# Proteome Comparison: A fine-grained tool for comparative genomics

In addition to the Protein Family Sorter that allows researchers to examine up to the protein families from up to 500 genomes at a time, PATRIC also provides a more detailed and fine-grained way to compare genomes. The Proteome Comparison tool is an enhancement of RAST's 'Sequence Based Comparison Tool' .[1] The Proteome Comparison tool allows users to readily identify insertions and deletions in up to nine target genomes compared with one reference genome, and examine the degree of homology among the genes. The tool colors each gene based on protein similarity using BLAST[2], and each gene is marked as being unique, a unidirectional best hit or a bidirectional best hit in comparison to the reference genome. The output also includes a whole-genome schematic colored by BLAST similarity and available as a Circos SVG image[3]. The resulting data table can also be downloaded for further analysis. The following workflow will demonstrate how to compare genomes using this tool

1. Click on the Services tab and then click on Proteome Comparison.



2. This will open up the Proteome Comparison landing page

Parameters 🚯	
FERENCE GENOME	
e.g. Mycobacterium tuberculosis H37Rv	-
D UP TO 8 GENOMES TO COMPARE (USE PLUS BUTTON TO ADD)	
e.g. M. tuberculosis CDC1551	- (
TIONAL EXTERNAL GENOME (PROTEIN FASTA FILE)	

3. In the text box below the words "REFERENCE GENOME" click on the funnel icon. This will open up a pop-up window that has two things to select (Public or My Genomes).

culosis H37Rv
JSE PLUS BUTTON TO ADD)
1551 🚽 🕁

#### 4. Deselect Public Genomes by clicking on the checkbox.

Parameters ()			
REFERENCE GENOME			
e.g. Mycobacteriu	m tuberculosis H37Rv	🛛 🚽 < Specify ge	enome name.
Include in Search	MPARE (USE PLUS BUTTON TO ADD)		
Public Genomes	s CDC1551	<b>~</b> 🗘	
My Genomes			
	J		

5. To see your private genomes, you will need to start typing the words you used to identify them when you annotated the genome. Here I start with "Acineto" and it will show me all of my private genomes that contain that text.

Parameters ()	
EFERENCE GENOME	
T Acineto	🔽 < Genome name.
Acinetobacter baumannii 1000160 [1310800.4]	
Acinetobacter baumannii [470.964]	0
Acinetobacter baumannii 1592897 [1310696.4]	v

6. Select the Acinetobacter baumannii 1592897 genome by clicking on it. Be sure that the entire name appears in the text box for REFERENCE GENOME.

Parameters ()	
REFERENCE GENOME	
▼ Acinetobacter baumannii 1592897	

7. Now we need to add the comparison genomes. In the text box below the words "ADD UP TO 8 GENOMES TO COMPARE" click on the funnel icon. This will open up a pop-up window that has two things to select (Public or My Genomes).

Parameters () REFERENCE GENOME	
▼ Acinetobacter baumannii 1592897	•
ADD UP TO 8 GENOMES TO COMPARE (USE PLUS BUTTON TO ADD)	Specify genome name.
Include in Search Public Genomes My Genomes	

#### 8. Deselect Public Genomes by clicking on the checkbox.

	· ·	
ADD UP TO 8 GENOMES TO C	OMPARE (USE PLUS BUTTON TO ADD)	
e.g. M. tuberculos	is CDC1551	l - 🗘
Include in Search Public Genomes My Genomes		

9. To see your private genomes, you will need to start typing the words you used to identify them when you annotated the genome. Here I start with "Acineto" and it will show me all of my private genomes that contain that text.

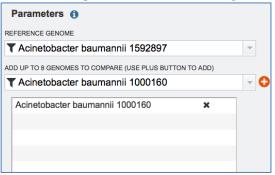
ADD UP TO 8 GENOMES TO COMPARE (USE PLUS BUTTON TO ADD)	
▼ Acinetobacter	🛛 🚽 🧲 The vi
Acinetobacter baumannii 1000160 [1310800.4]	
Acinetobacter baumannii [470.964]	
Acinetobacter baumannii 1592897 [1310696.4]	

10. Select the Acinetobacter baumannii 1000160 genome by clicking on it. Be sure that the entire name appears in the text box for ADD UP TO 8 GENOMES TO COMPARE.

ADD UP TO 8 GENOMES TO COMPARE (USE PLUS BUTTON TO ADD)			
▼ Acinetobacter baumannii 1000160	-	0	
	]		

11. You will need to click on the Plus Sign Icon that is at the end of the text box. This will add the genome to the box below. This box contains all the genomes that you

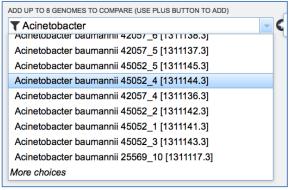
want to compare to the reference genome.



12. Now let's add some public genomes. You'll first have to click on the funnel icon, then deselect My Genomes and select Public Genomes.

ADD UP TO 8 GENOMES TO C	OMPARE (USE PLUS BUT	FTON TO ADD)	
Acinetobacter bau	umannii 1000160		- 0
Include in Search Public Genomes My Genomes	nnii 1000160	×	

13. If you start typing the word Acinetobacter, you will see that a lot of choices pop up.



14. To find a specific genome, you will probably need to type something unique to it, perhaps the strain name. Here I typed in "AYE" and then clicked on the line that had "Acinetobacter baumannii AYE VEB [509173.8]".

▼ AYE	-
Yersinia enterocolitica subsp. palearctica YE-P1 [13293	363.3]
Yersinia enterocolitica subsp. palearctica YE-149 [1329	365.3]
Yersinia enterocolitica subsp. palearctica YE-P4 [13293	364.3]
Yersinia enterocolitica subsp. palearctica YE-150 [1329	366.3]
Mycoplasma yeatsii 13926 [1188240.3]	
Neisseria shayeganii 871 [1032488.3]	
Metallosphaera yellowstonensis MK1 [671065.12]	
Acinetobacter baumannii AYE VEB [509173.8]	

#### 15. You will need to click the Plus Sign Icon to enter that genome into the comparison box below.

ADD UP TO 8 GENOMES TO COMPARE (USE PLUS BUTTON T	TO ADD)	
T Acinetobacter baumannii AYE		- 🗘
Acinetobacter baumannii AYE	×	
Acinetobacter baumannii 1000160	×	

# 16. Use the previous step to include the SDF genome ADD UP TO 8 GENOMES TO COMPARE (USE PLUS BUTTON TO ADD)

T Acinetobacter baumannii SD	-
Acinetobacter baumannii 2011SDAB2 [470.583]	
Acinetobacter baumannii 2011SDAB1 [470.582]	
Acinetobacter baumannii 2011SDAB3 [470.584]	
Acinetobacter baumannii SDF [509170.6]	

17. You should see one reference genome, and in this case three comparison genomes in the comparison genome box.

Proteome Comparison Protein sequence-based comparison using bi-	lirectional l	BLASTP.
Parameters ()		
REFERENCE GENOME		
▼ Acinetobacter baumannii 1592897		-
ADD UP TO 8 GENOMES TO COMPARE (USE PLUS BUTTON	TO ADD)	
T Acinetobacter baumannii SDF		- O
Acinetobacter baumannii SDF	×	
Acinetobacter baumannii AYE	×	
Acinetobacter baumannii 1000160	×	

18. If you had an annotation that was not part of PATRIC, you could upload it at this time using the OPTIONAL EXTERNAL GENOME (PROTEIN FASTA FILE) function by

clicking on the folder at the end of the text box. Directions will pop up that will show you how to upload the file. We aren't doing that in this exercise.



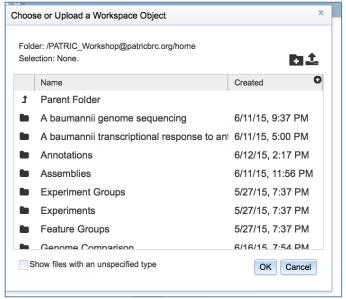
19. You will need to identify an output folder where the results will be placed.

OUTPUT FOLDER	
	-
OUTPUT NAME	
Output Name	1

20. At the end of the text box under OUTPUT FOLDER, click on the Folder icon



21. This will open a pop-up window that shows the folders in your workspace.



22. For this exercise, we want to name a new folder. In the upper right hand corner of the window you will see a Folder icon with a plus mark in it. Click on that.



23. Below the folder icon, a Text icon and a text box will appear

Choo	se or Upload a Workspace Object		х
	der: /PATRIC_Workshop@patricbrc.org/home ection: None.		<b>t</b> 1
	Name	Created	0
	Untitled Folder		

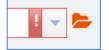
24. I entered the words "Proteome Comparison" in my text box. Once you name your folder, click the OK button at the bottom of the window. The pop-up window will disappear after you do this.

C	Choos	e or Upload a Workspace Object	х
		er: /PATRIC_Workshop@patricbrc.org/home ction: None.	n1
		Name	Created O
	Ľ	Proteome Comparison	
	t	Parent Folder	
		A baumannii genome sequencing	6/11/15, 9:37 PM
		A baumannii transcriptional response to ant	6/11/15, 5:00 PM
		Annotations	6/12/15, 2:17 PM
		Assemblies	6/11/15, 11:56 PM
		Experiment Groups	5/27/15, 7:37 PM
		Experiments	5/27/15, 7:37 PM
l		Footuro Groups	5/27/15 7.27 DM
	S	now files with an unspecified type	OK Cancel

25. Now we need to choose that folder as your OUTPUT FOLDER for the results of your comparison.

OUTPUT FOLDER	
	- 5-
OUTPUT NAME	
Output Name	1

26. Click on the folder icon at the end of the text box.



27. This will open up a pop-up window that has all of your folders. I have a lot of folders, and I selected the one I named "Proteome Comparison."

/home/ Experiments	
/home/ Genome Groups	J
/home/ <b>Experiment Groups</b>	
/home/ <b>Feature Groups</b>	
/home/ A baumannii transcriptional response to antibiotic stres	S
/home//Acinetobacter baumannii 1000160/ AMI_75_BR2	
/home//Acinetobacter baumannii 1000160/ CIP_25_BR2	
/home//Acinetobacter baumannii 1592897/ MHB_BR2	
/home/ Proteome Comparison	
/home/A baumannii transcriptional response to antibiotic stress Acinetobacter baumannii 34654	/
/home//Acinetobacter baumannii 34654/ CIP_75_BR2	
/home//Acinetobacter baumannii 34654/ MHB_BR2	
OUTPUT NAME	
Output Name	

28. Clicking on it enters the name into the text box under OUTPUT FOLDER. Now you will need to choose what to name your comparison

OUTPUT FOLDER	
Proteome Comparison	-
OUTPUT NAME	
Output Name	1

29. In the text box under OUTPUT NAME, enter text that you will recognize as specific to this comparison. I used "Ab 1592897 as reference". Then click the Submit button.

OUTPUT FOLDER	
Proteome Comparison	-
OUTPUT NAME	
Ab 1592897 as reference	
Reset Submit	

30. At the lower right hand corner of the page, you will see a box called Jobs with number following it. That tells you the number of submitted, in progress, queued and suspended jobs. Double click on the word "Jobs".



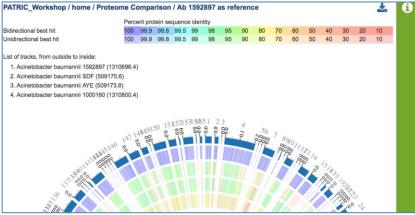
31. That will take you to the landing page for your PATRIC jobs. The green circle shows you that your comparison is in progress

	· ·				
Status	Submit	Арр	Output Name	Start	Completed O
<ul> <li>in-progress</li> </ul>	7/10/15, 8:03 AM	GenomeComparison	Ab 1592897 as reference	7/10/15, 8:03 AM	
<ul> <li>completed</li> </ul>	6/29/15, 1:57 AM	RNASeq	ab_34654_test	6/29/15, 1:57 AM	6/29/15, 3:34 AM

32. Once the job is complete, the circle will turn blue. Double click on that blue circle (Note: You can also access the comparison by clicking on WORKSPACE: HOME at the upper right hand corner of the page and finding the right folder, but this is quicker).

Status	Submit	Арр	Output Name	Start	Completed
completed	7/10/15, 8:03 AM	GenomeComparison	Ab 1592897 as reference	7/10/15, 8:03 AM	7/10/15, 8:07 AM

33. This will open up the results of your proteome comparison job. You will see a page that shows you the name, a list of the genomes compared, a figure that shows you're the percent sequence identity for both the uni- and bi-directional best BLAST hits, and a Circos diagram that shows the reference and comparison genomes.



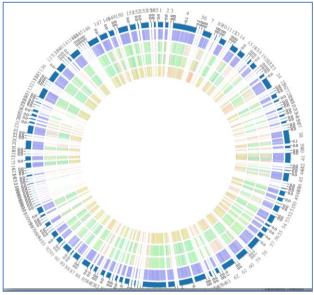
34. To further examine the results, you will need to download them. Click on the Download icon at the upper right corner, next to the green "i".



35. This will show you the download choices. Click on the SVG image.



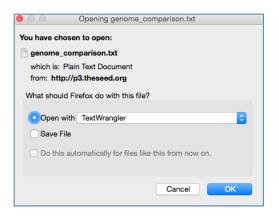
36. This will open up the visualization of the comparison results in Circos. The 1592857 genome is in a lot of contigs, and that is why you are seeing the fractured image. When you use a complete genome, the chromosome will be united into a single, uniform image.



37. To see the data behind the image you will need to download the table. Return to the download icon and click on Genome Comparison Table



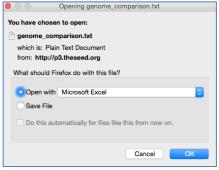
38. A pop-up window appears and I am given the choice to open it with TextWrangler, but I want to see it in excel so I click on the arrow following TextWrangler.



## 39. I choose Microsoft Excel.

	Choose Helper App	lication			
C > SS					
Favorites	Name	Date Modified	<ul> <li>✓ Size</li> </ul>	Kind	
All My Files	Multilities	Yesterday, 11:36 AM		Folder	
iCloud Drive	Microsoft Office 2011	Jul 7, 2015, 4:55 PM		Folder	
	Microsoft Document Connection	Jul 7, 2015, 4:58 PM	5.3 MB	Application	
Applications	🗙 Microsoft Excel	Jul 7, 2015, 4:58 PM	47.1 MB	Application	
Desktop	Microsoft Outlook	Jul 7, 2015, 4:58 PM	48.6 MB	Application	
	Microsoft PowerPoint	Jul 7, 2015, 4:58 PM	30.1 MB	Application	
Documents	Microsoft Word	Jul 7, 2015, 4:58 PM	56.7 MB	Application	

40. Now click the OK button at the bottom of the pop-up window.



41. This will open up an excel file with all the results, and there are a lot of them. Don't be discouraged by all the numbers and columns, this is valuable information and once you explore it you will see how useful it is.. We will go over these in class, but you note that it begins with your reference genome , and then shows you the comparison genomes across Row 1. The second row has the pertinent column heading below those genomes. Click on each one of those column headings so that you can understand the data you will see below them for each individual gene. Using the Freeze Pane functionality in excel works very nicely here.

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		genome ref genome ref genome i	ef_genome_ref_genome_ref	genome re	ef_genome_ref_ge	nome comp	genor comp_genor com	genor co	mp_genor o	omp_genor cor	np genor d	omp_gen	or comp_genor co	mp_genor.co	mp_genor com	p genor comp genor	comp_genor co	mp_genor comp_ge
1310696.4.ct	1	189 fig 1310696.4.peg.1	Transcription	272	841 +	bi	NC_010400	1777	315 fi	g 509170.6 AB	SDF1859 a	lkR	Transcription	0.968	0.587 bi	NC_010410	1973	315 fig 5091
1310696.4.cr	2	621 fig 1310696.4.peg.2	Peptidyl-prol	881	2746 -	bi	NC_010400	1776	621 fi	g 509170.6 AB	5DF1858 p	piD	Peptidyl-prol	0.963	0.998 bi	NC_010410	1974	621 fig 5091
1310696.4.ct	3	90 fig 1310696.4.peg.3	DNA-binding	2878	3150 -	bi	NC_010400	1775	90 fi	g 509170.6 AB	5DF1857 H	ирВ	DNA-binding	1	0.989 bi	NC_010410	1975	90 fig 5091
1310696.4.ct	4	151 fig 1310696.4.peg.4	polyhydroxya	3320	3775 -	bi	NC_010400	1774	151 fi	g 509170.6 AB	SDF1856		polyhydroxya	0.98	0.993 bi	NC_010410	1976	151 fig 5091
1310696.4.ct	5	215 fig 1310696.4.peg.5	hypothetical	3986	4633 +	bi	NC_010400	1773	215 fi	g 509170.6 AB	SDF1855		hypothetical	0.963	0.995 bi	NC_010410	1977	215 fig 5093
1310696.4.cr	6	157 fig 1310696.4.peg.6	Iron-sulfur cl	4742	5215 +	bi	NC_010400	1772	157 fi	g 509170.6 AB	5DF1854 k	scR	Iron-sulfur cl	0.994	0.994 bi	NC_010410	1978	157 fig 5093
1310696.4.cr	7	405 fig 1310696.4.peg.7	Cysteine des	5217	6434 +	bi	NC_010400	1771	405 fi	g 509170.6 AB	5DF1853 it	icS	Cysteine des	0.995	0.998 bi	NC_010410	1979	405 fig 509
310696.4.ct	8	128 fig 1310696.4.peg.8	Iron-sulfur cl	6501	6887 +	bi	NC_010400	1770	128 fi	g 509170.6 AB	5DF1852 it	scU	Iron-sulfur cl	0.992	0.992 bi	NC_010410	1980	128 fig 509
310696.4.ct	9	106 fig 1310696.4.peg.9	Iron binding	6918	7238 +	bi	NC_010400	1769	106 fi	g 509170.6 AB	SDF1851 is	scA	Iron binding	0.981	0.991 bi	NC_010410	1981	106 fig 509
310696.4.ct	10	172 fig 1310696.4.peg.10	Chaperone p	7324	7842 +	bi	NC_010400	1768	172 fi	g 509170.6 AB	SDF1850 h	scB	Chaperone p	0.975	0.913 bi	NC_010410	1982	172 fig 509
310696.4.ct	11	619 fig 1310696.4.peg.11	Chaperone p	7881	9740 +	bi	NC_010400	1767	620 fi	g 509170.6 AB	5DF1849	scA	Chaperone p	0.974	0.998 bi	NC_010410	1983	619 fig 509
310696.4.ct	12	112 fig 1310696.4.peg.12	Ferredoxin, 2	9759	10097 +	bi	NC_010400	1766	112 fi	g 509170.6 AB	5DF1848 f	dx	Ferredoxin, 2	0.991	0.991 bi	NC_010410	1984	112 fig 509
310696.4.ct	13	489 fig 1310696.4.peg.13	Adenylate cy	10146	11615 -	bi	NC_010400	1765	328 fi	g 509170.6.pe	2.1765		Adenylate cy	1	0.997 bi	NC_010410	1985	489 fig 509
310696.4.ct	14	144 fig 1310696.4.peg.14	Diadenosine	11668	12102 -	bi	NC_010400	1762	144 fi	g 509170.6 AB	5DF1844		Diadenosine	1	0.993 bi	NC_010410	1986	144 fig 509
310696.4.cr	15	1153 fig 1310696.4.peg.15	Transcription	12304	15765 -	bi	NC_010400	1761	1153 fi	g 509170.6 AB	5DF1843 m	nfd	Transcription	0.975	0.999 bi	NC_010410	1987	1153 fig 509
310696.4.ct	16	118 fig 1310696.4.peg.16	FIG00350213	15900	16256 -		-								bi	NC_010410	1988	118 fig 509
310696.4.ct	17	313 fig 1310696.4.peg.17	Predicted dy	16337	17278 -	bi	NC_010400	1759	180 fi	g 509170.6.pe	1759		Predicted dy	0.961	0.994 bi	NC_010410	1989	310 fig 509
310696.4.ct	18	62 fig 1310696.4.peg.18	expressed pr	17415	17603 -	uni	NC_010400	1756		g 509170.6.pe			expressed pr	0.839	0.339 uni	NC_010410	1990	259 fig 509
310696.4.ct	19	173 fig 1310696.4.peg.19	expressed pr	17672	18193 -	bi	NC 010400	1756		g 509170.6.pe			expressed pr	0.885	0.478 bi	NC 010410	1990	259 fig 509
310696.4.ct	20	168 fig 1310696.4.peg.20	Ribonuclease	18218	18724 -	bi	NC 010400	1752		g 509170.6 AB		raA	Ribonuclease	0.988	0.994 bi	NC 010410	1991	168 fig 509
310696.4.cr	21	221 fig 1310696.4.peg.21	Oxidoreduct	18736	19401 -	bi	NC 010400	1751		g 509170.6 AB			Oxidoreduct	0.959	0.995 bi	NC 010410	1992	221 fig 509
310696.4.cr	22	88 fig 1310696.4.peg.22	SSU ribosom	19614	19880 +	bi	NC 010400	1750		g 509170.6 AB		psT	SSU ribosom	1	0.989 bi	NC 010410	1993	88 fig 509
310696.4.ct	23	244 fig 1310696.4.peg.23	FIG00968626	19937	20671 -	bi	NC 010400	1749		g 509170.6 AB			FIG00968626	0.971	0.996 bi	NC 010410	1994	244 fig 509
310696.4.ct	24	285 fig 1310696.4.peg.24	3'(2'),5"-bispl	20729	21586 -	bi	NC 010400	1748		g 509170.6 AB		vsQ	3'(2'),5'-bispl	0.965	0.996 bi	NC 010410	1995	285 fig 509
310696.4.ct	25	122 fig 1310696.4.peg.25	FIG00350888	21896	22264 +	bi	NC 010400	1747		g 509170.6 AB			FIG00350888	0.984	0.992 bi	NC 010410	1996	122 fig 509
310696.4.cr	26	244 fig 1310696.4.peg.26	FIG00351092	22318	23052 -	bi	NC 010400	1746		g 509170.6 AB			FIG00351092	0.991	0.995 bi	NC 010410	1997	219 fig 509
310696.4.cr	27	576 fig 1310696.4.peg.27	Dipeptide tra	23085	24815 -	bi	NC 010400	1745		g 509170.6 AB			Dipeptide tra	0.964	0.998 bi	NC 010410	1998	576 fig 509
310696.4.cr	28	380 fig 1310696.4.peg.28	Oligopeptide	24828	25970 -	bi	NC 010400	1744		g 509170.6 AB			Oligopeptide	0.972	0.997 bi	NC 010410	1999	380 fig1509
310696.4.cr	29	312 fig 1310696.4.peg.29	Dipeptide tra	25978	26916 -	bi	NC 010400	1743		g 509170.6 AB			Dipeptide tra	0.965	0.997 bi	NC 010410	2000	312 fig 509
310696.4.cr	30	670 fig 1310696.4.peg.30	Thimet oligo	26929	28941 -	hi	NC 010400	1742		g 509170.6 AB			Thimet oligo	0.976	0.999 bi	NC 010410	2001	670 fig 509
310696.4.cr	31	606 fig 1310696.4.pcg.31	Dipeptide-bi	28963	30783 -	bi	NC 010400	1741		g 509170.6 AB			Dipeptide-bi	0.914	0.998 bi	NC 010410	2002	606 fig1509
310696.4.cr	32	610 fig 1310696.4.peg.32	Dipeptide-bi	30802	32634 -	bi	NC 010400	1740		g 509170.6 AB			Dipeptide-bi	0.929	0.998 bi	NC 010410	2003	594 fig 509
310696.4.cr	33	47 fig 1310696.4.peg.33	hypothetical	33132	33275 +				224								2000	
310696.4.cr	34	890 fig 1310696.4.peg.34	TonB-depend	33369	36041 +	ы	NC 010400	1739	871 6	g 509170.6 AB	SDF1822		Ton8-depend	0.761	0.999 bi	NC 010410	2004	871 fig 509
310696.4.cr	35	238 fig 1310696.4.peg.35	Ferric sidero	36152	36868 +	hi	NC 010400	1738		g 509170.6 AB			Ferric sidero	0.933	0.996 bi	NC 010410	2005	238 fig 509
310696.4.cr	36	292 fig 1310696.4.pcg.36	MotA/TolQ/I	36912	37790 +	bi	NC 010400	1737		g 509170.6 AB			MotA/TolQ/I	0.952	0.997 bi	NC 010410	2005	293 fig1509
310696.4.ct	30	140 fig 1310696.4.peg.30	Biopolymer t	37806	38228 +	bi	NC 010400	1736		g 509170.6 AB			Biopolymer t	0.932	0.997 bi	NC 010410	2006	140 fig 509
310696.4.cr	37	137 fg 1310696.4.peg.38	Biopolymer t	3/800	38661 +	bi	NC 010400	1735		g 509170.6 AB			Biopolymer t	0.988	0.993 bi	NC 010410	2007	140 fig 509
310696.4.ct	39	47 fig 1310696.4.peg.39	FIG0035085:	38717	38860 -				13/ 1	61262170.0 MD.	. 1010		supporyther t	0.376	0.555 01	010410	2000	207 lig 303
1310696.4.cr	40	4/ fig 1310696.4.peg.39 721 fig 1310696.4.peg.40	Malate synth	38/1/	41129 -	bi	NC 010400	1734	721 6	g 509170.6 AB	051917	6-9	Malate synth	0.99	0.999 bi	NC 010410	2009	721 fig 509
1310696.4.ct 1310696.4.ct	40			41500	41129 -	bi		1/34				jub (		0.99	0.999 bi	NC_010410 NC 010410	2009	
1310696.4.cr	41	380 fig 1310696.4.peg.41 339 fig 1310696.4.peg.42	ATPase, AFG	41500	42642 -	DI	NC_010400 NC_010400	10/0	380 1	g 509170.6 AB	our1/40		ATPase, AFG	0.984	0.997 01	NC_010410	2010	380 fig 509:

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