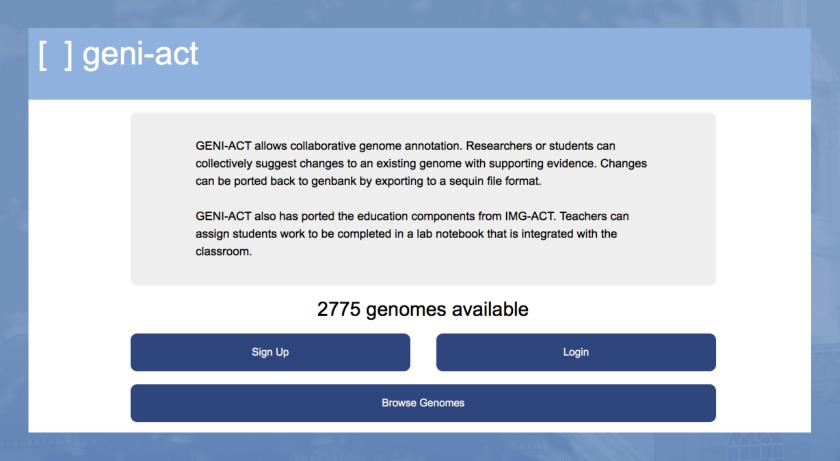
An Introduction to GENI-ACT



Part of the Vision of GEM:

- Engaging the wider WNY community and increasing its genomic and microbiomic literacy will help promote GEM-relevant science in multiple ways, including building public support for research funding and explaining the importance of public participation in clinical research.
- In tandem with their world-class research on these questions, the GEM faculty will develop new courses to educate both STEM and non-STEM oriented students alike, about these important topical issues. UB students will be involved in K-12 and community outreach GEM activities. By fostering partnerships between Buffalo area school teachers and students and researchers at UB, we will form stronger bonds between the community and the university.

https://www.buffalo.edu/genomeenvironmentmicrobiome/about-us/our-vision.html

What is GENI-ACT?

- GENI-ACT is the current version of what initially was the Integrated Microbial Genomes Annotation Collaboration Toolkit (IMG-ACT).
- IMG-ACT was developed by instructors from both research-intensive and predominately undergraduate institutions in collaboration with the Department of Energy-Joint Genome Institute (DOE-JGI) as a means to innovate and update undergraduate education and faculty development in 2008.
- The IMG-ACT program provided a cadre of tools, including access to a clearinghouse of genome sequences, bioinformatics databases, data storage, instructor course management, and student notebooks for organizing the results of their bioinformatic investigations.
- In the process, IMG-ACT made it feasible to provide undergraduate research opportunities to a greater number and diversity of students, in contrast to the traditional mentor-to-student apprenticeship model for undergraduate research, which can be too expensive and time-consuming to provide for every undergraduate.

http://journals.plos.org/plosbiology/article?id=10.1371/journal.pbio.1000448

History of the use of GENI-ACT at UB

- 2008-201: The Department of BCLS had a workforce development grant to give teachers enrichment training in fields related to clinical laboratory sciences and biotechnology
- 2008-2009: ~96 teachers from several counties came to UB for the enrichment training.
- In January of 2010 Dr. Rama Dey-Rao and Dr. Stephen Koury travel to the JGI to receive training in the use of MG-ACT.
- Summer of 2010: Teachers come to UB for 1 week of training in the use of GENI-ACT
- Fall of 2010 Students work with their teachers to annotate genes with guidance from Dr. Dey-Rao and Dr. Koury
- December 2010 students, teachers and their family come to UB for a Saturday minisymposium to present their findings and hear a talk by Dr. Marc Halfon





Timeline Since 2010:

- 2012- Used the workforce development grant experience as preliminary data for an NSF ITEST (Innovative Technology Experiences for Students and Teachers) Grant - not funded.
- 2013 Modified the grant for submission to NIH for a SEPA (Scientific Education Partnership Award)
 - One proposal per institution; one other group wanted to submit
 - · Upon suggestion from Ken Tramposch, Associate Vice President for Research, we partnered with the NY State AHEC (Area Health Education Center) System, housed in Family Medicine, and their two western NY Centers to revise the proposal
 - ENAHEC Erie Niagara AHEC
 - Rural AHEC 12 counties outside of Erie / Niagara
 - Resulted in the formation of the Western New York Genetics in Research and Healthcare Partnership - application submitted and a score was obtained that was likely to be funded.
 - But then came the.....

"United States budget sequestration of 2012"

- SEPA program funds were cut...and the SEPA program in general was threatened with discontinuation
- Program Officer could give no guidance other than to say no awards were going to be given
- Modied the grant as the "Western New York Genetics in Research Partnership" and resubmitted as an NSF ITEST Strategies project
 - Funded in the summer of 2013 for 3 years
- Spring 2014 SEPA program funds restored with the mandate that all projects that were going to be funded in the previous cycle be funded before another program announcement could be made.
 - June 1 2014 SEPA award given after modification of the original proposal to make is sufficiently different from the ITEST award; 5 year award.

NSF ITEST AWARD

Project website:

http://ubwp.buffalo.edu/wnygirp/

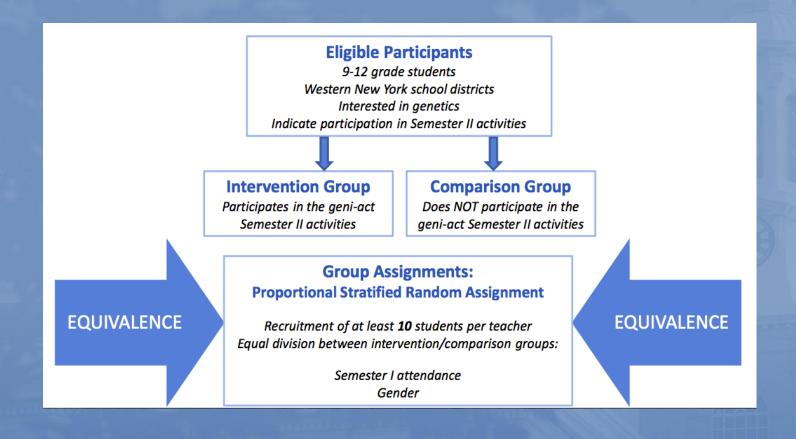
The STEM-related goals of the project are to:

- allow high school students and teachers to participate in scientific research
- stimulate the interest of students in pursuing careers in science and technology through the use of a unique, interactive learning environment combined with intensive support intervention (GENI-ACT)
- encourage teachers to include bioinformatics and genomics in their curriculum.

ITEST Project Overview:

- A week long summer training workshop for teachers (30 per year) to learn the fundamentals of gene annotation using the GENI-ACT system (along with 3 additional subsequent refresher training days).
- 3 fall semester activities to build interest among student participants and to recruit students to participate in the spring semester genome annotation exercises.
- Spring semester teacher guided genome annotation projects for 5 or more students per teacher (150 total per year). A control group of students will be included for comparison.
- A capstone symposium for student and teacher participants to present the results of their genome annotations.

Student Participant Selection



GENI-ACT Overview

[] geni-act

GENI-ACT allows collaborative genome annotation. Researchers or students can collectively suggest changes to an existing genome with supporting evidence. Changes can be ported back to genbank by exporting to a sequin file format.

GENI-ACT also has ported the education components from IMG-ACT. Teachers can assign students work to be completed in a lab notebook that is integrated with the classroom.

2775 genomes available

Sign Up Login **Browse Genomes**

Signing Up For GENI-ACT

[] 9	geni-act
geni-act :: re	egister
1	an account below. If you are a student in a class, make sure to enter the class-id. First Name: Last Name: Email: Password: Password Confirm: Class Token:
	ith a valid class-id are created immediately. Otherwise the account is made as edged user until validated by an administrator.

If you want to experiment With GENI-ACT you can use this token: GEM 2016

Email me at stvkoury@buffalo.edu after signing up and I will make some gene assignments for you.

Request Course Instructor status from the system adminstrator, Dr. Brad Goodner at: GoodnerBW@hiram.edu

Workshops can be arranged for any interested faculty.



Profile

Courses

Name	School	Term
MT447/547 - Introduction to Microbial Genome Annotation	University at Buffalo	Spring 2014
MGAN Workshop Genes	University at Buffalo	Spring 2016
Biotechnology Techniques	Westfield Central School	Spring 2016
UB GEM - Try out GENI-ACT	University at Buffalo	Spring 2016

Instructor Course Management

geni-act :: courses :: MT 447/547 Spring 2016

MT 447/547 Spring 2016 **Change Course Properties** Info Period: Spring 2016 Write Access Date: Not specified. Name MT 447/547 Spring 2016 Course End Date: Not specified. Token MT447/547_Spring_2016 **Change Course Properties** Class Management [+] Students [+] Sections [+] Teaching Assistants [+] Student Teams

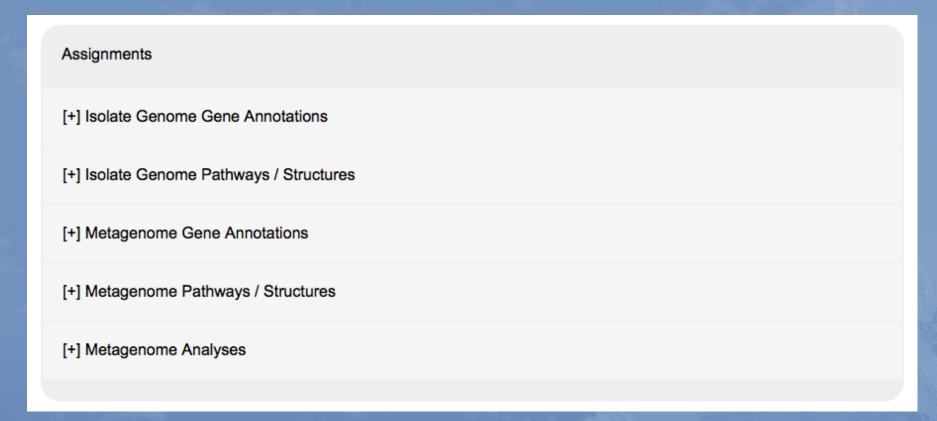
GENI-ACT Modules

Enabled Modules				
Basic Information	ON	Turn OFF		
Sequence-based Similarity Data	ON	Turn OFF		
Cellular Localization Data	ON	Turn OFF		
Alternative Open Reading Frame	ON	Turn OFF		
Structure-based Evidence	ON	Turn OFF		
Enzymatic Function	ON	Turn OFF		
Duplication and Degradation	ON	Turn OFF		
Horizontal Gene Transfer	ON	Turn OFF		
RNA	ON	Turn OFF		
Proposed Annotation	ON	Turn OFF		

Available Modules	
Basic Information	 DNA Coordinates: Use a <u>GENE Page</u> to enter coordinates Nucleotide Sequence / Length: Use a <u>GENE Page</u> to find the Nucleotide Sequence and Length Protein Sequence / Length: Use a <u>GENE Page</u> to find the Protein Sequence and Length
Sequence-based Similarity Data	 BLAST: Find the top hits in BLAST using NCBI BLAST CCD: Find COG Results from NCBI BLAST T-Coffee: Run a multiple sequence alignment using T-Coffee WebLogo: Find the sequence logo and analyze it
Cellular Localization Data	 Gram Stain: Research Pubmed to find the gram stain of the organism TMHMM: Plot the transmembrane topology SignalP: Plot the signal peptide graph LipoP: Predict lipoproteins and signal peptides PSORT-B: Predict protien localization Phobius: Plot the phobius posterior probabilities
Alternative Open Reading Frame	DNA Coordiantes: Change the DNA Coordinates if the ORF has been incorrectly determined

Structure-based Evidence	TIGRFAM: Search TIGRFAM Hidden Markov Model database for hits Pfam: Find Pfam families within a sequence PDB: Align sequences using Protein Data Bank
Enzymatic Function	KEGG: Find the KEGG Pathway and analyze the pathway map MetaCyc: Find the Metabolic Pathway and analyze EC Number: Use ExPASy ENZYME to locate the EC number and name
Duplication and Degradation	Paralogs: Use NCBI BLAST to find paralogs Pseudogene: Research to find if the gene is a pseudogene
Horizontal Gene Transfer	 Phylogenetic Tree: Use Phylogeny.fr to plot the phylogenetic tree from sequences Gene Context: Find the Ortholog Neighborhood Region using IMG Chromosome Viewer GC Heat Map: Analyze the GC heat map to find the characteristic and average GC%
RNA	Rfam: Find the Rfam number, score and pairwise alignment
Proposed Annotation	Note: Propose a new annotation to be placed in the note section of GENI-ACT and Genbank GBK

Types of Assignments in GENI-ACT



Ideas for implementation strategies in using GENI-ACT can be found at:

https://sites.google.com/a/augustana.edu/mgan workshop ccuri2014/home/implementation-strategies



Organism: Kytococcus sedentarius DSM 20547 CP001686

Locus: Ksed 00010

Instructions

The locus tag for the gene under investigation in this notebook

Basic online module

instructions

Basic Information

Sequence-based Similarity Data

Cellular Localization Data

Alternative Open Reading Frame

Structure-based Evidence

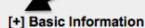
Enzymatic Function

Duplication and Degradation

Horizontal Gene Transfer

RNA

Clicking on a notebook section opens it for editing



- [+] Sequence-based Similarity Data
- [+] Cellular Localization Data
- [+] Alternative Open Reading Frame

The GENI-ACT Student Notebook

Locus Tag Links to Gene Page for the gene under investigation

Detailed Module Instructions Have Been Developed on our Project Websites.

Making Entries in the Lab Notebook

Basic Information	
dule Instructions	
ONA Coordinates	
o to the Gene Page	Clicking here will open the editor for data to be added to the notebook.
ONA coordinates 🗎 🕌	
DNA Sequence	
DNA Sequence go to the <u>Gene Page</u> Nucleotide sequence (FAS	TA format; see module Quick Links for instructions)
go to the Gene Page	STA format; see module Quick Links for instructions)
go to the <u>Gene Page</u> Nucleotide sequence (FAS	STA format; see module Quick Links for instructions)
go to the Gene Page	TA format; see module Quick Links for instructions)



DNA Sequence

go to the Gene Page

Nucleotide sequence (FASTA format; see module Quick Links for instructions)



Upload Image:

Browse... No file selected.

Upload

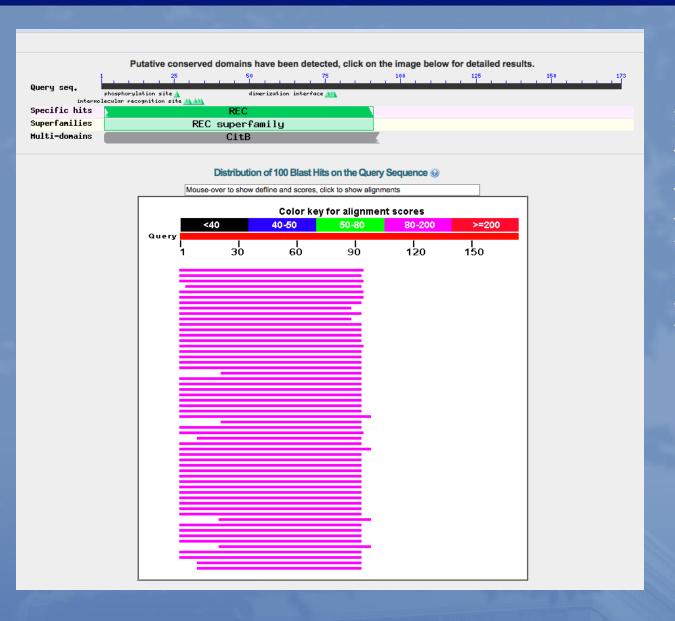
Overview of a filled in notebook and examples of student posters:

http://www.geni-act.com/lab_notebook/public/8ed2a45227a744df/

http://ubwp.buffalo.edu/wnygirp/student-research/

One example of wet lab experiment resulting from annotation results

- Illustrates how undergraduate research or MS level project or thesis research studies can be generated from annotation of a genome.
- http://geni-act.org/gene/locus/CP001686/Ksed_02850/



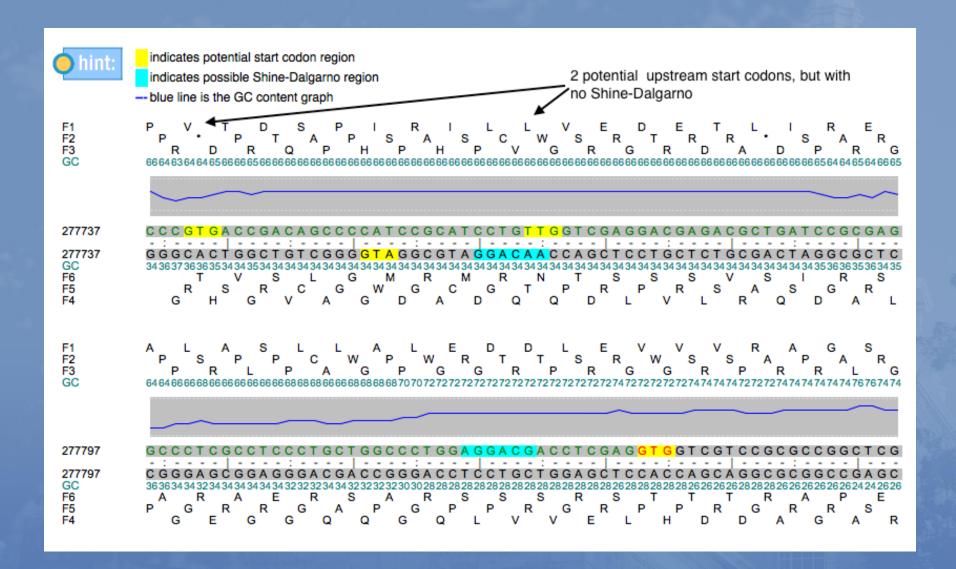
Ksed_02850: Example of an interesting finding leading to proposed wet lab experiments; annotated as a pseudogene.

T-Coffee multiple sequence alignment: Incorrect start codon?

Multiple sequence alignment

```
CLUSTAL W (1.83) multiple sequence alignment
Ksed 02850-aa
gi | 377537422 | dbj | GAB48067.1 |
gi 496022179 ref WP 008746686.1
                                     -----EPAP-AQPVTSRVRVLLADDEHLIRGALAALLALE
gi|502488441|ref|WP 012803990.1
                                 MSPARDSRTSPEPGTSPAOASEPADAALRVILVDDENLIRSALATMLSLE
gi|502715736|ref|WP 012950794.1
                                                         ----IRVLLADDENLIRSALAALLALE
  |654875439|ref|WP 028327408.1
                                       ----IRLLADDENMFRDALATLLGLO
  |738384255|ref|WP 036336310.1
                                         ----IRVLLADDENLVRSAFAALLGLE
gi | 739287555 | ref | WP 037150275.1
                                           ----IPVLLADDENLVRTAMAAMLDLE
gi | 750276589 | ref | WP 040562936.1
                                                   -----PIRILIADDETLFRDALATLLDLR
gi | 772758607 | ref | WP 045317119.1
gi 820816812 ref WP 046769457.1
                                          -----IRVLLADDENLIRSALRALLSLE
Ksed 02850-aa
                                  ----MVVRAGSVAEAKAVLR----STEVDVALLDLQLPDGDGIDLAVHL
gi 377537422 dbj GAB48067.1
                                 DDLEVVAOAASGPEAMALAR----AHEFDVALLDLOMPGADGIAVAETI
gi|496022179|ref|WP 008746686.1
                                 EDLEVVAEAASGAEALAMAR----ARTPDVAVLDLOMPDGDGVSVATAL
gi | 502488441 | ref | WP 012803990.1
                                 ADLDVRGEAATVADGLCLAE----AVQPDVAVLDLQLPDGDGLELAARI
gi|502715736|ref|WP 012950794.1
                                 EDLEVVAQAASGAEALAMAR----AHIPDVAVLDLOMPDRDGISVAGEL
  |654875439|ref|WP 028327408.1
                                 EDLLVVAQAASGPEAIAMAC----SHEVDVAVLDLQMPGADGIAVAESL
  |738384255|ref|WP 036336310.1
                                 DDLEVVAQASSGAEALAMAR----AHAPDVAVLDLQMPDRDGISVAGEL
gi|739287555|ref|WP 037150275.1
                                 TDIDVIADVESGEDLIAIWRRRVDRGESAAVAVIDLOMPGIDGIDTAIEL
gi | 750276589 | ref | WP 040562936.1
                                 DDLHVVAOAASGPEAVAMAR----AHRPDVALLDLOMPGODGIETAIAL
gi | 772758607 | ref | WP 045317119.1
                                 DDLEVVAOAATGAEALATAR-----VVRPDVAVIDLOMPDLDGVSVAOAL
gi 820816812 ref WP 046769457.1
                                 DGIEVVAEAATADEALAMAR----SFSPDVAVLDLQLPDRDGIALAAEL
```

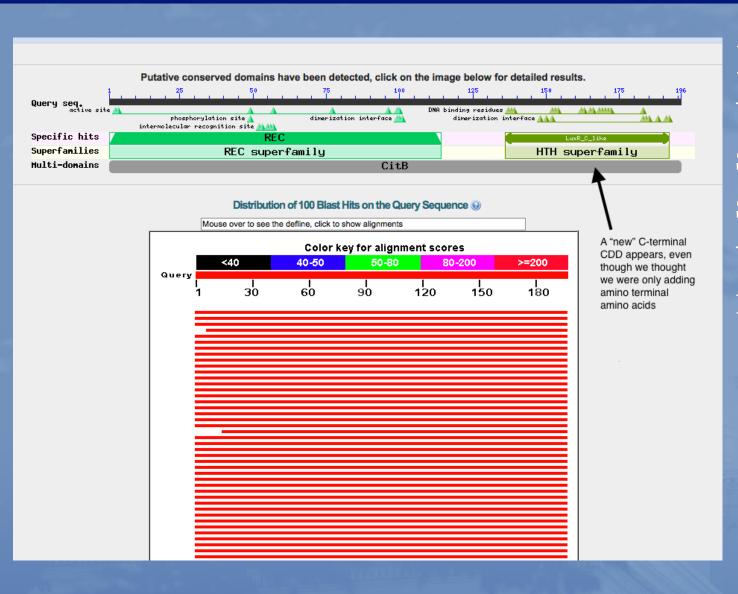
Alternate Open Reading Frame Viewer – IMG/EDU



Obtained the translation from the upstream in frame start codon from the text output of the AORF viewer

>645946474_1_ORF1 Translation of 645946474 in frame 1, ORF 1, threshold 80,206aa

LVEDETLIREALASLLALEDDLEVVVRAGSVAEAKAVLRSTEVDVALL DLQLPDGDGIDLAVHLGEVQPQAASLIITSHGRPGYLKRALESGVRG FLPKTVGRRALGEAVRTLAEGGRYVDQELAADALAAGASPLSAREA DVLELSADAAPVEEIAQRAHLSAGTVRNYLSAAVAKTGTSNRHEAAR **VARSKGWI**



Blast of first upstream start codon sequence: unexpected result!

This led to an align two sequences experiment in blast: frame shift "mutation" detected

■Download ∨ Graphics

645946474_1_ORF1 Translation of 645946474 in frame 1, ORF 1, threshold 80, 206aa Sequence ID: Icl|Query 240543 Length: 196 Number of Matches: 1

Dango	4 .	24	400	102	Graphics
range	4.	4	LO	103	Graphics

_										
Score		Expect	Method		I	dentities	Posi	tives	Gap	s
187 bits	s(476)	1e-64	Compositiona	al matrix ac	ijust. 1	08/168(64	%) 114/	/168(67%)	12/	168(7%)
Query	1		SVAEAKAVLRST SVAEAKAVLRST							60
Sbjct	24		SVAEAKAVLRST							83
Query	61		SGVRGFLPKTV							120
Sbjct	84		SGVRGFLPKTV							135
Query	121	WSSRPTE S+R	PRRWRRSPSGPT	CRPAR	CATTCR		TGTRPR	164		
Sbjct	136		OVLELSADAAPV					183		

■Download ∨ Graphics

Alt_Ksed_02850 DNA sequence consisting of 522 bases

Sequence ID: Icl|Query_142477 Length: 522 Number of Matches: 1

Range 1	l: 1 to	522 Graphics		▼ r	Next Match 🛕	Previous M
Score		Expect	Identities	Gaps	Strand	
957 bit	s(518) 0.0	521/522(99%)	1/522(0%)	Plus/Plu	S
Query	1	GTGGTCGTCCGCGCC	GGCTCGGTGGCCGAGGCG	AAGGCCGTGCTGCGCTCC.	ACCGAGGTG	60
Sbjct	1	GTGGTCGTCCGCGCC	GGCTCGGTGGCCGAGGCG	AAGGCCGTGCTGCGCTCC	ACCGAGGTG	60
Query	61	GACGTCGCCCTGCTG	GACCTGCAGCTGCCGGAC	GGCGACGGCATCGACCTC	GCGGTGCAC	120
Sbjct	61	GACGTCGCCCTGCTG	GACCTGCAGCTGCCGGAC	GGCGACGGCATCGACCTC	GCGGTGCAC	120
Query	121	CTGGGTGAGGTGCAG	CCGCAGGCGGCGAGCCTC	ATCATCACCAGCCACGGG	CGCCCCGGG	180
Sbjct	121	CTGGGTGAGGTGCAG	CCGCAGGCGGCGAGCCTC	ATCATCACCAGCCACGGG	ceccceee	180
Query	181	TACCTGAAGCGGGCG	CTGGAGTCCGGCGTCCGG	GGGTTCCTGCCCAAGACG	GTGGGACGC	240
Sbjct	181	TACCTGAAGCGGGC	ctecaetccecetcee	GGGTTCCTGCCCAAGACG	GTGGGACGC	240
Query	241	CGGCCCTCGCCGAG	GCGGTGCGGACCCTCGCG	GA-GGGGGCCGGTACGTG	GACCAGGAG	299
Sbjct	241	CGGGCGCTCGGCGAG	GCGGTGCGGACCCTCGCG	GAGGGGGCCGGTACGTG	GACCAGGAG	300
Query	300	CTGGCGGCCGACGCC	CTGGCCGCTGGCGCCTCC	CCGCTCAGCGCCCGGGAG	GCCGACGTG	359
Sbjct	301	CTGGCGGCCGACGCC	creecectecectcc	CCGCTCAGCGCCCGGGAG	GCCGACGTG	360
Query	360	CTGGAGCTCTCGGCC	GACGCCGCGCCGTGGAG	GAGATCGCCCAGCGGGCC	CACCTGTCG	419
Sbjct	361	CTGGAGCTCTCGGCC	GACGCCGCGCCGGTGGAG	GAGATCGCCCAGCGGCC	CACCTGTCG	420
Query	420	GCCGGCACGGTGCGC	AACTACCTGTCGGCGGCC	GTGGCGAAGACCGGCACC	TCCAACCGG	479
Sbjct	421	GCCGGCACGGTGCGC	AACTACCTGTCGGCGGCC	GTGGCGAAGACCGGCACC	TCCAACCGG	480
Query	480	CACGAGGCCGCGCG	GTCGCCCGGTCCAAGGGC	TGGATCTGA 521		
Sbjct	481	CACGAGGCCGCGCG	GTCGCCCGGTCCAAGGGC	TGGATCTGA 522		

Wet Lab Experiments Initiated –

- Kytococcus sedentarius DSM20547 obtained from ATCC
- Genomic DNA isolated
- PCR primers designed to amplify predicted coding region
- Will clone into an expression vector
- mRNA will be isolated to determine if expressed under normal growth conditions
- Further bioinformatic / wet lab studies to help narrow down potential function for testing in rescuing mutants

Collaborators:

Shannon Carlin-Menter, PhD Danise Wilson, MPH Rama Dey-Rao, PhD Norma Nowak, PhD Patricia Masso-Welch, PhD Karen Liermo, MS Lon Knappenberger, MS Oak Ridge Affiliated Universities - evaluation team

Brad Goodner, PhD - Hiram College Microbial Genome Annotators Network (MGAN)

Links of Interest:

GENI-ACT: www.geni-act.org

Video Tutorials: http://geni-

act.blogspot.com/p/welcome-to-geni-act-teaching-

resources.html

WNY Genetics in Research Partnership: http://ubwp.buffalo.edu/wnygirp/

WNY Genetics in Research and Health Care Partnership: http://ubwp.buffalo.edu/wnygirahcp/

GENI-ACT Manuals can be found at either of the above websites under Educational Resources