

Modification Form for Permit BIO-UWO-0082

Permit Holder: Miguel Valvano

Approved Personnel

(Please stroke out any personnel to be removed)

Daniel Aubert

Kinnari Patel

~~Emily Haggerty~~

Jennifer Tolman

S. Saldias

Slade Loutet

~~Karen Keith~~

~~Kendra Maloney~~

~~R. Flannagan~~

~~Ximena Ortega~~

Cristina Marolda

Additional Personnel

(Please list additional personnel here)

Robert Newman

Sarah Furlong

Xiang Ruan

Mohamed Hamad

Daniel Hynes

Alexander Skeldon

* PLEASE ATTACH A MATERIAL SAFETY DATA SHEET OR EQUIVALENT FOR NEW BIOHAZARDS.

** PLEASE ATTACH A BRIEF DESCRIPTION OF THE WORK THAT EXPLAINS THE BIOHAZARDS USED AND HOW THEY WILL BE USED.

Classification: 2

Date of last Biohazardous Agents Registry Form: Oct 2, 2006

Signature of Permit Holder: Miguel Valvano

BioSafety Officer(s): _____

Chair, Biohazards Subcommittee: _____

Modification Form for Permit BIO-UWO-0082

Permit Holder: Miguel Valvano

	Please stroke out any approved Biohazards to be removed below	Write additional Biohazards for approval below. *
Approved Microorganisms	Burkholderia cepacia and E.coli K-12 <i>Salmonella</i> sp <i>shigella</i> sp <i>Pseudomonas</i> sp	
Approved Cells	Human established microphages and Rodent (established and primary) microphage THP1, RAW, PU5.1 Established epithelial cells lines - A549	plasmids for Transfection pCDNA3-EGFP-cdc42(WT)
Approved Use of Human Source Material	L CACO-2 L Hep2 L BHK	L cdc42 (T17N) L cdc42 (Q61L) L Rae1 (Q61L)
Approved GMO		L Rae1 (T17N) L Raes (WT)
Approved use of Animals	inbred mice	
Approved Toxin(s)		

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

Classification: 2

Date of last Biohazardous Agents Registry Form: Oct 2, 2006

Signature of Permit Holder: Miguel Valvano

BioSafety Officer(s): _____

Chair, Biohazards Subcommittee: _____

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

Subject:Fwd: New MTA for Valvano lab plasmid request (Order 20536)

Date:Fri, 27 Feb 2009 11:45:20 -0500

From:Miguel Valvano <Miguel.Valvano@schulich.uwo.ca>

To:Jennifer Stanley <jstanle2@uwo.ca>

CC:cmarolda@uwo.ca

References:<49A7CFF5.6CC1.0022.0@schulich.uwo.ca>

Dear Jennifer

Below is the information we requested

Salmonella sp, Shigella sp and Pseudomonas sp. are used as model system to investigate the biosynthesis and assembly of the lipopolysaccharide molecule.

cell lines A549, CACO-2, Hep2, and BHK, are all epithelial cell lines that we use to test the ability of Burkholderia cenocepacia to attach and also to invade intracellularly.

The plasmids from Addgene that we currently have fall into two types. One type are plasmid vectors expressing various forms of small Rho-GTPases and they are used to transfect macrophages and follow the activation of the GTPases by fluorescen microscopy.

The other type are plasmids expressing a recombinant protein that we purify in the lab and that serves as a scaffold to prepare nandiscs. These are a lipid-protein complex where we reconstitute our LPS synthesis enzymes.

Let me know if you need any more information.

Thank you



Home | Request Plasmids | Information | Boxes | Search | Pricing | FAQ

Login | New User

About Addgene

Plasmid Cart

Recently Viewed



Price: \$65.00

Plasmid Links
Author's map
Sequence
From this article
CDC42 plasmids
Gary Bokoch Lab Plasmids
Other Links
M57296
NCBI: CDC42
CDC42 antibodies

This is no longer a plasmid entry
pcDNA3-EGFP-Cdc42-GFP
pcDNA3-EGFP-Rac1-T17N
pcDNA3-EGFP-RhoA-T17N
pcDNA3-EGFP-Rac1-G01L
pcDNA3-EGFP-Cdc42-WT

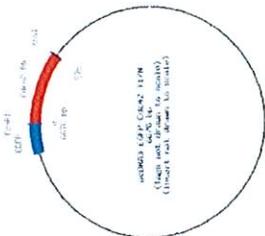
Browse > Gary Bokoch > Subauste et al > pcDNA3-EGFP-Cdc42-T17N

Plasmid 12976: pcDNA3-EGFP-Cdc42-T17N

- Cells/Species tested: Cdc42 DN
- Alternative names: Cdc42
- Insert size (bp): 576
- GeneBank/EMBL/EBI accession: M57296
- Current project: CDC42, G25K, CDC42Hs
- Species of interest: H. sapiens (human)
- Protein name: T17N Dominant negative EGFP
- Fluorescent proteins of tags: N terminal on backbone
- Formular: pcDNA3-EGFP (Search Vector Database)
- Vendor backbone: Invitrogen
- Gene source manufacturer: Mammalian expression
- Type of vector: 6100
- Barcode size (bp): 6100
- Cloning site: 5' EcoRI
- Gen: 203Bry-03
- Random cloning: No
- Cloning site 3': XhoI
- Cloning site 5': No
- Size (kilo) base pair (during cloning): No
- 5' Sequencing primer: T7 for EGFP (List of Sequencing Primers)
- 3' Sequencing primer: S66
- Bacterial resistance: Ampicillin
- High of low copy: High Copy
- Grow in standard E. coli @ 37C: Yes
- Sequence: View sequence
- Author's Map: View map
- Plasmid provided in: DH5a
- Participate in activity flow: Gary Bokoch
- Terms used: MTA

Addgene has sequenced a portion of this plasmid for verification. Click here for the sequencing result.

Click on map to enlarge



Addgene Rho family proteins modulate rapid apoptosis induced by cytotoxic T lymphocytes and Fas. Subauste MC et al (J Biol Chem. 2000 Mar 31; 275(13):9725-35. 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 12976" in your Materials and Methods section. This information allows Addgene to create a link from the plasmid page to your publication.

View Addgene plasmid data for this plasmid: pcDNA3-EGFP-Cdc42-T17N



Search for Plasmids:

Home Deposit Plasmids Request Plasmids Plasmid Tools Information Browse Search Privacy Policy FAQ

Login | New User

Plasmid Cart Your cart is empty.

Add to Cart

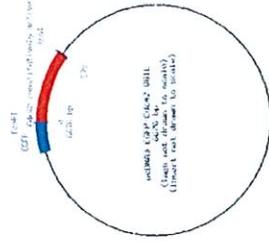
Recently Viewed pcDNA3-EGFP-Cdc42-Q61L Plasmid 12986

Plasmid 12986: pcDNA3-EGFP-Cdc42-Q61L

Contribuent name: Cdc42 constitutively active
Alternative names: Cdc42
Insert size (bp): 576
GeneBank/EMBL/ID of insert: M57298
Contig/contig aliases: CDC42, G25K, CDC42hs
Species of interest: H. sapiens (human)
Plasmid maintenance/destination: Q61L Constitutively active EGFP
Fusion partners or tags: EGFP
Terminal: N terminal on backbone
pcDNA3-EGFP (Search Vector Database)
Vector backbone: pcDNA3-EGFP
Bacteriome manufacturer: Invitrogen
Type of vector: Mammalian expression
Baculovirus size (bp): 6100
Cloning site 5: EcoRI
Site of restriction enzyme cleavage: No
Cloning site 3: XhoI
Site of restriction enzyme cleavage: No
5' Sequencing primer: T7 for EGFP (List of Sequencing Primers)
3' Sequencing primer: Sp6
Bacterial resistance: Ampicillin
High or low copy: High Copy
Cloning in standard E. coli @ 37C: Yes
Suspension: View sequence
Supplier's Map: View map
Plasmid provided in: DH5a
Principal investigator: Gary Bokoch
Terms and Licenses: MTA

Addgene has sequenced a portion of this plasmid for verification. Click here for the sequencing result.

Click on map to enlarge



Article: Rho family proteins modulate rapid apoptosis induced by cytotoxic T lymphocytes and Fas. Subauste MC et al. (J Biol Chem. 2000 Mar 31; 275(13):19725-33. 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 12986" in your Materials and Methods section. This information allows Addgene to create a link from the plasmid page to your publication.

Home | Contact | Terms of Use | Privacy Policy | Help | About Us | Addgene



Home Deposit Plasmids Request Plasmids Plasmid Tools About Addgene

Search for Plasmids:

Advanced Search Plasmid Tools

Login | New User

Plasmid Cart

Recently Viewed

Add to Cart

Price \$65.00

Plasmid Name
Author's map
Sequence
Related Plasmids
From this article
RAC1 plasmids
Gary Bokoch Lab Plasmids
Gene Links
AF498964
NCBI RAC1
RAC1 antibodies

This is a derivative expression vector
pcDNA3-EGFP-Rac1-T7M
pcDNA3-EGFP-RhoA-Q61L
pcDNA3-EGFP-Rac1
pcDNA3-EGFP-RhoA-Cell
pcDNA3-EGFP-RhoA-T15N

Print Friendly Email

Plasmid 12981: pcDNA3-EGFP-Rac1-Q61L

Rac1 constitutively active

Rac1
579
AF498964
RAC1, MIG5, TC-25, p21-Rac1, MGC11543

H. sapiens (human)
O61L Constitutively active EGFP

N terminal on backbone
pcDNA3-EGFP (Search Vector Database)
Invitrogen
Mammalian expression

6100
EcoRI
XhoI

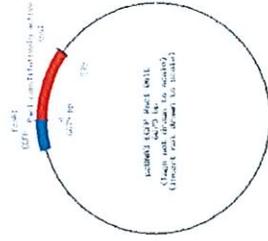
T7 for EGFP (List of Sequencing Primers)
Sp6
Ampicillin
High Copy

View sequence
Auditor's Map
DH5a

Gary Bokoch
MTA

Addgene has sequenced a portion of this plasmid for verification. Click here for the sequencing result

Click on map to enlarge



Article: Rho family proteins mediates rapid apoptosis induced by cytotoxic T lymphocytes and Fas. Subauste WC et al | J Biol Chem. 2000 Mar 31; 275(13):9725-33. 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 12981" in your Materials and Methods section. This information allows Addgene to create a link from the plasmid page to your publication.



Search for Plasmids:

Request Plasmids Browse Search Pricing FAQ

Login | New User

About Addgene Plasmid Cart

Print Friendly Email

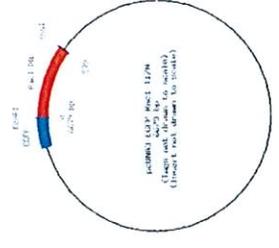
Plasmid 12982: pcDNA3-EGFP-Rac1-T17N

GenBank name	Rac1 DN
Accession number	Rac1
Insert size (bp)	579
GeneBank Entry ID of insert	AF498964
GeneBank accession	RAC1, MIG5, TC-25, p21-Rac1, MGC111543
Species of (yep/ye)	H. sapiens (human)
Relevant modifications	T17N Dominant negative
Fusion proteins or tags	EGFP
Terminal	N terminal on backbone
Vector backbone	pcDNA3-EGFP (Search Vector Database)
Biological manufacturer	modified pcDNA3
Type of vector	Mammalian expression
Backbone size (bp)	6100
Cloning site 5'	EcoRI
Site digested during cloning	No
Cloning site 3'	XhoI
Site digested during cloning	No
5' Sequencing primer	T7 for EGFP (List of Sequencing Primers)
3' Sequencing primer	Sp6
Bacterial resistance	Ampicillin
High or low copy	High Copy
Gene in standard E. coli strain	Yes
Sequence	View sequence
Author's Map	View map
Plasmid Provided In	DH5a
Principal Investigator	Gary Bokoch
Terms and Licenses	MTA

This is a mammalian expression vector
pcDNA3-EGFP-Rac1-061L
pcDNA3-EGFP-RhaA-T19N
pcDNA3-EGFP-Rac1-rt
pcDNA3-EGFP-Cdc42-T17N
pcDNA3-EGFP-RhaA-063L

Addgene has sequenced a portion of this plasmid for verification. Click here for the sequencing result

Click on map to enlarge



Article: Rho family proteins modulate rapid apoptosis induced by cytotoxic T lymphocytes and Fas. Subauste WC et al. (J Biol Chem. 2000 Mar 31; 275(13):925-33. 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 12982" in your Materials and Methods section. This information allows Addgene to create a link from the plasmid page to your publication.



Home Deposit Plasmids Request Plasmids Plasmid Family
 Identification Browse Search Pricing FAQ
 Search for Plasmids:

Print Friendly Email

Plasmid 12980: pcDNA3-EGFP-Rac1-wt

GenBank ID: [F64884](#)
 Insert Size (bp): 579
 Sequencing: [AF498964](#)
 GenBank ID: [F64884](#)
 Gen request: [af498964](#)
 Species: [H. sapiens \(human\)](#)
 Feature positions: [see map](#)
 Terminal: [N terminal on backbone](#)
 Vector backbone: [pcDNA3-EGFP \(Search Vector Database\)](#)
 Expression: [in vitro](#)
 Mammalian expression: [Yes](#)
 Barcode size (bp): [6100](#)
 Cloning site 5': [EcoRI](#)
 Site: [BamHI](#)
 Cloning site 3': [XhoI](#)
 Site: [Not digested during cloning](#)
 5' Sequencing primer: [T7 for EGFP \(List of Sequencing Primers\)](#)
 3' Sequencing primer: [Sp6](#)
 Antibiotic resistance: [Ampicillin](#)
 High of low copy: [High Copy](#)
 Gateway destination: [E. coli \(p\) 37C](#)
 Sequences: [View sequence](#)
 Author's Map: [View map](#)
 Plasmid provided in: [DH5a](#)
 Principal investigator: [Gary Bokoch](#)
 Terms and Conditions: [MTA](#)

Login | New User

Your cart is empty

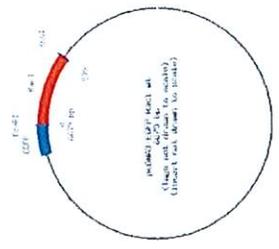
Recently Viewed

- pcDNA3-EGFP-Rac1 Plasmid 12980
- Addgene EGFP-Rac1 Plasmid 12981
- Addgene EGFP-Rac1 Plasmid 12982

Price: \$65.00

Plasmid type:
Author's map:
Sequence:
Related Plasmids:
From this article:
RAC1 plasmids:
City Research Lab Plasmids:
GenBank IDs:
AF498964
NCBI: RAC1
RAC1 antibodies

This is downloadable
pcDNA3-EGFP-Rac1-001L
pcDNA3-EGFP-Rac1-T7N
pcDNA3-EGFP-Rac1-wt
pcDNA3-EGFP-Cad2-wt
pcDNA3-EGFP-RhoA-T5N



Article: [Rho family proteins modulate rapid apoptosis induced by cytotoxic T lymphocytes and Fts. Subauste MC et al. \(J Biol Chem. 2000 Mar 31; 275\(13\):9725-33. 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 12980" in your Materials and Methods section. This information allows Addgene to create a link from the plasmid page to your publication.

Home | Contact Us | Terms of Use | Privacy Policy | About Us | Addgene EGFP-Rac1 Plasmid 12980

Addgene has sequenced a portion of this plasmid for verification. [Click here](#) for the sequencing result

[Click on map to enlarge](#)



Home > Emergency Preparedness > Laboratory Security > Material Safety Data Sheets (MSDS) - Infectious Substances
> Pseudomonas spp. (excluding B. mallei, B. pseudomallei) - Material Safety Data Sheets (MSDS)

Pseudomonas spp. (excluding B. mallei, B. pseudomallei) - Material Safety Data Sheets (MSDS)

MATERIAL SAFETY DATA SHEET - INFECTIOUS SUBSTANCES

SECTION I - INFECTIOUS AGENT

NAME: *Pseudomonas* spp. (excluding *B. mallei*, *B. pseudomallei*)

SYNONYM OR CROSS REFERENCE: *P. aeruginosa*, *P. cepacia*

CHARACTERISTICS: Family Pseudomonadaceae, gram negative bacillus, aerobic, non-spore forming, some pigmented (pyocyanin, fluorescein), motile by polar flagella, variety of toxins produced

SECTION II - HEALTH HAZARD

PATHOGENICITY: Opportunistic pathogen, greatest risk of disease in the immunocompromised; most medical conditions arise from colonization of pathogen in the respiratory and urinary tracts or due to deep disseminated infections leading to pneumonia and bacteremia; chronic respiratory infections among cystic fibrosis patients; eye infections (especially in contact lens wearers); nosocomial infections causing severe and often fatal infections (case fatality in susceptible populations is 30%), increasingly associated with bacterial meningitis, abscesses, endocarditis

EPIDEMIOLOGY: Worldwide; increasing in frequency in recent years; commonly a nosocomial infection associated with contaminated instruments; 16% of nosocomial pneumonia, 12% of hospital acquired urinary-tract infections; rarely causes community acquired infections in immunocompetent patients

HOST RANGE: Humans, animals, plants

INFECTIOUS DOSE: Not known

MODE OF TRANSMISSION: Direct contact with contaminated water, aerosols or aspirations, by contact of mucous membranes with discharges from infected conjunctivae or upper respiratory tract of infected persons through contaminated objects (improperly sterilized medical equipment, contaminated IV fluids) or fingers;

INCUBATION PERIOD: Variable depending on infection; eye infection - 24 to 72 hours

COMMUNICABILITY: Can be transmitted during course of active infection

SECTION III - DISSEMINATION

RESERVOIR: Saprophyte - soil, water, decomposing matter; infected animals and humans; infected solutions - I.V., soaps, eye drops, humidifiers; organism thrives in moist conditions

ZOONOSIS: None

VECTORS: None

SECTION IV - VIABILITY

DRUG SUSCEPTIBILITY: Sensitive to extended spectrum penicillins, aminoglycosides,

cephalosporins, fluoroquinolones, polymyxins and monobactams; aminoglycoside with a beta-lactam penicillin is the first line of treatment

DRUG RESISTANCE: Multidrug resistant strains are on the rise

SUSCEPTIBILITY TO DISINFECTANTS: Susceptible to many disinfectants - 1% sodium hypochlorite, 70% ethanol, 2% glutaraldehyde, formaldehyde; few reports of this bacteria growing in disinfectant solutions; alcohol-containing disinfectants recommended for resistant strains

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

SURVIVAL OUTSIDE HOST: Survives for several months in water with minimal nutrients

SECTION V - MEDICAL

SURVEILLANCE: Bacteriological identification of infection

FIRST AID/TREATMENT: Antibiotic therapy - aggressive treatment is necessary to avoid chronic infections; drainage of wounds; local application of antibiotic ointment or drops

IMMUNIZATION: None

PROPHYLAXIS: Antibiotic prophylaxis, not usually administered

SECTION VI - LABORATORY HAZARDS

LABORATORY-ACQUIRED INFECTIONS: No reported infections to date

SOURCES/SPECIMENS: Clinical specimens - respiratory secretions, wound exudates, blood, urine; environmental specimens - water, infected solutions (IV, disinfectants, soap)

PRIMARY HAZARDS: Accidental parenteral inoculation; direct contact of mucous membranes with infected materials; inhalation of infectious aerosols and ingestion also present a hazard

SPECIAL HAZARDS: None

SECTION VII - RECOMMENDED PRECAUTIONS

CONTAINMENT REQUIREMENTS: Biosafety level 2 practices, containment equipment and facilities for activities involving suspected or known infectious specimens and cultures

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

OTHER PRECAUTIONS: Good personal hygiene, frequent hand washing and the avoidance of rubbing eyes as a precautionary measure against eye infections

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 before clean up and disposal (30 min)

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

STORAGE: In sealed containers that are appropriately labelled

SECTION IX - MISCELLANEOUS INFORMATION

Date prepared: March, 2001

Prepared by: Office of Laboratory Security, PHAC

Although the information, opinions and recommendations contained in this Material Safety Data Sheet are compiled from sources believed to be reliable, we accept no responsibility for the accuracy, sufficiency, or reliability or for any loss or injury resulting from the use of the information. Newly discovered hazards are frequent and this information may not be completely up to date.

Copyright ©
Health Canada, 2001

Date Modified: 2001-05-14



Home > Emergency Preparedness > Laboratory Security > Material Safety Data Sheets (MSDS) - Infectious Substances
> Salmonella spp. (excluding S. typhi, S. choleraesuis, and S. paratyphi) - Material Safety Data Sheets (MSDS)

Salmonella spp. (excluding S. typhi, S. choleraesuis, and S. paratyphi) - Material Safety Data Sheets (MSDS)

MATERIAL SAFETY DATA SHEET - INFECTIOUS SUBSTANCES

SECTION I - INFECTIOUS AGENT

NAME: *Salmonella* spp. (excluding *S. typhi*, *S. choleraesuis*, and *S. paratyphi*)

SYNONYM OR CROSS REFERENCE: Salmonellosis

CHARACTERISTICS: Family Enterobacteriaceae; gram negative rod; motile, aerobic and facultatively anaerobic; serological identification of somatic and flagellar antigens; over 2000 serotypes capable of causing disease

SECTION II - HEALTH HAZARD

PATHOGENICITY: Salmonellosis is an acute gastroenteritis; acute infectious disease with sudden onset of abdominal pain, diarrhea, nausea and vomiting; dehydration may be severe in infants and elderly; deaths are uncommon except in very young or very old or debilitated/immunocompromised; morbidity may be high; food borne disease; may progress to more serious septicemia, includes focal infections, abscesses, endocarditis, pneumonia; may also cause typhoid like enteric fever; some cases develop reactive arthritis (Reiter's syndrome) which may become chronic

EPIDEMIOLOGY: Worldwide, more extensively in North America and Europe; higher incidence rate for infants and young children; small outbreaks in general population; large outbreaks in hospitals, institutions, nursing homes, restaurants; 2-3 million infections annually in USA, most go unreported; *S. enteritidis* and *S. typhimurium* are the most common in North America

HOST RANGE: Humans; domestic and wild animals, birds

INFECTIOUS DOSE: 100 - 1,000 organisms - ingestion; varies with multiple factors

MODE OF TRANSMISSION: By ingestion of directly or indirectly contaminated food, from infected animals or food by infected animal or person; from animal feeds and fertilizers prepared from contaminated meat scraps; fecal-oral transmission from person to person; direct contact with pets such as reptiles, birds, turtles, tortoises

INCUBATION PERIOD: Six to 72 hours, usually about 12-36 hours

COMMUNICABILITY: Communicable throughout course of infection; several days to several weeks; temporary carriers can continue for several months; antibiotic therapy can prolong period of communicability; 1% of infected adults and 5% of infected children excrete organism for over 1 year

SECTION III - DISSEMINATION

RESERVOIR: Humans - patients and carriers; domestic and wild animals - poultry (*S. enteritidis*), swine (*S. choleraesuis*), cattle, rodents, horses (*S. dublin*); pets - tortoises, turtles, chicks, dogs, cats; some strains associated with specific animal reservoir

ZOONOSIS: Yes - direct or indirect contact with animals (most are via ingestion); pets especially birds, reptiles, turtles tortoises

VECTORS: None

SECTION IV - VIABILITY

DRUG SUSCEPTIBILITY: Sensitive to ampicillin, amoxicillin, TMP-SMX, chloramphenicol, fluoroquinolones; many strains are antibiotic, or multi-drug resistant; drug susceptibility testing must be performed

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: Survives for long periods in the environment

SECTION V - MEDICAL

SURVEILLANCE: Monitor for gastrointestinal symptoms; confirm by stool culture, isolation from blood during acute stages

FIRST AID/TREATMENT: For enterocolitis - rehydration and electrolyte replacement; for enteric fever or septicemia - antibiotic therapy

IMMUNIZATION: None

PROPHYLAXIS: Not usually administered

SECTION VI - LABORATORY HAZARDS

LABORATORY-ACQUIRED INFECTIONS: 48 reported laboratory infections with *Salmonella* spp.

SOURCES/SPECIMENS: Feces, blood, urine; food, feed and environmental materials

PRIMARY HAZARDS: Ingestion, parenteral inoculation; importance of aerosol exposure not known

SPECIAL HAZARDS: None

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

PROTECTIVE CLOTHING: Laboratory coat; gloves when contact with infected 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

STORAGE: In sealed containers that are appropriately labelled

SECTION IX - MISCELLANEOUS INFORMATION

Date prepared: March, 2001

Prepared by: Office of Laboratory Security, PHAC

Although the information, opinions and recommendations contained in this Material Safety Data Sheet are compiled from sources believed to be reliable, we accept no responsibility for the accuracy, sufficiency, or reliability or for any loss or injury resulting from the use of the information. Newly discovered hazards are frequent and this information may not be completely up to date.

Copyright ©
Health Canada, 2001

Date Modified: 2001-05-15



Home > Emergency Preparedness > Laboratory Security > Material Safety Data Sheets (MSDS) - Infectious Substances
> Shigella spp. - Material Safety Data Sheets (MSDS)

Shigella spp. - Material Safety Data Sheets (MSDS)

MATERIAL SAFETY DATA SHEET - INFECTIOUS SUBSTANCES

SECTION I - INFECTIOUS AGENT

NAME: *Shigella* spp.

SYNONYM OR CROSS REFERENCE: Group A - *S. dysenteriae*, Group B - *S. flexneri*, Group C - *S. boydii*, Group D - *S. sonnei*; Shigellosis, Bacillary dysentery

CHARACTERISTIC: Family Enterobacteriaceae; Gram negative rod, non-encapsulated, non-sporogenous, non-motile; serological identification of somatic antigens; four serogroups historically treated as species; *S. dysenteriae* may produce enterotoxin (Shiga toxin)

SECTION II - HEALTH HAZARD

PATHOGENICITY: Acute disease of large and small intestine; diarrhea, fever, nausea, and sometimes toxemia, vomiting, cramps and tenesmus; stools contain blood, mucus and pus; alterations in consciousness may occur; mild and asymptomatic infections occur; severity of illness depends on host, dose and serotype - *S. dysenteriae* infections have up to 20% case fatality rate in hospitalized patients, while *S. sonnei* infections have negligible fatality rate; *S. flexneri* precipitate reactive arthritis (Reiter's syndrome) in some patients

EPIDEMIOLOGY: Worldwide; 2/3 of cases and most deaths are children under 10 years; common during weaning period; 10-40% secondary attack rates in households; outbreaks under conditions of crowding and poor sanitation; endemic in tropical and temperate climates

HOST RANGE: Humans, primates (outbreaks have occurred in colonies)

INFECTIOUS DOSE: 10-200 organisms by ingestion

MODE OF TRANSMISSION: By direct or indirect fecal-oral transmission from a patient or carrier; poor hygiene practices spread infection to others by direct physical contact or indirectly by contaminating food; water, milk, cockroach, and fly-borne transmission may occur as the result of direct fecal contamination; sexual transmission in homosexual men

INCUBATION PERIOD: One to 7 days, usually 1-3 days

COMMUNICABILITY: Communicable during acute infection and until agent is no longer present in feces, usually within 4 weeks after illness; asymptomatic carriers may transmit infection; the carrier state may persist for months or longer (although rarely)

SECTION III - DISSEMINATION

RESERVOIR: Humans are the only significant reservoir; outbreaks have occurred in primate colonies

ZOONOSIS: None

VECTORS: Flies (mechanical only)

SECTION IV - VIABILITY

DRUG SUSCEPTIBILITY: Sensitive to one or more of TMP-SMX, ampicillin, chloramphenicol,

ciprofloxacin, ofloxacin; multidrug resistant (MDR) strains are common

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: Feces up to 11 days; flies - up to 12 days; water - 2 to 3 days; shirts of patients - 8 days

SECTION V - MEDICAL

SURVEILLANCE: Monitor for symptoms; confirm by stool culture

FIRST AID/TREATMENT: Fluid and electrolyte replacement; conduct antibiotic susceptibility tests and administer antibiotic therapy if illness is severe (avoid drugs which slow intestinal motility)

IMMUNIZATION: None

PROPHYLAXIS: Administration of antibiotics for prophylaxis generally not recommended

SECTION VI - LABORATORY HAZARDS

LABORATORY-ACQUIRED INFECTIONS: at least 81 reported cases of shigellosis acquired from a laboratory before 1991

SOURCES/SPECIMENS: Feces; rarely, blood of infected humans and animals

PRIMARY HAZARDS: Ingestion or parenteral inoculation; importance of aerosols exposure not known

SPECIAL HAZARDS: Experimentally infected guinea pigs, other rodents and non-human primates are a proven source of infection

SECTION VII - RECOMMENDED PRECAUTIONS

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

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: March, 2001

Prepared by: Office of Laboratory Security, PHAC

Although the information, opinions and recommendations contained in this Material Safety Data Sheet are compiled from sources believed to be reliable, we accept no responsibility for the accuracy, sufficiency, or reliability or for any loss or injury resulting from the use of the information. Newly discovered hazards are frequent and this information may not be completely up to date.

Copyright ©

Health Canada, 2001

Date Modified: 2001-04-23

Cell Biology

ATCC® Number:	CCL-185™	<input type="button" value="Order this Item"/>	Price:	\$256.00
Designations:	A549		Depositors:	M Lieber
<u>Biosafety Level:</u>	1		Shipped:	frozen
Medium & Serum:	<u>See Propagation</u>		Growth Properties:	adherent epithelial

Organism:	<i>Homo sapiens</i> (human)	Morphology:	
-----------	-----------------------------	-------------	---

Source: **Organ:** lung
Disease: carcinoma

Cellular Products: keratin

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

[Related Cell Culture Products](#)

Isolation: **Isolation date:** 1972

Applications: transfection host([technology from amaxa Roche FuGENE® Transfection Reagents](#))

Reverse Transcript: negative

Amelogenin: X, Y

CSF1PO: 10,12

D13S317: 11

D16S539: 11,12

DNA Profile (STR): D5S818: 11

D7S820: 8,11

THO1: 8,9.3

TPOX: 8,11

vWA: 14

Cytogenetic Analysis: This is a hypotriploid human cell line with the modal chromosome number of 12, occurring in 24% of cells. Cells with 64 (22%), 65, and 67 chromosome counts also occurred at relatively high frequencies; the rate with higher ploidies was low at 0.4%. There were 6 markers present in single copies in all cells. They include der(6)t(1;6) (q11;q27); ?del(6) (p23); del(11) (q21), del(2) (q11), M4 and M5. Most cells had two X and two Y chromosomes. However, one or both Y chromosomes were lost in 40% of 50 cells analyzed. Chromosomes N2 and N6 had single copies per cell; and N12 and N17 usually had 4 copies.

Isoenzymes: G6PD, B

Age: 58 years

Gender: male

Ethnicity: Caucasian

Cell Biology

ATCC® Number:	HTB-37™	Order this Item	Price:	\$264.00
Designations:	Caco-2		Depositors:	J Fogh
<u>Biosafety Level:</u>	1		Shipped:	frozen
Medium & Serum:	See Propagation		Growth Properties:	adherent epithelial

Organism:	<i>Homo sapiens</i> (human)	Morphology:	
-----------	-----------------------------	-------------	---

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

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

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

[Related Cell Culture Products](#)

Restrictions: The cells are distributed for research purposes only. The Memorial Sloan-Kettering Cancer Center releases the line subject to the following: 1.) The cells or their products must not be distributed to third parties. Commercial interests are the exclusive property of Memorial Sloan-Kettering Cancer Center. 2.) Any proposed commercial use of these cells must first be negotiated with The Director, Office of Industrial Affairs, Memorial Sloan-Kettering Cancer Center, 1275 York Avenue, New York, NY 10021; phone (212) 639-6181; FAX (212) 717-3439.

Applications: transfection host ([technology from amaxa Roche FuGENE® Transfection Reagents](#))

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

Virus Susceptibility: Human immunodeficiency virus 1

Tumorigenic: YES

Reverse Transcript: N

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

Cell Biology

ATCC® Number: **CCL-23™**

Price: **\$264.00**

[Additional information about this cell line](#)

Designations: HEp-2
Biosafety Level: 2 [Cells contain Papovavirus]
 Medium & Serum: [See Propagation](#)

Depositors: AE Moore
 Shipped: frozen
 Growth Properties: adherent
 epithelial

Organism: *Homo sapiens* (human)

Morphology: 

Source: **Organ:** HeLa contaminant

Cellular Products: keratin

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

[Related Cell Culture Products](#)

Virus Susceptibility: Human adenovirus 3
 Human poliovirus 1
 Vesicular stomatitis virus

Reverse Transcript: negative
 Amelogenin: X
 CSF1PO: 9,10
 D13S317: 12,13.3
 D16S539: 9,10

DNA Profile (STR): D5S818: 11,12
 D7S820: 8,12
 TH01: 7
 TPOX: 8,12
 vWA: 16,18

Cytogenetic Analysis: Occasional polyploids. Several marker chromosomes were observed along with frequent minutes, and often 2 large chromosomes with subterminal centromeres. HeLa Marker Chromosomes: One copy of M2, two-four copies of M3 and one copy of M4 as revealed by G-banding patterns.

Isoenzymes: G6PD, A

HeLa Markers: Y

Comments: Cells of this line contain HeLa marker chromosomes, and were derived via HeLa contamination. This line was originally thought to be derived from an epidermoid carcinoma of the larynx, but was subsequently found, based on isoenzyme analysis, HeLa marker chromosomes, and DNA fingerprinting, to have been established via HeLa cell contamination. The cells are positive for keratin by immunoperoxidase staining.

Cell Biology

ATCC® Number: **CCL-10™** [Order this Item](#) Price: **\$264.00**
 Designations: BHK-21 [C-13] Depositors: I Macpherson
Biosafety Level: 1 Shipped: frozen
 Medium & Serum: [See Propagation](#) Growth Properties: adherent
 fibroblast

Organism: Mesocricetus auratus (hamster, Syrian golden) Morphology: 

Source: **Organ:** kidney
Disease: normal
Cell Type: fibroblast

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

[Related Cell Culture Products](#)

Isolation: **Isolation date:** March, 1961
 testing [\[92346\]](#) [\[92389\]](#)

Applications: transfection host ([technology from amaxa Roche FuGENE® Transfection Reagents](#))

Virus Susceptibility: Human adenovirus 25
 reovirus 3
 Vesicular stomatitis virus
 Human poliovirus 2

Reverse Transcript: negative

Cytogenetic Analysis: Chromosome Frequency Distribution 50 Cells: 2n = 44. This is a pseudodiploid line with the tetraploidy occurring at 4%. The karyotype is 44,XY,-6,-15,6q+,15q+ in a majority of cells analyzed. The markers 6q+ and 15q+ occurred in most cells. An occasional monosomic or trisomic condition for a normal chromosome was also detected.

Age: 1 day old newborn

HeLa Markers: N

The parent line of BHK-21(C-13) was derived from the kidneys of five unsexed, 1-day-old hamsters in March, 1961, by I.A. Macpherson and M.G.P. Stoker. [\[21411\]](#)

Comments: Following 84 days of continuous cultivation, interrupted only by an 8-day preservation by freezing, clone 13 was initiated by single-cell isolation. [\[25963\]](#)
 This line has been used as a host for transformation with expression vectors containing selectable and amplifiable marker DNAs (e.g., Factor VIII, see ATCC [CRL-8544](#)).

The World Organization for Animal Health (OIE) uses the cells for routine

**THE UNIVERSITY OF WESTERN ONTARIO
 BIOHAZARDOUS AGENTS REGISTRY FORM
 Revised Biosafety Committee: October 25, 2004**

This form must be completed by each Principal Investigator holding a grant administered by the University of Western Ontario where the use of biohazardous infectious agents are described in the experimental work proposed. The form must also be completed if animal work is proposed involving the use of biohazardous agents or animal carrying zoonotic agents infectious to humans. Containment Levels will be required in accordance with Laboratory Biosafety Guidelines, 2nd edition, 1996, Health Canada (HC) 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 (Stevenson-Lawson Building, Room 60) for forward to the Biohazard Subcommittee. For questions regarding this form, please contact the Biosafety Coordinator at extension 81135. If there are changes to the information on this form (excluding grant title and funding agencies) modifications must be completed and sent to Occupational Health and Safety.
 See website: www.uwo.ca/humanresources

PRINCIPAL INVESTIGATOR Miguel A. Valvano
 SIGNATURE *Miguel A. Valvano*
 DEPARTMENT Microbiology and Immunology
 ADDRESS DSB3014; Lab SDRI-201-208
 PHONE NUMBER 83436/83427
 EMAIL mvalvano@uwo.ca

Location of experimental work to be carried out: Building(s) SDRI Room(s) 201-208

*For work being performed at Institutions affiliated with the University of Western Ontario, the Safety Officer for the Institution where experiments will take place must sign the form prior to it being sent to Occupational Health and Safety (See Section 12.0, Approvals). For research being done at Lawson Health Research Institute, London Regional Cancer Centre, Child and Parent Research Institute or Robarts Research Institute, University Biosafety Committee members can also sign as the Safety Officer.

TITLE OF GRANT(S):
Bacteria-host cell interactions of Burkholderia cepacia CCFP. Funded
Cationic antimicrobial peptide resistance and stress responses in Burkholderia cepacia CCFP. Applied
Genetics of lipopolysaccharide assembly in Gram-negative bacteria CIHR, Applied

PLEASE ATTACH A BRIEF DESCRIPTION OF YOUR WORK, SUCH A THE RESEARCH GRANT SUMMARY(S) THAT EXPLAINS THE BIOHAZARDS USED.

FUNDING AGENCY/AGENCIES Canadian Cystic Fibrosis Foundation/CIHR

- Names of all personnel working under Principal Investigators supervision in this location:
- i) C. Marolda, E. Haggerty, K. Patel, D. Aubert, J. Tolman
 - ii) J. Lamothe, K. Mafoney, X. Ortega, R. Flannagan,
 - iii) S. Saldias, K. Keith, S. Loutet
 - iv) _____
 - v) _____

1.0 Microorganisms

1.1 Does your work involve the use of microorganisms? YES NO
 If no, please proceed to Section 2.0

1.2 Please complete the table below: (See also note at the bottom of page)

Name of Microorganism	Is the microorganism known to be a human pathogen? YES/NO	Is the microorganism known to be an animal pathogen? YES/NO	Is the microorganism known to be a zoonotic agent? YES/NO	Maximum quantity to be cultured at one time?
Burkholderia cepacia	<input checked="" type="checkbox"/> Yes <input type="checkbox"/> No	<input checked="" type="checkbox"/> Yes <input type="checkbox"/> No	<input type="checkbox"/> Yes <input checked="" type="checkbox"/> No	500 ml
Escherichia coli K-12	<input type="checkbox"/> Yes <input checked="" type="checkbox"/> No	<input type="checkbox"/> Yes <input checked="" type="checkbox"/> No	<input type="checkbox"/> Yes <input checked="" type="checkbox"/> No	up to 10 liters
	<input type="checkbox"/> Yes <input type="checkbox"/> No	<input type="checkbox"/> Yes <input type="checkbox"/> No	<input type="checkbox"/> Yes <input type="checkbox"/> No	
	<input type="checkbox"/> Yes <input type="checkbox"/> No	<input type="checkbox"/> Yes <input type="checkbox"/> No	<input type="checkbox"/> Yes <input type="checkbox"/> No	

1.3 For above named organism(s) circle HC or CFIA Containment Level required. 1 **2** 3

1.4 Source of microorganism? Burkholderia is naturally in the soil; we also work with clinical isolates from patients with cystic fibrosis. E. coli K-12 is strictly a lab organism

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 cells that will be grown in culture in the table below

Cell Type	Is this cell type used in your work?	Established or Primary *
Human macrophages	<input checked="" type="checkbox"/> Yes <input type="checkbox"/> No	<input checked="" type="checkbox"/> Established <input type="checkbox"/> Primary
Rodent macrophages	<input checked="" type="checkbox"/> Yes <input type="checkbox"/> No	<input checked="" type="checkbox"/> Established <input checked="" type="checkbox"/> Primary
Non-human primate	<input type="checkbox"/> Yes <input checked="" type="checkbox"/> No	<input type="checkbox"/> Established <input type="checkbox"/> Primary
Other (specify)		

* i.e. derived from fresh tissue

2.3 Supplier of primary cell culture tissue Bone marrow

2.4 List specific cell lines to be used and source/supplier: THP1, RAW, PU5.1

2.5 For above named cell types(s) circle HC or CFIA containment level required 1 **2** 3

1.2 (Cont.)

a) B. cepacia is an opportunistic bacterium that is only known to cause infections in humans who are immunocompromised. There is only one report in the literature about a case of cow mastitis in an isolated herd in Spain.

b) E. coli K-12 is the workhorse of molecular biologists: absolutely harmless.

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 if the following will be used in the laboratory

- ◆ Human blood (whole) or other bodily fluids YES NO If YES, Specify _____
- ◆ Human blood (fraction) or other bodily fluids YES NO If YES, Specify _____
- ◆ Human organs (unpreserved) YES NO If YES, Specify _____
- ◆ Human tissues (unpreserved) YES NO If YES, Specify _____

3.3 Is human source known to be infected with and infectious agent YES NO
If YES , please name infectious agent _____

3.4 For above named materials circle HC or CFIA containment level required. 1 2 3

4.0 Genetically Modified Organisms and Cell lines

4.1 Will genetic modifications be made to the organism or cell line ? YES NO
If no, please proceed to Section 5.0

4.2 Will genetic sequences from the following be involved:

- ◆ HIV YES NO
if YES specify _____
- ◆ HTLV 1 or 2 or genes from any CDC class 1 pathogens YES NO
if YES specify _____
- ◆ Other human or animal pathogen and or their toxins YES NO
if YES specify _____

4.3 Will intact genetic sequences be used from

- ◆ SV 40 Large T antigen YES NO If YES specify _____
- ◆ Known oncogenes YES NO If YES specify _____

4.4 Will a live vector(s) (viral or bacterial) be used for gene transduction YES NO
If YES name virus _____

4.5 List specific vector(s) to be used: _____

4.6 Will virus be replication defective YES NO

4.7 Will virus be infectious to humans or animals YES NO

4.8 Will this be expected to increase the Containment Level required YES NO

5.0 Human Gene Therapy Trials

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

If no, please proceed to Section 6.0

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

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

5.3 How will the virus be administered? _____

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

5.5 Has human ethics approval been obtained? YES NO

6.0 Animal Experiments

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

If no, please proceed to section 7.0

6.2 Name of animal species to be used inbred mice

6.3 AUS protocol # 2006-051-03

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

7.0 Use of Animal species with Zoonotic Hazards

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

- ◆ Pound source dogs YES NO
- ◆ Pound source cats YES NO
- ◆ Sheep or goats YES NO
- ◆ Non- Human Primates YES NO If YES specify species _____
- ◆ Wild caught animals YES NO If YES specify species _____

col # _____

8.0 Biological Toxins

8.1 Will toxins of biological origin be used? YES NO

If no, please proceed to Section 9.0

8.2 If YES, please name the toxin _____

8.3 What is the LD₅₀ (specify species) of the toxin _____

9.0 Import Requirements

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

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

9.3 Has an import permit been obtained from CFIA for animal pathogens? YES NO Not required

9.4 Has the import permit been sent to OHS? YES NO
If yes, Permit # _____ Copy(ies) of the permit(s) are sent to OHS upon approval by HC, but we have none active at present

10.0 Training Requirements for Personnel named on Form

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

- ◆ Biosafety
- ◆ Laboratory and Environmental/Waste Management Safety
- ◆ WHMIS

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

SIGNATURE Verified by e-mail (attached)

11.0 Containment Levels

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

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

11.3 If yes, please give the date and permit number: Spring 2004 - I have never received a permit number
B10-UWO-008220

12.0 Approvals

UWO Biohazard Subcommittee

Signature [Signature] Date 2 Oct. '06

Safety Officer for Institution where experiments will take place

Signature Jennifer Stanley Date Sept 29/06

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

Signature _____ Date _____

G. SUMMARY OF PROPOSED RESEARCH

Please provide a 1-page summary of the rationale, general objectives, and specific goals of the proposed research.

Burkholderia cepacia complex (Bcc) infections are an important concern to the health of patients with cystic fibrosis (CF) for three principal reasons: (i) infected patients often deteriorate more rapidly and can develop a life-threatening necrotizing pneumonia (cepacia syndrome); (ii) treatment of these infections is usually very difficult because *B. cepacia* isolates are intrinsically resistant to most clinically relevant antibiotics; and (iii) unlike *P. aeruginosa*, some Bcc isolates are transmissible from patient to patient.

While there is an urgent need to develop new therapies to combat these bacteria, it is also imperative to gain a clear understanding of the pathogenesis of the respiratory infection in CF patients. **Our long-term goals are to discern how Bcc (particularly *B. cenocepacia*) persist in the airways of CF patients, and ultimately to devise strategies for prevention of chronic infection and its deleterious effects on pulmonary tissue.** We have shown that Bcc isolates survive within amoebae and macrophages. Prolonged intracellular survival of Bcc bacteria in phagocytic cells occurs within a modified acidic vacuole that does not fuse with lysosomes, and also in the presence of an oxidative burst. Intracellular survival is **not** associated **with bacterial replication**. These properties distinguish Bcc bacteria from other "classical" intracellular microbes and provide a new paradigm of bacteria-host cell interactions.

We are particularly interested in *B. cenocepacia* isolates from the ET12 lineage since they comprise the most prevalent Bcc complex species in Canada. Thus, we propose to investigate three important questions arising from our research: **(i) How do intracellular *B. cenocepacia* redirect the maturation of phagosomes? (ii) How do *B. cenocepacia* resist oxidative killing mechanisms? (iii) How do infected macrophages respond to intracellular infection by *B. cenocepacia*?**

Our central hypothesis is that the combined effects of intracellular survival and bacterial resistance to oxidative damage in CF lung tissue contribute to enhance the persistence of *B. cenocepacia* in the airways and their ability to elicit a sustained inflammatory response.

Therefore, we propose to achieve the following specific objectives:

1) Characterization of intracellular survival of *B. cenocepacia* in macrophages. This involves: a) Construction of mutants in genes encoding type III and type IV secretory systems, mutants in genes encoding putative effector molecules, and the characterization of these mutants in relation to intracellular survival; and b) Assessing bacterial intracellular survival within macrophages *in vivo* employing the rat agar bead and the *CFTR*^{-/-} mouse lung infection models.

2) Characterization of macrophage responses to intracellular *B. cenocepacia*. This involves: a) Determining the role of the catalase/oxidase KatA and KatB enzymes in survival of *B. cenocepacia*; b) Investigating the interactions of the *B. cenocepacia*-containing vacuoles (BCVs) with the NADPH oxidase complex; c) Examining the role of reactive nitrogen species in the intracellular survival of *B. cenocepacia*; and d) Characterizing macrophage cytokine expression and surface receptor expression profiles in *B. cenocepacia*-infected macrophages.

We believe that macrophage-*B. cenocepacia* interactions are a key component in the pathogenesis of infections in patients with CF. It is becoming increasingly evident that tissue damage following sustained inflammation of the airways is a hallmark in the deterioration of the respiratory function in these patients. We feel the proposed research may result in a detailed understanding of macrophage-*B. cenocepacia* interactions and will pave the way for the development of novel approaches to curtail infection and/or inflammation induced by these bacteria. The experiments build on our previous research and we have generated sufficient preliminary data supporting most of the experimental approaches proposed. We have established the majority of the new tools necessary to achieve the experimental goals and enlisted key collaborators in the areas where we need additional expertise.

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

Subject:Re: Shigella containment requirements?

Date:Thu, 05 Mar 2009 08:24:59 -0500

From:ImportZoopath <ImportZoopath@inspection.gc.ca>

To:Jennifer Stanley <jstanle2@uwo.ca>

Hello Jennifer,

Shigella spp. is considered CL2. They only need a CL2 lab to work with it (though would need a permit to import it).

I hope that helps.

Have a good day,

-Andrew Halliday

Office of Biohazard Containment & Safety, CFIA | Bureau
du confinement des biorisques et de la sécurité, ACIA
Government of Canada | Gouvernement du Canada
59 Camelot, Ottawa ON K1A0Y9
Phone/Tél.: (613) 221-7068
Fax/ Téléc.: (613) 228-6129
ImportZoopath@inspection.gc.ca

Please visit our website at:

<http://www.inspection.gc.ca/english/sci/bio/bioe.shtml>

Veillez visiter notre site internet au:

<http://www.inspection.gc.ca/francais/sci/bio/biof.shtml>