

Annotation of the Helicobacter pylori Genome from DNA Coordinates 45733 to 50762

Rebekah Boy, Samatha Freiberger, Lindsey Nudds, Sorcha Lester and Lawrence Hohl

East Rochester Junior/Senior High School and the Western New York Genetics in Research and Healthcare Partnership

We were given a segment of the Helicobacter pylori 83 genome to analyze whether the particular genes 0045. 0046. 0049. and 0050 were correctly annotated. We used the GENI-ACT modules to analyze these genes, and the data indicated that the proposed annotations for all four of these genes were correct. The proposed an notations are that gene 0045 codes for a competence protein, which is involved in trans-mem brane transport; gene 046 codes for 4-hv droxv be nzo ate octapr env ltra nsfera se, which is involved in ubiquinone-8 synthesis; gene 0049 codes for pho sph ati dyls eri ne d ecar box yla se, w hich is in volv ed in aminophospholipid synthesis; and gene 0050 codes for quinolinate synthase, which is involved in NAD synthesis.

Introduction

Helicobacter pylori 83 is a gram-negative bacteria, which burrows into the epithelial cells of the stomach (Amie va et al., 2008). It contains a hydrogenase, allowing it to gain energy through the oxidation of molecular hydrogen produced in the intestine (Olson et al., 2002). It also produces urease to neutralize the gastric acid around itself, which produces ammonia and damages the surrounding epithelial cells, inflaming the cell sand often cau sing gastric and duodenal ulcers. O ther chemicals it produces, such as protease and lipase, have a similar effect (Smoot, 1997). H. pylori has been shown to be the main direct cause of distal gastricadenocarcinoma and gastric lymphoma, but only 15% of the bacteria develop these diseases. Research into this bacterium and how it becomes cancerous may lead to a cure for these forms of gastric cancer (Atherton, 2006)



Figure I - This picture shows the helical shape of Helicobader pylori 83, and shows the flagella that help it to burrow into the membrane of the stomach



Figure II - This diagram shows the locus tags of the genes analyzed in this annotation relative to each other and the DNA coordinates

Methods and Materials

Modules of the GENI-ACT (http://www.geni-act.org/) were used to complete Heli cobacter pylori 83 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?

Helicobacter pylori 83 0045:

The function of this gene is that it codes for a protein of the competence family that assists with transport across the cell membrane

Helicobacter pylori 83 0046

The function of this gene is that it aids in the attachment of a polyprenyl group to para-hydroxybenzoate as a step in the biosynthesis of ubiquinone-8.

Helicobacter pylori 83 0049:

The function of this gene is that it plays a central role in the biosynthesis of aminophospholipids by converting phosphatidyl serine to phosphatidylethanolamine.

Helicobacter pylori 83 0050:

The function of this gene is that it codes for guinolinate synthase, an enzyme that aids in NAD synthesis.

The gene 0045 in Helicobacter pylori 83 codes for a com E protein of the competence family. This protein assists with transport across the cell membrane and provides the materials needed for the synthesis reactions of genes 0046, 0049, and 0050. This protein is present in the cytoplasmic membrane of the cell.



Figure III - This graph shows that the gene Helicobacter 83 0045 which codes for a protein of the competence family possesses 9 transmembrane helices, proving that it assists with transmembrane transportation



Figure IV - This diagram shows a portion of a WebLogo of gene 0050, showing the similarity between the gene sequence in Helicobacter pylori 83 and the same gene in other species. This segment shows that this gene is well conserved since many of the species have the same amino acids in the same places

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Sbjct	1	MIALSNALS	RVFGSVAGYEFP	SFIQKGINALYVKIF	KIDLSEFEPLENYK	SLNALFTRSL	60	
Query	61	KKERPFDKA	PNACIAPCDALI	TECAFLDNDSALQIK	GMPYKAHELVGEIN	PLSPSFFYVN	120	
Sbjct	61	KKERPFDKT	PNACIAPCDALI	TECAFLDNDSALQIK	GMPYKAHELVGEIN	PLSPSFFYVN	120	
Query	121	FYLSPKDYH	HYHAPCDLEILE	ARCFAGKLLPVNKPS	LYKNKNLFVGNERV	ALVAKDIQGN	180	
Sbjct	121	FYLSPKDYH	HYHAPCDLEILE	ARCFAGKLLPVNKPS	L+KNKNLFVGNERV	ALVAKDIQGN	180	
Query	181	RLYFVAVGA	LNVGKMRFNFDK	NIQTNAKAHLTQTYS	YNPPIKVKKGDNLG	NFEMGSTIVL	240	
Sbjct	181	RLYFVAVGA	LNVGKMRFNFDK	NIQTNAKAHLTQYS	VNPPIKVKKGDNLG	NFEMGSTIVL	240	
Query	241	FIQNTAFKD	LKEKSVKFGESI	GEFHAN 267				

F+ÖNTAFKDLKEKSVKFGESIGEFHAN Sbjct 241 FVÖNTAFKDLKEKSVKFGESIGEFHAN 267

Figure V - This diagram shows a BLAST search comparing the protein sequence of gene 0049 to a sequence known to convert phosphatidylserine to phosphatidylethanolamine. The sequences are

exceptionally similar to each other, which proves that they most likely are the same

Range	1: 1 to	336	GenPep	Graphic					Ψ N	ext Match	A Prev	ious Match
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Sbjct	1	MPT	DNDLKT	SIVELLED	LDALL	AHFYOK	EIVELA	HYTGDSLELA	KIAS	SDKNLIVE	rč 60	
Query	61	GVH	FNGESV	KALAFNKQ	/IMPK	LSCCSMA	MIDSHY	YDRSVHLLKE	YGVK	EFYPITYIN	IS 120	
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Query	181	NSK	EEIKNA	DVVCYNGF	SVHO	FKLEDI	FYRORY	PDILIAVHPE	CEPS	VVQNADFSG	S 240)
Sbjct	181	NSP	EEIKNA	DVVCYNGF	SVHQ	LFKLEDI	FYRORY	PDILIAVHPE	CEPS	VVSNADFSG	\$ 240)
Query	241	TSQ	LIEFVE	KLSPHQKV	AIGTE	SNLVNRL	AKRNEG	NTFILSSTLA	LCPT	MNETTLKDI	F 300)
Shiet	241	T50	TTEFUE	VYORGE, TH	TOTE	UNTWART	CARENHO	NTFILSSTIA	FCPT	MIETTLKDI	F 300	

- Query 301 EVLKAYKNHRAFNAIELKOEVARLAKLALIKOMELS EVLKAYKNHRA+NAIELKOEVARLAKLALIKOMELS Sbjet 301 EVLKAYKNHRAINAIELKOEVARLAKLAIIKOMELS

Figure VI - This diagram shows a BLAST search comparing the protein sequence of gene 0050 to a sequence known to code for the enzyme quinolinate synthase. The two sequences are extremely similar, proving that this gene very likely codes for quinolinate synthase



Figure VII - The loci under investigation in this diagramare Helicobader pylori 83 genes 0045,0046,0049, and 0050, as shown in Figure II. This diagram shows the annotated genes on the DNA of multiple organisms. It is evident that the gene neighborhoods for these loci are very well conserved.

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

Gene Locus	Geni-Act Gene Products	Gene Function
0045	Competence protein	Assists with transport across cell membrane
0046	4-hydroxybenzoate octaprenyltransferase	Aids in the attachment of polyprenyl group to para- hydroxybenzoate as a step in the biosynthesis of ubiquinone-8
0049	Phosphatidylserine decarboxylase	Plays central role in biosynthesis of aminophospholipids
0050	Quinolinate synthetase	Aids in NAD synthesis

References

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