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Annotation of the *Kytococcus Sedentarius* Genome from Locus Tags Ksed_08460 to Ksed_08490

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Abstract

A group of four consecutive genes from the microorganism *Kytococcus sedentarius* (Ksed_08460 - Ksed_08490) were annotated using the collaborative genome annotation website GENI-ACT. The Genbank proposed gene product name for each gene was assessed in terms of the general genomic information, amino acid sequence-based similarity data, structure-based evidence from the amino acid sequence, and cellular localization data. The Genbank proposed gene product name 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.

Introduction

Kytococcus sedentarius is an aerobic, non-motile, non-encapsulated, and non-endospore forming gram-positive coccoid bacterium, found predominantly in tetrad formation. According to Sims et al. (2009), *Kytococcus sedentarius* is a microorganism of interest for several reasons. This bacterium is a natural source of the antibiotics monensin A and monensin B (Sims et al., 2009). *Kytococcus sedentarius* has been shown to be the cause of a number of opportunistic infections including valve endocarditis, hemorrhagic pneumonia, and pitted keratolysis (Sims et al., 2009). Finally, the phylogeny of this microorganism is a source of interest, as it is a member of the family *Dermacoccaceae* within the actinobacterial suborder *Micrococcineae*, which has yet to have been thoroughly studied utilizing bioinformatics (Sims et al., 2009).

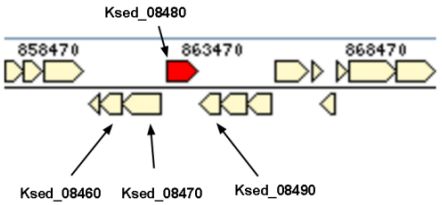

Figure I - Gene neighborhood of the four *Kytococcus sedentarius* genes annotated.

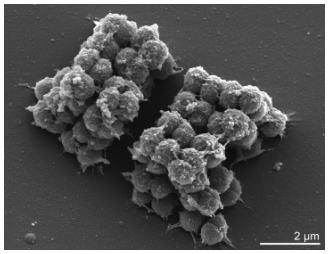

Figure II - Scanning electron Micrograph of *Kytococcus sedentarius*

Methods and Materials

Modules of the GENI-ACT (http://www.geni-act.org/) were used to complete *Kytococcus sedentarius* 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- Structure-Based Evidence	TIGRfam, Pfam, PDB	Are there functional domains in my protein?
Module 4- Cellular Localization Data	Gram Stain, TMHMM, SignalP, PSORT, Phobius	Is my protein in the cytoplasm, secreted or embedded in the membrane?

Results

Kytococcus sedentarius 08460:

The initial proposed product of this gene by GENI-ACT was a branched-chain amino acid permease. This gene product proposal was supported by the top BLAST hits for the amino acid sequence, the presence of well-curated protein functional domains within the amino acid sequence, the transmembrane topography of the amino acid sequence, and the cellular location of the amino acid sequence. As such, the proposed annotation is a branched-chain amino acid permease.

Kytococcus sedentarius 08470:

The initial proposed product of this gene by GENI-ACT was a cell envelope-related transcriptional attenuator. This gene product proposal was supported by the top BLAST hits for the amino acid sequence, the presence of well-curated functional domains within the amino acid sequence, the cellular location of the amino acid sequence, and the enzymatic function of the amino acid sequence. Attenuators are responsible for regulating the simultaneous transcription and translation of DNA. As such, the proposed annotation is a cell envelope-related transcriptional attenuator.

Kytococcus sedentarius 08480:

The initial proposed product of these genes by GENI-ACT was an succinoglycan biosynthesis protein. This gene product proposal was supported by the top BLAST hits for the amino acid sequences, this protein is needed for the making of succinoglycan, this protein is added to the second sugar (glucose) to make succinoglycan.
This protein is involved in exopolysaccharide biosynthesis (Making Polysaccharides that are that are exported and important in biofilms and in toxicity), which is part of glycan metabolism.

Kytococcus sedentarius 08490 :

The initial proposed product of this gene by GENI-ACT was a marker for gastrointestinal microbiota function. This gene product proposal was supported by the top BLAST hits for the amino acid sequence. This gene is involved with butanoate metabolism. As such, the proposed annotation is a marker for gastrointestinal microbiota function.

Figure III - Transcriptional Regulator Protein

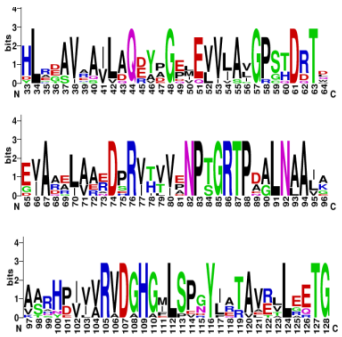

Figure IV – This is part of *Kytococcus sedentarius* 08480 WebLogo area that is showing regions of conserved amino acids (the single large letters), this also shows areas with non-conserved amino acids (the stacked small letters).

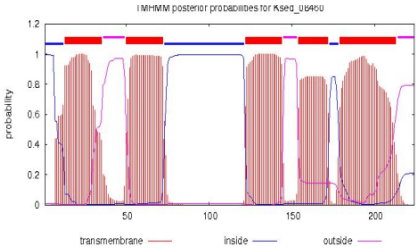

Figure V– This graph shows the TMHMM results for Ksed_08460. The red shows the portions of the protein embedded in the membrane. Five transmembrane domains are shown in the graph. The blue and pink lines indicate the portions of the protein that loop inside (blue) and outside (pink) of the membrane.

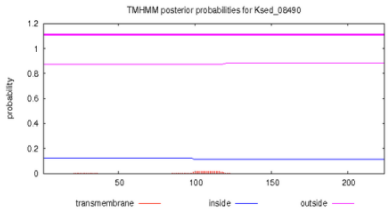

Figure VI - This graph shows the TMHMM results for Ksed_08490. This is an example of a graph with zero predicted transmembrane helices.

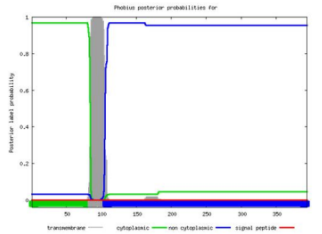

Figure VII - This is a Phobius graph that shows in *Kytococcus sedentarius* 08470 there is one transmembrane helix.

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 Product	Proposed Annotation
08460	Branched-chain amino acid permease.	Branched-chain amino acid permease.
08470	Cell envelope-related transcriptional attenuator	Cell envelope-related transcriptional attenuator
08480	Succinoglycan biosynthesis protein	Succinoglycan biosynthesis protein
08490	Gastrointestinal microbiota function	Gastrointestinal microbiota function

References

Sims et al. (2009). Complete genome sequence of Kytococcus sedentarius type strain (541T). *Standards in Genomic Sciences*, 12 - 20.

Acknowledgments

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