Annotation of the *Pseudomonas aeruginosa* Genome LES431 Locus Tags T223_00150 to T223_00180

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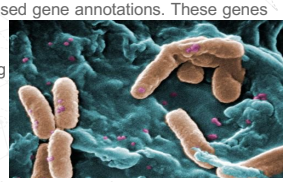
Abstract

A group of 3 neighboring genes from the microorganism *Pseudomonas aeruginosa* were annotated using the collaborative genome annotation website GENI-ACT. The Genbank proposed gene product names did not differ significantly from the proposed gene annotations. These genes were of research interest due to their medical significance. They are being investigated as possible drug targets. The enzymes produced by the studied genes can be targeted for medical treatment of infections caused by *Pseudomonas aeruginosa.*



Introduction

Pseudomonas aeruginosa is both an aerobic and anaerobic, motile, encapsulated, and non-endospore forming gram negative coccobacillus bacterium, found predominantly in complex groupings called “mucoids” that secrete adhesive polysaccharides, forming a sticky biofilm. This organism is classified as a gammaproteobacteria, as it is a gram-negative pathogen belonging to the group Pseudomonadaceae. It was first discovered by French chemist Carle Gessard in 1882 through an experiment that identified the microbe by its water soluble pigments that turned blue-green under exposure to ultra-violet light.

Pseudomonas aeruginosa is a microorganism of interest for several reasons. This bacterium can thrive in most environments, especially on moist surfaces, which allows it to effectively colonize in the lungs, urinary tract, and kidneys. *Pseudomonas aeruginosa* has been linked as an agent of a number of opportunistic infections including cystic fibrosis, postoperative infections in radial keratotomy patients, and is associated with osteomyelitis involving puncture wounds of the foot for which diabetic patients are at a higher risk. With low phosphate levels, this bacteria has been known to activate from benign symbiont to express lethal toxins inside the intestinal tract which either damages or kills the host. With a large resistance island of over 50 resistance genes within the genome, this bacteria has a low antibiotic susceptibility caused by multidrug efflux pumps and chromosomally encoded antibiotic resistance genes. Due to the lack of effective treatment, clinical trials for next-generation antibiotics are currently underway.

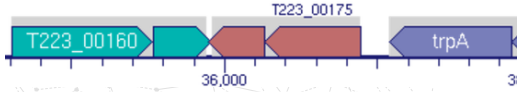


Figure 1. The locus tags and relative position of the genes under investigation in this research include T223_00160 (Erin Congdon), T223_00175 (Kathryn Sirianni), and T223_00180 (Zachary Greenfield).

Methods

Modules of the GENI-ACT (http://www.geni-act.org/) were used to complete Pseudomonas aeruginosa genome annotation. The modules are described below:

Modules	Activities	Questions Investigated
Module 1- Basic Information Module	DNA Coordinates and Sequence, Protein Sequence	What is the sequence of my gene and protein? Where is it located in the genome?
Module 2- Sequence-Based Similarity Data	Blast, CDD, T-Coffee, WebLogo	Is my sequence similar to other sequences in Genbank?
Module 3- Cellular Localization Data	Gram Stain, TMHMM, SignalP, PSORT, Phobius	Is my protein in the cytoplasm, secreted or embedded in the membrane?
Module 4- Alternative Open Reading Frame	IMG Sequence Viewer For Alternate ORF Search	Has the amino acid sequence of my protein been called correctly by the computer?
Module 5- Structure-Based Evidence	TIGRfam, Pfam, PDB	Are there functional domains in my protein?
Module 6- Enzymatic Function	KEGG, MetaCyc, E.C. Number,	In what process does my protein take part?
Module 7- Gene Duplication/ Gene Degradation	Paralog, Pseudogene	Are there other forms of my gene in the bacterium? Is my gene functional?
Module 8- Evidence for Horizontal Gene Transfer	Phylogenetic Tree,	Has my gene co-evolved with other genes in the genome?
Module 9- RNA	RFAM	Does my gene encode a functional RNA?

Results

T223_00160:
The initial proposed product of this gene by GENI-ACT was a transcriptional regulator. This gene product proposal was supported by the top BLAST hits for the amino acid sequence and the presence of well-curated protein functional domains. As such, the proposed annotation is a transcriptional regulator that was determined at the completion of the second module.

T223_00175:
The initial proposed product of this gene by GENI-ACT was a virulence factors putative positive transcription regulator BvgA. This gene product proposal was supported by the top BLAST hits for the amino acid sequence, the presence of well-curated functional domains and the cellular location of the protein. As such, the proposed annotation is a virulence factors putative positive transcription regulator BvgA that was determined by the completion of the fourth module.

T223_00180:
The initial proposed product of these genes by GENI-ACT was a tryptophan synthase, subunit alpha. This gene product proposal was supported by the top BLAST hits for the amino acid sequences, the presence of well-curated functional domains, the cellular location of the proteins, and the enzymatic function of the gene product. As such, the proposed annotation is tryptophan synthase, subunit alpha.

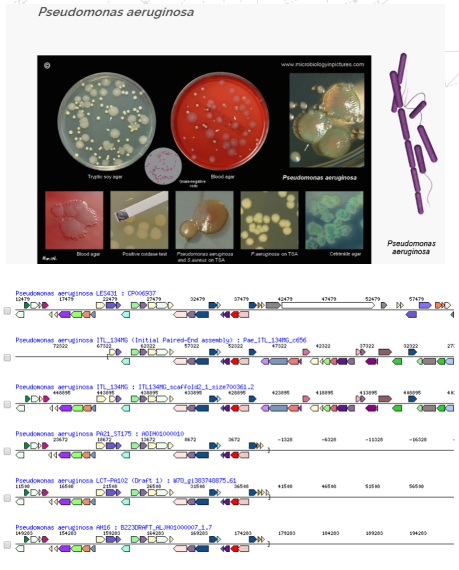


Figure 2 –Pseudomona aeruginosa LES431) gene neighborhood

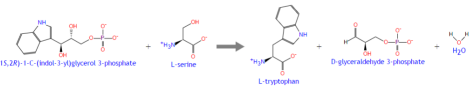


Figure 3 – T223_00180 reaction mechanism.

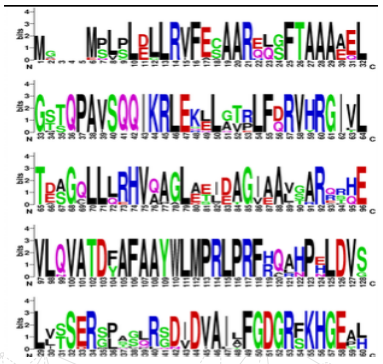
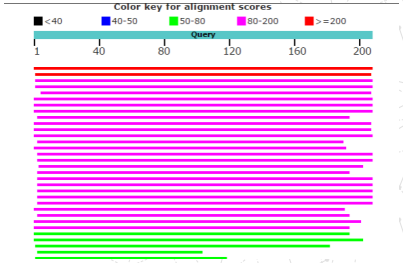


Figure 4 – Weblogo T223_00160.



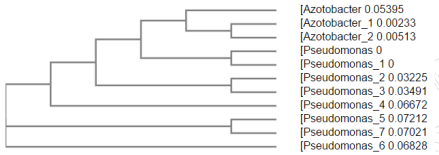
T223_00175 blast alignment

Phylogenetic Tree

This is a Neighbour-joining tree without distance corrections.

Download Phylogenetic Tree Data

Branch length: Cladogram Real



Conclusion

The GENI-ACT proposed gene product did not differ significantly from the proposed gene annotation for each of the genes in the group and as such, the genes appear to be correctly annotated by the computer database.

Gene Locus	Geni-Act Gene Products	Proposed Annotation
00160	Transcriptional Regulator	Transcriptional Regulator
00175	Virulence Factors Putative Positive Transcription Regulator BvgA	Virulence Factors Putative Positive Transcription Regulation BvgA
00180	Tryptophan Synthase, Subunit Alpha	Tryptophan Synthase, Subunit Alpha

References

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http://eol.org
Dunn MF, Niks D, Ngo H, Barends TR, Schlichting I (June 2008) “Tryptophan synthase: the working of a channeling nanomachine”. *Trends in Biochemical Scuences. 33*

Acknowledgments

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