



## **CoBRA** Project Overview

Prepared for: **RBC Cochrane**

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## COCHRANE BIO-RESEARCHERS IN ACTION PROJECT OVERVIEW

### Background

A healthy forest is diverse. It has trees and plants of different ages, species, heights and genetic make-up. Diversity provides good quality wildlife habitat and helps limit the size of wildfires and insect outbreaks.

In the forests of the mountain national parks, the activity of mountain pine beetle plays a role in creating forest diversity. Healthy forests are in a constant state of change in part through natural disturbances like avalanches, landslides, floods, fire and the activity of insects and diseases. Forests have evolved with these processes for thousands of years. Without them, the natural balance is lost.

While mountain pine beetle are a natural part of the southern Rocky Mountain ecosystem, recent beetle outbreaks are larger than those of the past. Decades of fire suppression have created large tracts of older pine forest that provide a highway for beetle expansion. The lack of fire, combined with a recent warming trend, means that the beetles are now occurring where they haven't been observed before: farther east, farther north and at higher elevations. This is cause for concern as the mountain national parks form the margin between the beetle outbreak in British Columbia and commercial forests in Alberta.

It is important to note that the beetle itself doesn't kill the trees and that the detrimental effects associated with the mountain beetle are produced by *Grosmannia Clavigera*, or blue stain fungus (BSF). This fungus causes irreversible damage to the trees internal systems by converting the trees natural defences into a carbon based food source, clogging the various nutrient channels within the tree and finally by using those channels as a means to spread and eventually kill the tree.

Our project will focus on building a bacterial plasmid that will produce and secrete the chitinase enzyme which will in turn be able to break down the chitin rich membranes of the BSF while leaving the tree unaffected and thus able to use its own defences to deal with the mountain pine beetle.

A graphic explaining the interplay between the beetle, fungi and tree is attached as **Appendix A** along with a team photo.

### Objective

Our project goal is to determine if a cloned chitinase cDNA can be successfully expressed in transgenic E.coli. Using recombinant DNA techniques our team will create an entirely new DNA biobrick, this biobrick will be placed in the pSB1C3 vector and contain a specific inducible or constitutive promoter (LacI or TetR), a specific cDNA (one of three class 1 chitinases; PgeChia1-1, PgeChia1-2 and PcChia1-1), and a stop codon or terminator gene. This construct as previously mentioned will be placed into lab grade Top 10 and k12 lab strain E.coli bacteria thus allowing these new, genetically altered bacteria to successfully produce and secrete the chitinase enzyme thus showing proof of concept.

It is important to note that our engineered bacteria will never be used outside of a controlled lab setting.

A visual of our hypothesized plasmid is attached as **Appendix B**.

### Geographical Area Served

In 2012, International Genetically Engineered Machine Foundation (iGEM) spun out of MIT and became an independent nonprofit organization located in Cambridge, Massachusetts, USA. The iGEM Foundation fosters scientific research and education through organizing and operating the iGEM Competition, the premier student synthetic biology competition. It also fosters scientific research and education by establishing and operating the

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## COCHRANE BIO-RESEARCHERS IN ACTION PROJECT OVERVIEW

Registry of Standard Biological Parts, a community collection of biological components. The organization promotes the advancement of science and education by developing an open community of students and practitioners in schools, laboratories, research institutes, and industry. The iGEM community has a long history of involving students and the public in the development of the new field of synthetic biology. (source: <http://igem.org/About>)

Because of the iGEM platform our project has the capacity to serve the students of Cochrane High School, our local community of Cochrane Alberta, Canadian research institutes, and other international and national organizations. Our project looks to create a viable solution to a major industrial problem which has the capacity to cripple the Canadian lumber industry if left unchecked.

### Benefits

This project benefits the students of Cochrane High School directly as they will learn from the scientific inquiry method and new field of synthetic biology education. As the popularity of the project increases, schools from across the county will begin to take notice of our high schools' unprecedented steps towards environmental protection and conservation. Our project can be found at [http://igem.org/Team\\_List?year=2014&division=high\\_school](http://igem.org/Team_List?year=2014&division=high_school)

Our approach to the project has meant that we have made contact with researchers and institutions such as Dr. Joerg Bohlmann, UBC Michael Smith Laboratories; Janice Cooke University of Alberta, and Matt Bryman Network Manager The NSERC TRIA Network (TRIA-Net).

### Budget and Funding

Total Cost of Project: ~\$45,000

We are currently pursuing various local and national grant applications, these applications include:

- Alta Genetics
- ATA
- AB Pacific
- AB Forest Genetic Research Council
- Forest Resource Improvement Association of Alberta (FRIAA)

In addition to these grant programs we are also pursuing local sources of funding from a variety of community organizations and local businesses.

A comprehensive budget and breakdown of expenses is attached as **Appendix C**

### Canadian Revenue Number

Our charitable donation number is 10790 7750RR0001.

### Donation Recognition

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## COCHRANE BIO-RESEARCHERS IN ACTION PROJECT OVERVIEW

Companies and local sponsors will be recognized for their generosity by; being highlighted in any media coverage we receive over the duration of our project, being featured on our projects wiki page as well as on our school website, and finally, being listed as a sponsor on our iGEM project booth at MIT.

### Project Promotion

We will host a series of collaborative open houses with the community to educate them on the benefits and viability of synthetic biology. At this event we will highlight the involvement and contributions of our sponsors and their key role in the success of our project. In addition to these events, we are looking at posting a bi-weekly column in our local newspapers to inform the community of our projects standing and to continually educate the community on the revolutionary new science that is synthetic biology.

### Goals and Measurement of Success

Our goal and measurement of success in this project is to raise awareness and foster public understanding of synthetic biology. We wish to highlight the benefits of synthetic biology, and also the place that the students of today have in this new and innovative branch of science.

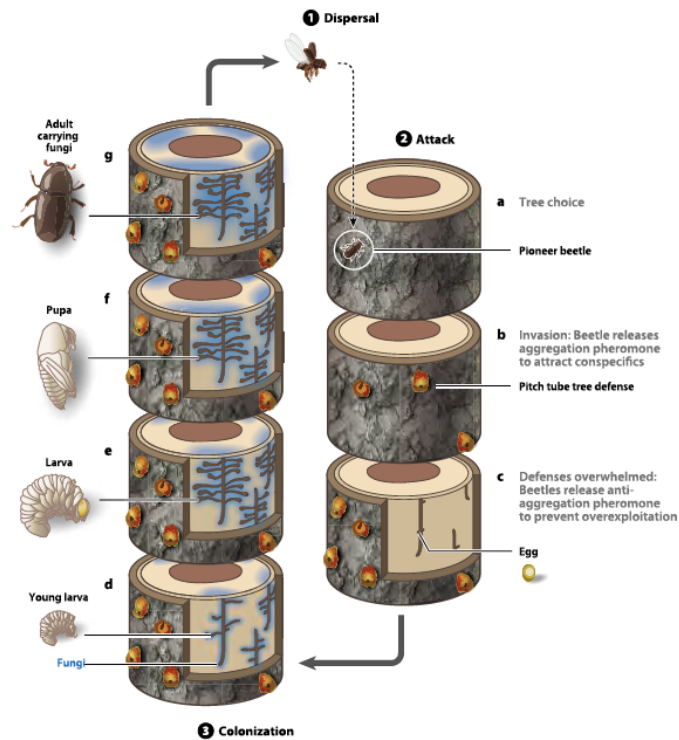
A measure of our success will be the amount of support we get from our local community. Being that one of the goals as specified by iGEM is to develop an open community of students and practitioners, we have already experienced great successes in our research phase by forming contacts with professors, associate professors, and grad students at the University of Calgary, University of Alberta and the University of British Columbia as well as the Canadian TRIA Project.

### Potential Successes at iGEM:

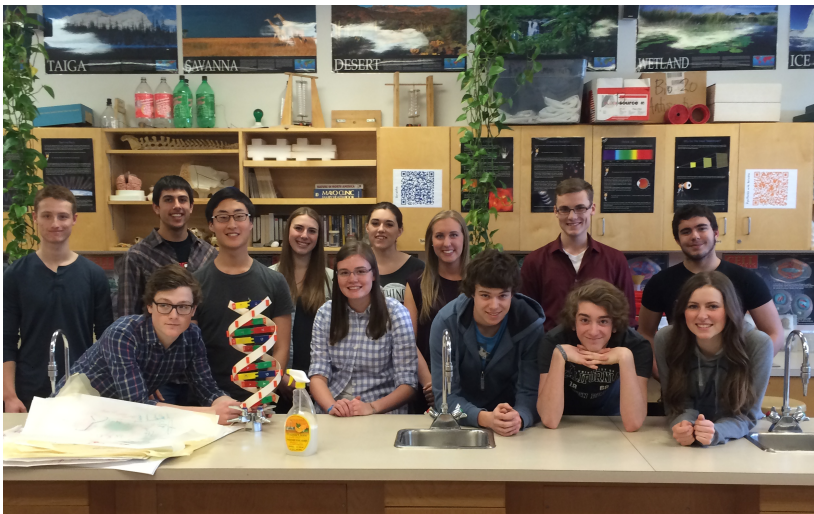
Our project has promise to receive awards in the following categories:

- Best Human Practices Advance
  - Best Model
  - Best New Standard
  - Best New BioBrick Part or Device, Engineered
  - Best Wiki: Best Poster, Regional Level: Best Presentation
  - Best Parts Collection
  - Most Improved Registry Part
  - Best Environmental Project
  - Best Poster: Best Presentation
  - Best Wiki
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## APPENDIX A PROJECT VISUAL AND TEAM PHOTO



Diana L. Six and Michael J. Wingfield (2011) *Annu. Rev. Entomol.* 56:255–72

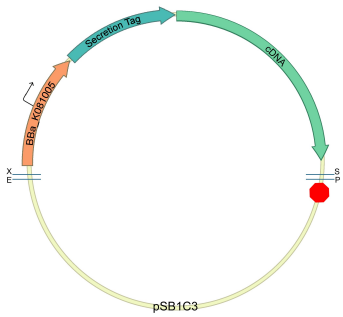
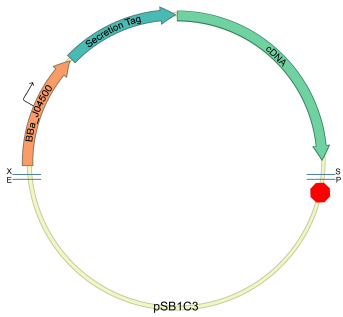


**Back Row (L to R):** Marius Anastasiu, Matt Pipa, Lauren Van Dyke, Katie Goetjen, Kayla Sage, Jacob Rovere, and Joao Areias de Moraes

**Front Row (L to R):** Adam Sibbald, Richard Lee, Autumn Bernard, Ben Luft, Kristian Smits, and Katrina Berube

**Missing:** Rhett Devlin and Cassandra Bourchier

## APPENDIX B PLASMID VISUAL

	BioBrick #1	BioBrick #2
<b>Visual</b>		
<b>Vector</b>	pSB1C3	pSB1C3
<b>cDNA</b>	PgeChia1-1. PgeChia1-2, PcChia1-1	PgeChia1-1. PgeChia1-2, PcChia1-1
<b>Promotor + RBS</b>	TetR + B0030 (BBa_K081005)	LacI + B0034 (BBa_J04500)
<b>Secretion Tag</b>	TBD	TBD
<b>Terminator</b>	BBa_B0015	BBa_B0015

## APPENDIX C BUDGET

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**COCHRANE BIO-RESEARCHERS IN ACTION PROJECT OVERVIEW**
**UPCOMING EXPENDITURES**

Description	Unit Price
Lab Coats	\$ 1,260
Water Bath	\$ 1,500
Hotel and Airfaire	\$ 16,000
iGEM Jamboree Registration	\$ 3,300
Transportation in Boston	\$ 500
<b>Total</b>	<b>\$22,560</b>

**EXPENDITURE THUS FAR**

Description	Unit Price
Lab Materials	\$ 173
Vinyl Gloves	\$ 338
Lunch (Feb. 7th)	\$ 331
Extension Cord	\$ 42
BioTech Equipment	\$ 3,664
iGEM Registration Free	\$ 1,500
Fridge	\$ 40
Autoclave	\$ 3,000
PCR Machine	\$ 8,000
Centrifuge	\$ 1,000
Pipets	\$ 500
Gell Electrophoresis Unit	\$ 2,000
DNA Synthesis and Optimization	\$ 700
<b>Total</b>	<b>\$21,288</b>

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**COCHRANE BIO-RESEARCHERS IN ACTION PROJECT OVERVIEW**
**Funds Secured**

Description	Unit Price
Alberta Innovates Grant	\$ 19,000
Member Fee	\$ 1,500
Private Donations	\$ 3,900
Cu-SEE Equipment Donation	\$ 14,000
<b>Total</b>	<b>\$38,400</b>

**Totals**

Description	Unit Price
Upcoming Expenditures	\$ 22,560
Funds Secured - Expenditures Made	\$ 17,112
Contingency Fund	\$ 3,000
<b>Amount Needed</b>	<b>\$8,448</b>

\*All amounts included in this Budget are current as of March 28th 2014. This data is subject to change as our project progresses.

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