

Chryseobacterium haifense DSM 19056^T, A Genomic Report

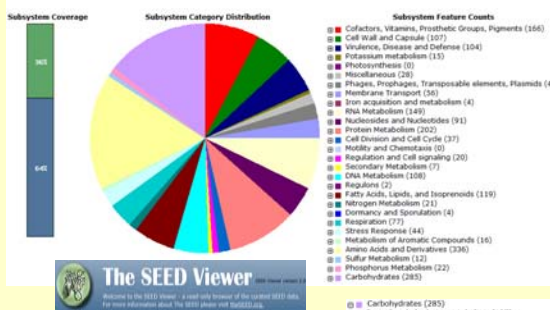
Thomas Sontag, Jeffrey D. Newman, Lycoming College Biology Department, Williamsport, PA



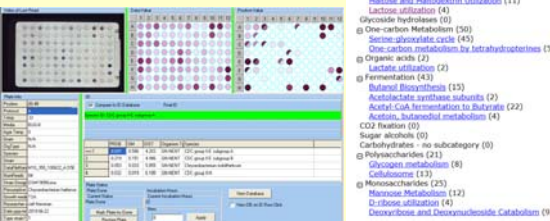
Abstract

The phylogenetic position of a number of bacteria within the family *Flavobacteriaceae* has been questioned. To address the question, the whole genomes of several organisms were sequenced, and this project is focused on *Chryseobacterium haifense* DSM 19056. The advances in next generation sequencing (NGS) technologies have caused a decrease in cost for whole genome sequencing. This decreased cost has led to more genomes being sequenced and in the process has caused a large demand for bioinformatics tools to handle the genomic data. To analyze the genomic data, the 930,000 reads were assembled in several steps, using several different software packages to refine the assembly to fewer than 700 contiguous sequences. Automated annotation using the Rapid Annotation using Subsystem Technologies (RAST) server identified the organism's genes and known pathways which were compared to its phenotypes. The Reciprocal Orthology Score Average (ROSA) genomic similarity calculator showed that *Chryseobacterium haifense* is as different from "true" *Chryseobacterium* as other separate genera are which has led to the conclusion that *Chryseobacterium haifense* does not belong within the *Chryseobacterium* genus.

RAST/SEED Subsystem Analysis of *C. haifense*



Biolog GenIII Plate Results

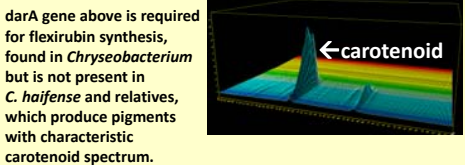


avg control	22.6	gelatin	72.2
dextrin	26.4	glycyl-L-proline	75.2
D-maltose	89.2	L-alanine	6.8
D-trehalose	6.8	L-arginine	8.6
D-cellobiose	12.8	L-aspartic acid	39.4
gentiobiose	9.6	L-glutamic acid	98.4
sucrose	37.2	L-histidine	14.8
D-tartrate	6.8	L-phenylalanine acid	9.2
stachyose	6.6	L-serine	9
pos control	99	lincosamine	10.8
pH 6	98.6	guanidine HCl	11.8
pH 5	16.8	niaproof 4	15.6
D-raffinose	21.4	pectin	97
is-D-lactose	16.8	D-galacturonic acid	27.4
D-melibiose	6.8	L-galacturonic acid lactone	3.8
β-methyl-D-glucoside	6.6	D-gluconic acid	16.2
D-salicin	4.8	D-glucosaminic acid	22.8
N-acetyl-D-glucosamine	4.6	glucuronamide	23.2
N-acetyl-β-D-mannosamine	4.6	muslic acid	17
N-acetyl-D-galactosamine	7.8	quinic acid	10.8
N-acetylneuraminic acid	4.8	D-saccharic acid	9.6
1% NaCl	58	vancomycin	10.8
4% NaCl	20.8	tetraazolum violet	45.6
8% NaCl	22.8	tetrazolum blue	92.8
is-D-glucose	43.6	p-hydroxy-phenylacetic acid	13.2
D-mannose	22.6	methyl pyruvate	7.2
D-fructose	59.4	D-lactic acid methyl ester	16
D-galactose	10.6	L-lactic acid	10.8
3-methyl glucose	9.8	citric acid	12.4
D-fucose	9.2	α-keto-glutaric acid	11.2
L-fucose	12	D-malic acid	4.6
L-rhamnose	8.4	L-malic acid	9
inosine	5	β-bromo-succinic acid	4.6
1% Na-lactate	9.8	malonic acid	14.8
fucose acid	11	malic acid	13.8
D-serine	59.6	Kristalline	97.6
D-sorbitol	10.4	β-cam-40	87.6
D-mannitol	6.2	α-amino-butyric acid	12
D-arabitol	4.8	α-hydroxy-butyric acid	14
myo-inositol	5	β-hydroxy-D-l-butyric acid	14.2
glycerol	5	α-keto-butyric acid	7.6
D-glucoside-β-PO4	83.6	acetoacetic acid	43.6
D-fructose-β-PO4	16.4	propionic acid	14.2
D-aspartic acid	4.6	acetic acid	7.2
D-serine	7.2	formic acid	8.4
treoleandomycin	9.2	atitram	97.6
rifamycin SV	10	Na-butyrate	20.2
minocycline	14	Na bromate	22.8

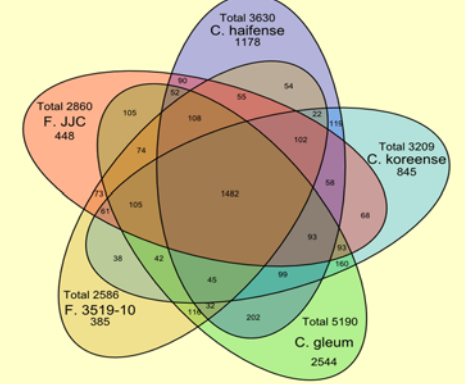
RAST/SEED Sequence Based Comparison

Reference	Chryseobacterium gleum F93, ATCC 33910 (332327.3)
Comparison Organism 1	Chryseobacterium haifense DSM 19056 (421825.12) [BaseCallPut]
Comparison Organism 2	Chryseobacterium gleum CGUG 49869 (232234.6) [BaseCallPut]
Comparison Organism 3	Flavobacteriaceae bacterium JS19 (531844.6) [BaseCallPut]
Comparison Organism 4	Flavobacteriaceae bacterium JJC (531842.7) [BaseCallPut]

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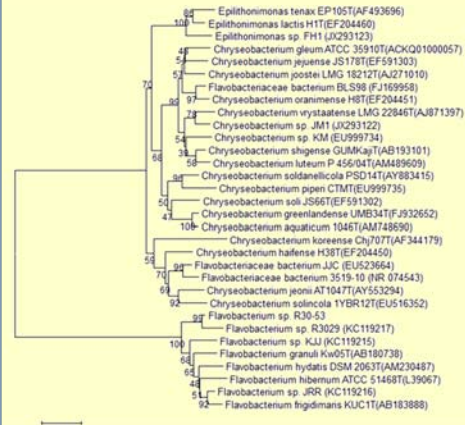
Gene Distribution Venn Diagram



Venn Diagram Conclusions

There are 102 genes shared among *C. haifense*, *C. koreense*, *F. JJC* and *F. 3519-10* that are missing in *C. gleum*, which indicates the presence of genetic differences. The genes have not yet been analyzed to determine if they contribute to phenotypic differences between the two groups. All of the organisms share 1482 genes. *C. gleum* has 2544 genes that are not shared with any others in the comparison. *C. gleum* has significantly more genes, due to its larger genomic sequence, than the other organisms in the comparison, which suggests a significant difference between the two groups.

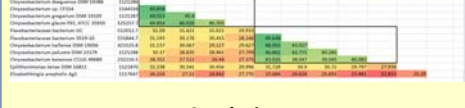
Phylogenetic Tree based on 16S rRNA Gene



Average Amino Acid Identity (AAI) Matrix



Reciprocal Orthology Score Average (ROSA) Matrix



Conclusions

- 16S rRNA phylogeny and genomic analysis (ROSA) indicate that *Chryseobacterium haifense* DSM 19056 should not be classified as a member of the genus *Chryseobacterium*.
- Based on the Bacteriological Code, the genus *Kaistella* should be reinstated with *K. koreensis* and the type species
- Many current *Chryseobacterium* spp. should be moved into the genus *Kaistella*

Acknowledgements

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References

- Aziz, R.K. et al. (2008). The RAST server: Rapid annotations using subsystems technology. *BMC Genomics* 9:75.
- Hantsis-Zacharov E, and Halpern M. (2007). *Chryseobacterium haifense* sp. nov., a psychrotolerant bacterium isolated from raw milk. *USM J* 57: 2344-8.
- Kämpfer, P., et al. (2009). Description of *Chryseobacterium anthrapi* sp. nov. to accommodate clinical isolates biochemically similar to *Kaistella koreensis* and *Chryseobacterium haifense*, proposal to reclassify *Kaistella koreensis* as *Chryseobacterium koreense* comb. nov. and amended description of the genus *Chryseobacterium*. *USM J* 59, 2421-8.
- Kim, M. et al