

**The University of Western Ontario**  
**BIOLOGICAL AGENTS REGISTRY FORM**  
**Approved Biohazards Subcommittee: August 12, 2011**  
**Biosafety Website: [www.uwo.ca/humanresources/biosafety/](http://www.uwo.ca/humanresources/biosafety/)**

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

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

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

Electronically completed forms are to be submitted to Occupational Health and Safety, (OHS), (Support Services Building, Room 4190 or to [jstanle2@uwo.ca](mailto:jstanle2@uwo.ca)) for distribution to the Biohazards Subcommittee. For questions regarding this form, please contact the Biosafety Officer at extension 81135 or [biosafety@uwo.ca](mailto:biosafety@uwo.ca). If there are changes to the information on this form (excluding grant title and funding agencies), contact Occupational Health and Safety for a modification form. See website: [www.uwo.ca/humanresources/biosafety/](http://www.uwo.ca/humanresources/biosafety/).

Please ensure that all questions are fully and clearly answered. Failure to do so will lead to the form being returned, which will cause delays in your approval and frustration for you and your colleagues on the Committee.

**If you are re-submitting this form as requested by the Biohazards Subcommittee, please make modifications to the form in bold print, highlighted in yellow. Please re-submit forms electronically.**

|                            |   |
|----------------------------|---|
| PRINCIPAL INVESTIGATOR:    | <b>Dr. Argyrios Margaritis</b>              |
| DEPARTMENT:                | <b>Chemical and Biochemical Engineering</b> |
| ADDRESS:                   | <b>TEB 377</b>                              |
| PHONE NUMBER:              | <b>519-661-2146</b>                         |
| EMERGENCY PHONE NUMBER(S): | “                      ”                    |
| EMAIL:                     | <b>amarg@uwo.ca</b>                         |

Location of experimental work to be carried out :

|                            |                                      |
|----------------------------|--------------------------------------|
| Building : <b>SEB, TEB</b> | Room(s): <b>2035, 2033 &amp; 313</b> |
| Building : _____           | Room(s): _____                       |
| Building : _____           | Room(s): _____                       |

**\*For work being performed at Institutions affiliated with the University of Western Ontario, the Safety Officer for the Institution where experiments will take place must sign the form prior to its being sent to the University of Western Ontario Biosafety Officer (See Section 15.0, Approvals).**

FUNDING AGENCY/AGENCIES: **NSERC**

GRANT TITLE(S): **LIGNOCELLULOSIC BUTANOL PRODUCTION FROM ANGRICULTURAL WASTE RESIDUES USING SOLVENTOGENIC CLOSTRIDIA**

UNDERGRADUATE COURSE NAME(IF APPLICABLE): \_\_\_\_\_

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

| Name                     | UWO E-mail Address     | Date of Biosafety Training |
|--------------------------|------------------------|----------------------------|
| <b>Dr. Peter Kilonzo</b> | <b>pkilonz2@uwo.ca</b> | <b>2005-Present</b>        |
| Postdoc                  | _____                  | _____                      |
| _____                    | _____                  | _____                      |
| _____                    | _____                  | _____                      |



### **Retrieval of Frozen Bacterial Stocks**

**Never thaw frozen bacterial stocks in DMSO or glycerol. Use sterile loop, sterile wooden stick, or sterile disposable pipette to scratch the surface of the stock. Streak appropriate agar plates (e.g., LB agar plates) for single colonies. Recap the frozen stock and return it to storage at -80°C. Incubate the plate overnight at 37°C. The colonies on a plate can be used for up to 1 week to inoculate cultures. Plates should be stored upside down at 4°C during this time.**

**All preparation work is done within the laminar sterile hood Biosafety cabinet level 1. Unused liquid media are collected and stored in labeled waste bottles. Petri-dishes that have the remaining of solid media are autoclaved and stored in special bags for disposal. Remains of fermentation broths are autoclaved at 121°C for 60 min prior to disposal. Wire loops, wooden sticks, or pipettes used during cell culture preparation are normally sterilized by autoclaving under similar conditions.**

**Clostridium beijerinckii will be used to ferment soluble starch to butanol, acetone and ethanol. The anaerobic fermentation data will be used to develop a new kinetic model that relates the growth of Clostridium beijerinckii cells and production of butanol, acetone and, ethanol at different pH conditions and starch concentrations.**

**Please include a ONE page research summary or teaching protocol in lay terms.  
Forms with summaries more than one page will not be reviewed.**

#### **Abstract:**

**Clostridium acetobutylicum ATCC 824 and C. beijerinckii are grown on a variety of different sugars [D-(+)-glucose, D-(+)-xylose, D-(+)-mannose, D-(+)-galactose, D-(-)-arabinose, and D-(+)-cellobiose] found in agricultural waste hydrolysates and assayed for acetone, butanol, and ethanol (ABE) production. The order of sugar utilization by the culture is D-(+)-glucose > D-(+)-cellobiose > D-(+)-mannose > D-(+)-xylose > D-(-)-arabinose > D-(+)-galactose. The high D-(+)-glucose utilization of 98% results in higher cell density of 2.49 g/L with the highest growth rate of 0.293 h<sup>-1</sup>, than other sugars. In this system, ABE concentration of 18.3 g/L, is triggered by a total acid concentration of 11.5 g/L, but growth cessation takes place at a total butanol and acid concentration of 24.1 g/L. Although the culture utilizes 92% sugar from D-(+)-cellobiose and 86% from D-(+)-xylose, the resultant biomass concentration (1.53 and 1.82 g/L) is very low. In these systems, a total acid concentration between 14 and 15 g/L triggers 19.0 and 13.9 g/L ABE g/L from D-(+)-cellbiose and D-(+)-xylose, respectively. Relatively high yield (0.33 g/g), productivity (0.47 g/L.h), and low growth rate of 0.069 h<sup>-1</sup> results from the D-(+)-cellobiose system. In both systems, growth cessation occurs at a total butanol and acid concentration between 25 and 27 g/L. Culture grown on mixed sugars utilizes 70-90% total sugars. A total acid concentration between 23 and 28 g/L triggers only 13-16 g/L ABE, with relatively high yield and productivity of 0.37 and 0.43 g/L.h, whereas growth inhibition occurs at a total butanol and acid concentration between 30 and 39 g/L.**

**This research continues to obtain more kinetic data in order to develop the kinetic model for cell growth of Clostridium acetobutylicum at different sugar concentrations.**

**acetobutylicum ATCC 824**

#### **2 Abstract**

**Production of glucoamylase by recombinant Saccharomyces cerevisiae C468/pGAC9 (ATCC 20690) in a continuous stirred tank bioreactor is studied at different dilution rates. Plasmid stability is found to be growth (dilution rate) dependent; it increases with the dilution rate. Bioreactor productivity and specific productivity also increase with the dilution rate. A kinetic equation is used to model the plasmid stability kinetics. The growth rate ratio between plasmid-carrying and plasmid-free cells decreases from 1.397 to 1.215, and segregational instability or probability of plasmid loss from each cell division decreases from 0.059 to 0.020 as the dilution rate increases from 0.10 to 0.37 1/h. The specific growth rates increase with dilution rate, while the growth rate difference between plasmid-carrying and plasmid-free cell populations is negligible. This is attributed to the low copy number of the hybrid plasmid pGAC9. Thus, the growth rate has no significant effect on plasmid instability. The proposed kinetics is consistent with experimental results, and the model simulates the experimental data well.**

## 1.0 Microorganisms

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

Do you use microorganisms that require a permit from the CFIA?  YES  NO

If YES, please give the name of the species \_\_\_\_\_

What is the origin of the microorganism(s)? \_\_\_\_\_

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

*Please attach the CFIA permit.*

Please describe any CFIA permit conditions:

N/A

1.2 Please complete the table below:

| Full Scientific Name of Biological Agent(s)* (Be specific) | Is it known to be a human pathogen? YES/NO                             | Is it known to be an animal pathogen? YES/NO                           | Is it known to be a zoonotic agent? YES/NO                             | Maximum quantity to be cultured at one time? (in Litres) | Source/Supplier | PHAC or CFIA Containment Level   |
|--|--|--|--|--|-----------------|--|
| <i>Clostridium acetobutylicum</i> ATCC 824*                | <input type="checkbox"/> Yes<br><input checked="" type="checkbox"/> No | <input type="checkbox"/> Yes<br><input checked="" type="checkbox"/> No | <input type="checkbox"/> Yes<br><input checked="" type="checkbox"/> No | 210 mL   | ATCC            | <input checked="" type="checkbox"/> 1 <input type="checkbox"/> 2<br><input type="checkbox"/> 2+ <input type="checkbox"/> 3 |
| <i>Clostridium beijerinckii</i> **                         | <input type="checkbox"/> Yes<br><input checked="" type="checkbox"/> No | <input type="checkbox"/> Yes<br><input checked="" type="checkbox"/> No | <input type="checkbox"/> Yes<br><input checked="" type="checkbox"/> No |  |                 | <input checked="" type="checkbox"/> 1 <input type="checkbox"/> 2<br><input type="checkbox"/> 2+ <input type="checkbox"/> 3 |
| <i>Saccharomyces cerevisiae</i> ***                        | <input type="checkbox"/> Yes<br><input checked="" type="checkbox"/> No | <input type="checkbox"/> Yes<br><input checked="" type="checkbox"/> No | <input type="checkbox"/> Yes<br><input checked="" type="checkbox"/> No |  |                 | <input checked="" type="checkbox"/> 1 <input type="checkbox"/> 2<br><input type="checkbox"/> 2+ <input type="checkbox"/> 3 |
|  | <input type="checkbox"/> Yes<br><input type="checkbox"/> No            | <input type="checkbox"/> Yes<br><input type="checkbox"/> No            | <input type="checkbox"/> Yes<br><input type="checkbox"/> No            |  |                 | <input type="checkbox"/> 1 <input type="checkbox"/> 2<br><input type="checkbox"/> 2+ <input type="checkbox"/> 3            |
|  | <input type="checkbox"/> Yes<br><input type="checkbox"/> No            | <input type="checkbox"/> Yes<br><input type="checkbox"/> No            | <input type="checkbox"/> Yes<br><input type="checkbox"/> No            |  |                 | <input type="checkbox"/> 1 <input type="checkbox"/> 2<br><input type="checkbox"/> 2+ <input type="checkbox"/> 3            |
|  | <input type="checkbox"/> Yes<br><input type="checkbox"/> No            | <input type="checkbox"/> Yes<br><input type="checkbox"/> No            | <input type="checkbox"/> Yes<br><input type="checkbox"/> No            |  |                 | <input type="checkbox"/> 1 <input type="checkbox"/> 2<br><input type="checkbox"/> 2+ <input type="checkbox"/> 3            |
|  | <input type="checkbox"/> Yes<br><input type="checkbox"/> No            | <input type="checkbox"/> Yes<br><input type="checkbox"/> No            | <input type="checkbox"/> Yes<br><input type="checkbox"/> No            |  |                 | <input type="checkbox"/> 1 <input type="checkbox"/> 2<br><input type="checkbox"/> 2+ <input type="checkbox"/> 3            |
|  | <input type="checkbox"/> Yes<br><input type="checkbox"/> No            | <input type="checkbox"/> Yes<br><input type="checkbox"/> No            | <input type="checkbox"/> Yes<br><input type="checkbox"/> No            |  |                 | <input type="checkbox"/> 1 <input type="checkbox"/> 2<br><input type="checkbox"/> 2+ <input type="checkbox"/> 3            |

\*Please attach a Material Safety Data Sheet or equivalent from the supplier if the bacterium used is not on this link:  
[http://www.uwo.ca/humanresources/docandform/docs/ohs/CFIA\\_Ecoli\\_list.pdf](http://www.uwo.ca/humanresources/docandform/docs/ohs/CFIA_Ecoli_list.pdf)

Additional Comments: \* **Production of biofuel-biobutanol from sugars**  
 \*\* **Production of biofuel biobutanol from starch**  
 \*\*\***Production of enzymes from sugars**

## 2.0 Cell Culture

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

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

| Cell Type         | Is this cell type used in your work?                     | Source of Primary Cell Culture Tissue | AUS Protocol Number |
|-------------------|--|---------------------------------------|---------------------|
| Human             | <input type="checkbox"/> Yes <input type="checkbox"/> No |                                       | Not applicable      |
| Rodent            | <input type="checkbox"/> Yes <input type="checkbox"/> No |                                       |                     |
| Non-human primate | <input type="checkbox"/> Yes <input type="checkbox"/> No |                                       |                     |
| Other (specify)   | <input type="checkbox"/> Yes <input type="checkbox"/> No |                                       |                     |

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

| Cell Type         | Is this cell type used in your work?                     | Specific cell line(s)* | Containment Level of each cell line | Supplier / Source of cell line(s) |
|-------------------|--|------------------------|-------------------------------------|-----------------------------------|
| Human             | <input type="checkbox"/> Yes <input type="checkbox"/> No |                        |                                     |                                   |
| Rodent            | <input type="checkbox"/> Yes <input type="checkbox"/> No |                        |                                     |                                   |
| Non-human primate | <input type="checkbox"/> Yes <input type="checkbox"/> No |                        |                                     |                                   |
| Other (specify)   | <input type="checkbox"/> Yes <input type="checkbox"/> No |                        |                                     |                                   |

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

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

Additional Comments: \_\_\_\_\_

## 3.0 Use of Human Source Materials

3.1 Does your work involve the use of human source materials?  YES  NO  
 If no, please proceed to Section 4.0

3.2 Indicate in the table below the Human Source Material to be used.

| Human Source Material                      | Source/Supplier /Company Name | Is Human Source Material Infected With An Infectious Agent?<br>YES/UNKNOWN | Name of Infectious Agent (If applicable) | PHAC or CFIA Containment Level (Select one)   |
|--|-------------------------------|--|--|---|
| Human Blood (whole) or other Body Fluid    |                               | <input type="checkbox"/> Yes<br><input type="checkbox"/> Unknown           |  | <input type="checkbox"/> 1 <input type="checkbox"/> 2<br><input type="checkbox"/> 2+ <input type="checkbox"/> 3 |
| Human Blood (fraction) or other Body Fluid |                               | <input type="checkbox"/> Yes<br><input type="checkbox"/> Unknown           |  | <input type="checkbox"/> 1 <input type="checkbox"/> 2<br><input type="checkbox"/> 2+ <input type="checkbox"/> 3 |
| Human Organs or Tissues (unpreserved)      |                               | <input type="checkbox"/> Yes<br><input type="checkbox"/> Unknown           |  | <input type="checkbox"/> 1 <input type="checkbox"/> 2<br><input type="checkbox"/> 2+ <input type="checkbox"/> 3 |
| Human Organs or Tissues (preserved)        |                               | Not Applicable   |  | Not Applicable  |

Additional Comments: \_\_\_\_\_

**4.0 Genetically Modified Organisms and Cell lines**

4.1 Will genetic modifications be made to the microorganisms, biological agents, or cells described in Sections 1.0 and 2.0?  YES  NO If **NO**, please proceed to Section 5.0

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

| Bacteria Used for Cloning *         | Plasmid(s) ** | Source of Plasmid                          | Gene Transformed or Transfected                            | Will there be a change due to transformation of the bacteria?  | Will there be a change in the pathogenicity of the bacteria after the genetic modification? | What are the consequences due to the transformation of the bacteria?  |
|-------------------------------------|---------------|--|--|--|---|---|
| <b>Saccharomyces cerevisiae 468</b> | <b>pGAC9</b>  | <b>yeast 2µ plasmid (2µ micron circle)</b> | <b>LEU gene glucoamylase gene from Aspergillus awamori</b> | <b>The S. cerevisiae host strain C468 (aleu2-3 leu2-112 his311 his3-15 mal-) (ATCC 62995) is haploid, with auxotrophic markers for leucine and histidine and carries mutation (mal-) blocking the utilization of maltose as carbon source.</b> |   | <b>Therefore, the host cell is complementary to the leucine prototrophy by inserting the selectable marker (LEU 2) into the expression plasmid and the presence of the glucoamylase gene on the plasmid allows the host cell to grow on maltose</b> |

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

\*\* Please attach a plasmid map.

\*\*\*No Material Safety Data Sheet is required for the following strains of E. coli:

[http://www.uwo.ca/humanresources/docandform/docs/ohs/CFIA\\_Ecoli\\_list.pdf](http://www.uwo.ca/humanresources/docandform/docs/ohs/CFIA_Ecoli_list.pdf)

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

YES, complete table below  NO

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

\* Please attach a Material Safety Data Sheet or equivalent.

4.3.1 Will virus be replication defective?  YES  NO

4.3.2 Will virus be infectious to humans or animals?  YES  NO

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

**5.0 Will genetic sequences from the following be involved?**

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

5.1 Is any work being conducted with prions or prion sequences?  NO  YES

Additional Comments: \_\_\_\_\_

## 6.0 Human Gene Therapy Trials

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

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

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

6.4 How will the biological agent be administered?

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

6.6 Has human ethics approval been obtained?  YES, number:  NO  PENDING

## 7.0 Animal Experiments

7.1 Will live animals be used?  YES  NO If NO, please proceed to section 8.0

7.2 Name of animal species to be used

7.3 AUS protocol #

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

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

## 8.0 Use of Animal species with Zoonotic Hazards

8.1 Will any animals with zoonotic hazards or their organs, tissues, lavages or other body fluids including blood be used (see list below)?  YES  NO - If NO, please proceed to section 9.0

8.2 Will live animals be used?  YES  NO

8.3 If YES, please specify the animal(s) used:

- |                             |  |                             |
|-----------------------------|--|-----------------------------|
| ◆ Pound source dogs         | <input type="checkbox"/> YES                     | <input type="checkbox"/> NO |
| ◆ Pound source cats         | <input type="checkbox"/> YES                     | <input type="checkbox"/> NO |
| ◆ Cattle, sheep or goats    | <input type="checkbox"/> YES, species            | <input type="checkbox"/> NO |
| ◆ Non-human primates        | <input type="checkbox"/> YES, species            | <input type="checkbox"/> NO |
| ◆ Wild caught animals       | <input type="checkbox"/> YES, species & colony # | <input type="checkbox"/> NO |
| ◆ Birds                     | <input type="checkbox"/> YES, species            | <input type="checkbox"/> NO |
| ◆ Others (wild or domestic) | <input type="checkbox"/> YES, specify            | <input type="checkbox"/> NO |

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

## 9.0 Biological Toxins and Hormones

9.1 Will toxins or hormones of biological origin be used?  YES  NO If NO, please proceed to Section 10.0

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

9.3 What is the LD<sub>50</sub> (specify species) of the toxin or hormone

9.4 How much of the toxin or hormone is handled at one time\*?

9.5 How much of the toxin or hormone is stored\*?

9.6 Will any biological toxins or hormones be used in live animals?  YES  NO  
If YES, Please provide details:

\*For information on biosecurity requirements, please see:  
[http://www.uwo.ca/humanresources/docandform/docs/healthandsafety/biosafety/Biosecurity\\_Requirements.pdf](http://www.uwo.ca/humanresources/docandform/docs/healthandsafety/biosafety/Biosecurity_Requirements.pdf)

Additional Comments: \_\_\_\_\_

## 10.0 Insects

10.1 Do you use insects?  YES  NO - If NO, please proceed to Section 11.0

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

10.3 What is the origin of the insect?

10.4 What is the life stage of the insect?

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

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

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

## 11.0 Plants

- 11.1 Do you use plants?  YES  NO - If NO, please proceed to Section 12.0
- 11.2 If YES, please give the name of the species.
- 11.3 What is the origin of the plant?
- 11.4 What is the form of the plant (seed, seedling, plant, tree...)?
- 11.5 What is your intention?  Grow and maintain a crop  "One-time" use
- 11.6 Do you do any modifications to the plant?  YES  NO  
If yes, please describe:
- 11.7 Please describe the risk (if any) of loss of the material from the lab and how this will be mitigated:
- 11.8 Is the CFIA permit attached?  YES  NO  
If YES, Please attach the CFIA permit & describe any CFIA permit conditions:

## 12.0 Import Requirements

- 12.1 Will any of the above agents be imported?  YES, country of origin  NO  
If NO, please proceed to Section 13.0
- 12.2 Has an Import Permit been obtained from HC for human pathogens?  YES  NO
- 12.3 Has an import permit been obtained from CFIA for animal or plant pathogens?  YES  NO
- 12.4 Has the import permit been sent to OHS?  YES, please provide permit #  NO

## 13.0 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.

**An X in the check box indicates you agree with the above statement...**   
**Enter Your Name** \_\_\_\_\_ **Date:** \_\_\_\_\_

**14.0 Containment Levels**

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

14.2 Has the facility been certified by OHS for this level of containment?  
 YES, location and date of most recent biosafety inspection: **MARCH 01/2011**  
 NO, please certify  
 NOT REQUIRED for Level 1 containment

14.3 Please indicate permit number (not applicable for first time applicants):

**15.0 Procedures to be Followed**

15.1 Are additional risk reduction measures necessary beyond containment level 1, 2, 2+ or 3 measures that are unique to these agents?  YES  NO  
If **YES** please describe:

15.2 Please outline what will be done if there is an exposure to the biological agents listed such as a needlestick injury or an accidental splash:  
**In case of mucous membrane or skin exposure wash well with soap and water. Complete accident/incident report. Seek medical attention if needed.**

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

*An X in the check box indicates you agree with the above statement...*

Enter Your Name \_\_\_\_\_ Date:                       
                      
15.4 Additional Comments:                       
                    

**16.0 Approvals**

1) UWO Biohazards Subcommittee: SIGNATURE: \_\_\_\_\_  
Date: \_\_\_\_\_

2) Safety Officer for the University of Western Ontario SIGNATURE: \_\_\_\_\_  
Date: \_\_\_\_\_

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

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

Special Conditions of Approval:

## Bacteria

ATCC<sup>®</sup> Number: **824<sup>TM</sup>** [Order this Item](#) Price: **\$205.00**

Organism: *Clostridium acetobutylicum* McCoy et al. emend. Keis et al. deposited as *Granulobacter pectinovorum* (Stormer) Beijerinck

Designations: [CCRC 10639, CCUG 42182, DSM 792, IAM 19013, IFO 13948, JCM 1419, KCTC 1790, L.S. McClung 2291, LMG 5710, McCoy and McClung strain W, NCCB 29024, NCCB 84048, NCIMB 8052, VKM B-1787]

Isolation: plant-derived foodstuff (corn meal)

Depositor: ER Weyer

Biosafety Level: 1

Shipped: freeze-dried

Growth Conditions: ATCC medium 2107: Reinforced clostridial broth (modified)

**Temperature:** 37.0°C

**Atmosphere:** Anaerobic

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 Links ▶**

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## Bacteria

ATCC<sup>®</sup> Number: **858<sup>TM</sup>** [Order this Item](#) Price: **\$255.00**

Organism: *Clostridium beijerinckii* Donker emend. Keis et al.  
 Designations: LMD 25.10 [NCIB 11373, NCTC 2264, VPI 11896]  
 Isolation: pasteurized garden soil  
 Depositor: J van der Toorn  
 History: ATCC <<<--J van der Toorn<<<--H.J.L. Donker 7 (<<<-- A.J. Kluver <<<-- H.J.L. Donker)  
Biosafety Level: 1  
 Shipped: freeze-dried  
 Growth Conditions: ATCC medium38: Beef liver medium for anaerobes  
**Temperature:** 37.0°C  
 Duration: anaerobic  
 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.  
 References: 5543: Donker HJLBijdrage tot de Kennis der Botersuur-, butylacohlen acetonigistingen Ph.D. thesis, Delft Univ. Technol., 1926

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## Fungi ,Yeasts and Yeast Genetic Stock

ATCC® Number: **62995™**  Price: **\$275.00**

Organism: *Saccharomyces cerevisiae* Meyen ex E.C. Hansen deposited as *Saccharomyces cerevisiae* Hansen, teleomorph

Alternate State: *Candida robusta* Diddens et Lodder

Designations: NRC 5140 [ATCC 66527, C468, CMCC 1398, LL20, NCYC 1445]

Depositors: RK Latta

Biosafety Level: 1

Shipped: frozen

Genotype/ORF/  
Gene Name: MATalpha leu2-3 leu2-112 his3-11 his3-15 [psi+] [cir+]

Growth Conditions: ATCC medium 1245: YEPD  
**Temperature:** 25.0°C

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 [20995] [20124]

Mating Type: alpha

Karyotype: Ploidy: haploid

Comments: Expression of *Aspergillus awamori* glucoamylase gene [20314] [21188]

Sensitive to *Kluyveromyces lactis* toxin [20534]

Subcollection: Yeasts

References:

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## Clone

ATCC<sup>®</sup> Number: **20690** [Order this Item](#) Price: **\$275.00**

Designation: pGAC9 [pYepGAC9]  
 Depositors: Cetus Corp., A Belt, Cetus Corp.  
 Insert Source: *Saccharomyces cerevisiae* Meyen ex E.C. Hansen  
 DNA: cDNA  
 Insert Information: Insert lengths(kb): 2.109999895095825  
 Gene product: glucoamylase  
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.  
 Size (kb): 10.6700000762939500  
 Vector: pAC1 (plasmid)  
 Promoters: Promoter enolase I  
 Construction: pBR322, peno 46, LEU2, 2 micron  
 Marker(s):LEU2,ampR  
 Vector: Construct size (kb): 10.67000007629395  
 Features: marker(s): ampR, LEU2  
 promoter: enolase I  
 replicon: pMB1, 2 micron  
 terminator: enolase I  
 Comments: Production of glucoamylase [[12209](#)]  
 The insert contains the full-length cDNA and 3' poly(A), minus the four introns. [[12209](#)]  
 Media Description: [ATCC medium 1212](#): Yeast synthetic minimal medium  
 References: 12209: Nunberg J, et al. Glucoamylase cDNA. US Patent 4,794,175 dated Dec 27 1988

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