisms for commonly used culture media. London, UK:British Standards Institution;British Standard DD CEN ISO/TS 11133:2003.

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BioServices

Bio-materials management; basic repository to complex partnership-level services

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Biological Reference Material and Consensus Standards for the life science community

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Canada

Yersinia enterocolitica, Yersinia pseudotuberculosis - Material Safety Data Sheets (MSDS)

MATERIAL SAFETY DATA SHEET - INFECTIOUS SUBSTANCES

SECTION I - INFECTIOUS AGENT

NAME: ***Yersinia enterocolitica, Yersinia pseudotuberculosis***

SYNONYM OR CROSS REFERENCE: Yersiniosis, enterocolitis, pseudotuberculosis

CHARACTERISTICS: Gram negative rod-shaped to ovoid bacilli, some strains produce a heat-stabile enterotoxin (ST), serologic identification of somatic antigens (*Y. pseudotuberculosis* - O-group 1 strains most frequent>90%)

SECTION II - HEALTH HAZARD

PATHOGENICITY: Acute enteric disease manifested by acute watery diarrhea, enterocolitis, acute mesenteric lymphadenitis mimicking appendicitis, fever, headache, pharyngitis, anorexia, vomiting, erythema nodosum, arthritis, iritis, cutaneous ulceration, hepatosplenic abscesses, osteomyelitis and septicemia; *Y. enterocolitica* - gastroenterocolitis syndrome; *Y. pseudotuberculosis* - abdominal pain, higher case fatality rate in immunocompromised individuals

EPIDEMIOLOGY: Worldwide; 2/3 of *Y. enterocolitica* cases occur among infants and small children; 3/4 of *Y. pseudotuberculosis* cases involve 5 to 20 year olds; highest rate during cold season in temperate climates; epidemics associated with hospitals and schools as well as contaminated vehicles (milk)

HOST RANGE: *Y. pseudotuberculosis* is primarily a zoonotic disease of wild and domesticated birds and mammals, with humans as incidental hosts; *Y. enterocolitica* has been recovered from a wide variety of animals without signs of disease (fatal outbreak in chinchillas); household pets - sick puppies and kittens; pigs

INFECTIOUS DOSE: 10^6 organisms

MODE OF TRANSMISSION: Fecal-oral transmission by contact with infected persons or animals, or by eating and drinking fecally contaminated food and water; nosocomial transmission has been reported; transmission by infected blood products has been reported

INCUBATION PERIOD: Probably 3 to 7 days, generally under 10 days

COMMUNICABILITY: Fecal shedding at least as long as symptoms exist; untreated cases may excrete organism for 2 to 3 months; chronic carrier state exists

SECTION III - DISSEMINATION

RESERVOIR: Principal reservoirs are domestic animals; *Y. enterocolitica* has been recovered from healthy animals and from primates with acute enteric disease; pigs may be an important reservoir through pork products, especially head meats; *Y. pseudotuberculosis* is widespread among many species of avian and mammalian hosts

ZOONOSIS: Yes, by contact with infected animals

VECTORS: None

SECTION IV - VIABILITY

DRUG SUSCEPTIBILITY: Sensitive to many antibiotics; may be resistant to penicillin and its semisynthetic derivatives

SUSCEPTIBILITY TO DISINFECTANTS: Susceptible to many disinfectants - 1% sodium hypochlorite, 70% ethanol, 2% glutaraldehyde, iodines, phenolics, formaldehyde

PHYSICAL INACTIVATION: Sensitive to moist heat (121° C for at least 15 min) and dry heat (160 -170° C for at least 1 hour)

SURVIVAL OUTSIDE HOST: Water - 20 days; beets - 1 to 2 days; linen - 18 hours; seawater - up to 105 days (winter); soil - 540 days

SECTION V - MEDICAL

SURVEILLANCE: Monitor for symptoms; confirmation by serologic agglutination tests; circulating antibodies; stool samples; ELISA

FIRST AID/TREATMENT: Antibiotic therapy may be helpful for gastrointestinal symptoms; definitely indicated for septicemia or other invasive disease

IMMUNIZATION: None

PROPHYLAXIS: None

SECTION VI - LABORATORY HAZARDS

LABORATORY-ACQUIRED INFECTIONS: No reported infections to date

SOURCES/SPECIMENS: Blood, feces, urine,

PRIMARY HAZARDS: Ingestion, accidental parenteral inoculation

SPECIAL HAZARDS: Contact with infected animals

SECTION VII - RECOMMENDED PRECAUTIONS

CONTAINMENT REQUIREMENTS: Biosafety level 2 practices, containment equipment and facilities for activities with cultures or potentially infectious clinical materials; animal biosafety level 2 practices and facilities for activities involving infected animals

PROTECTIVE CLOTHING: Laboratory coat; gloves when direct contact with infectious materials is unavoidable

OTHER PRECAUTIONS: Good personal hygiene and frequent hand washing

SECTION VIII - HANDLING INFORMATION

SPILLS: Allow aerosols to settle; wearing protective clothing, gently cover spill with paper towels and apply 1% sodium hypochlorite, starting at perimeter and working towards the centre; allow sufficient contact time (30 min) before clean up

DISPOSAL: Decontaminate before disposal; steam sterilization, chemical disinfection, incineration (animal wastes)

STORAGE: In sealed containers that are appropriately labeled

SECTION IX - MISCELLANEOUS INFORMATION

Date prepared: January, 2001

Prepared by: Office of Laboratory Security, PHAC

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Date Modified: 2001-03-05

Hosts

ATCC® Number:	**53323** Order this Item	Price:	**$190.00**
Organism:	*Escherichia coli* (Migula) Castellani and Chalmers		
Designations:	JM109		
Depositors:	Crop Genetics International N.V.		
Genotype:	F' traD36 proA+ proB+ lacIq delta(lacZ)M15 delta(lac-proAB) supE44 hsdR17 recA1 gyrA96 thi-1 endA1 relA1 e14- lambda-		
Growth Conditions:	ATCC medium 1065: LB medium **Temperature:** 37.0°C		
Biosafety Level:	1		
Shipped:	frozen		
Permits/Forms:	In addition to the MTA mentioned above, other ATCC and/or regulatory permits may be required for the transfer of this ATCC material. Anyone purchasing ATCC material is ultimately responsible for obtaining the permits. Please click here for information regarding the specific requirements for shipment to your location.		
Applications:	transformation host		
Comments:	Bacteriophage host This is a host for M13 and other filamentous bacteriophages		
References:	10266: Yanisch-Perron C, et al. Improved M13 phage cloning vectors and host strains: nucleotide sequences of the M13mp18 and pUC19 vectors. Gene 33: 103-119, 1985. PubMed: 2985470		

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Return to Top

-------- Original Message --------
Subject: Re: Biological Agents Registry Form: Creuzenet
Date: Tue, 30 Nov 2010 11:21:53 -0500
From: Carole Creuzenet <ccreuzen@uwo.ca>
To: Jennifer Stanley <jstanle2@uwo.ca>

Hi Jennifer,
Here is the revised BARF form attached. I will also put a hardcopy through the mail.
Note that I added the new personnel that was hired AFTER the last form was submitted.
I hope all is good now
CC

Carole CREUZENET, PhD
Associate Professor
Graduate co-Chair
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Dept of Microbiology and Immunology
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Tel: 519 661 3204
Fax: 519 661 3499
e-mail: ccreuzen@uwo.ca
Web site: http://publish.uwo.ca/~ccreuzen/

On 26/11/2010 2:54 PM, Jennifer Stanley wrote:

Hi Dr. Creuzenet -

Your recent submission was reviewed at the November Biohazards Subcommittee meeting.

The source of Helicobacter pylori should be stated in section 1.2. "Other Canadian labs" is not specific enough.
The list of personnel may not be a complete representation of individuals working in the lab?
"No" should be checked in tables 2.2 and 2.3 where applicable.
Table 1.2 should include E. coli DH5alpha, JM109
Table 4.2 should include gene(s) transfected, if possible

Please send the revisions to me.

Thanks, and have a great week-end.
Jennifer

-------- Original Message --------
Subject: Re: Biological Agents Registry Form: Creuzenet
Date: Thu, 21 Oct 2010 10:37:22 -0400
From: Carole Creuzenet <ccreuzen@uwo.ca>
To: Jennifer Stanley <jstanle2@uwo.ca>

Hi Jennifer,
Sorry, it is grant writing time and teaching time, so this skipped my mind.

- For CJ, yes 1 liter is about what we grow at one time. This bacterium rather grows on plates, so we only use the broth for very specialized applications.

- For HP, no MSDS available as far as I know. It is very closely related to CJ.

- For the genes, we have so many and they just go by numbers/codes which do not provide any information in terms of safety. The ones we have encode enzymes that modify sugars, so none of them encodes any toxins. Examples are: CJ1427, CJ1428, CJ1429, Cj1430, HP0366, HP0840, WcbK, WcaG, DmhA, DmhB, Cj1121c, Cj1123c, CJ1293, CJ1293, WbpM.
Others encode various proteins involved in secretion: HP0235, HP0231, HP0253, HP0879.

I hope this helps
CC

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