

Is Sharing Caring?

Elucidating the Effects of the
Presence of CRISPR-Cas Systems
on Rates of Horizontal Gene
Transfer Using Network Analysis

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MolBiol 4C12 Thesis



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Biology Department,
McMaster University

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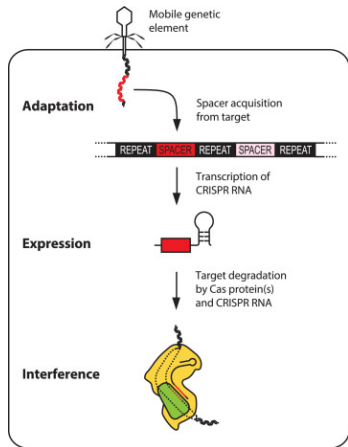
1. CRISPR-Cas systems
2. Horizontal Gene Transfer
3. Phylogenomic Networks
4. Do CRISPR Systems Affect Horizontal Gene Transfer?
5. My Project
6. Results

CRISPR-Cas systems

What Are They?

What Are They?

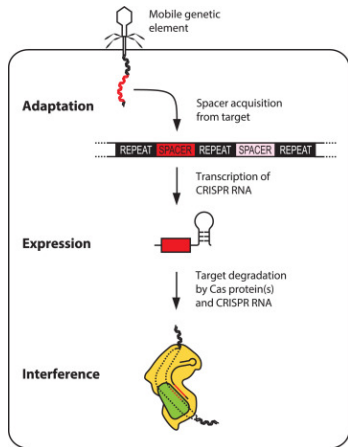
- Adaptive Bacterial Immune System



(Rath et al., 2015)

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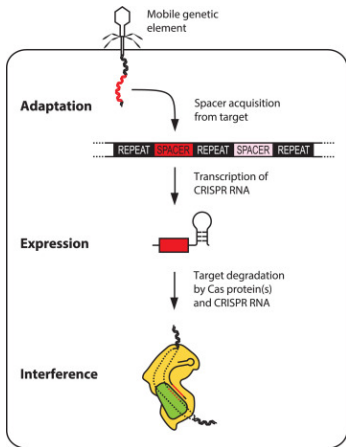
- Adaptive Bacterial Immune System
- Protects against foreign DNA



(Rath et al., 2015)

What Are They?

- Adaptive Bacterial Immune System
- Protects against foreign DNA
- Requires Cas proteins and CRISPR loci

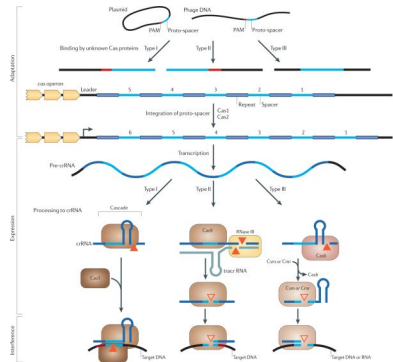


(Rath et al., 2015)

Diversity & Ubiquity

Diversity & Ubiquity

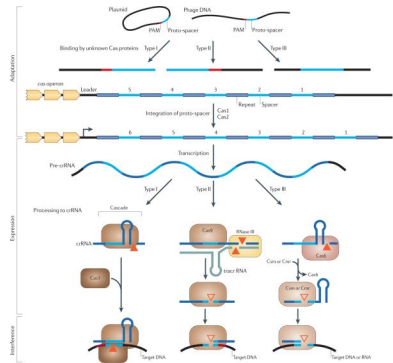
- 45% of bacteria have CRISPR loci ($n = 6782$) (Grissa, I. and Drevet, C. and Couvin, D., 2017)



(Makarova et al., 2011)

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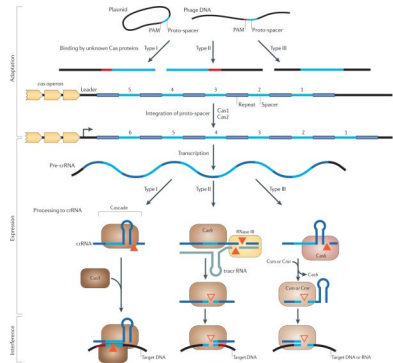
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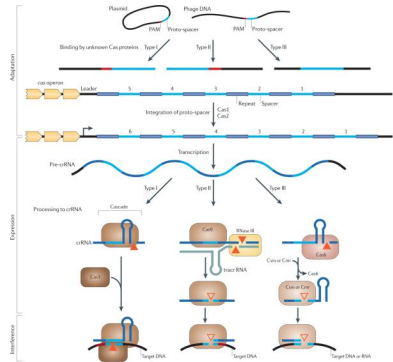
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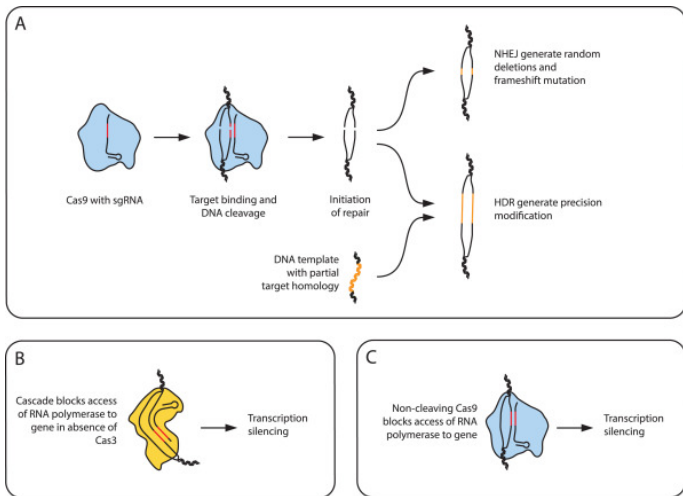
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- 3 Main Types, multiple sub types (Bondy-Denomy and Davidson, 2014)
- CRISPR arrays represent unique life history of an organism
- 11% – 28% are false or orphaned CRISPR loci (Zhang and Ye, 2017)



(Makarova et al., 2011)

Biotech Application

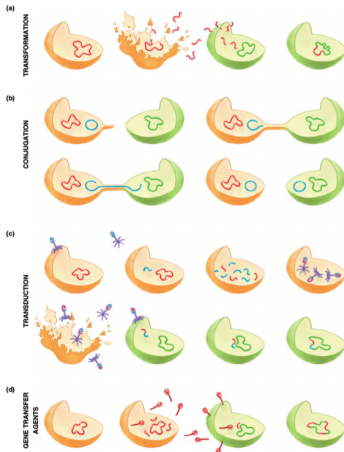
Biotech Application



(Rath et al., 2015)

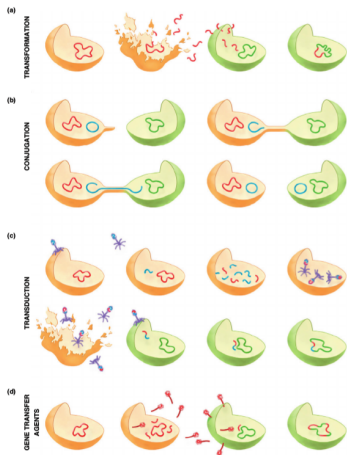
Horizontal Gene Transfer

Mechanisms



(Popa and Dagan, 2011)

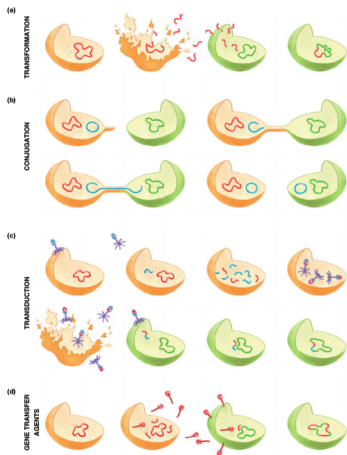
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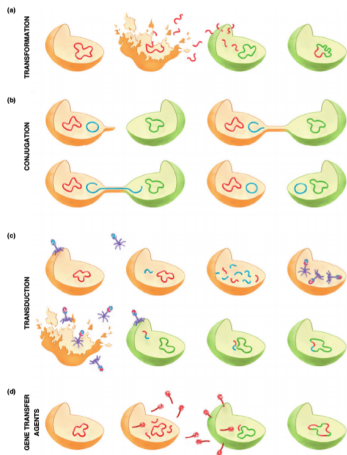
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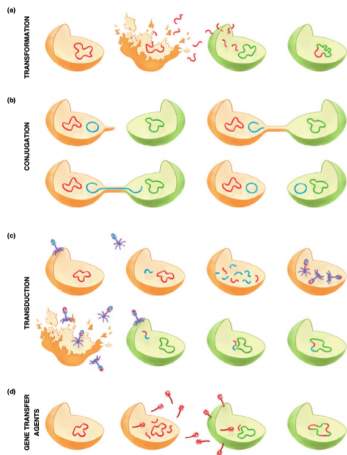
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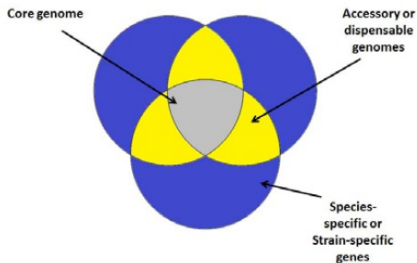


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- **CRISPR-Cas directly affects HGT** (Popa and Dagan, 2011)

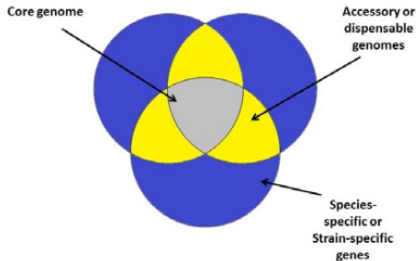
Pan-Genomes

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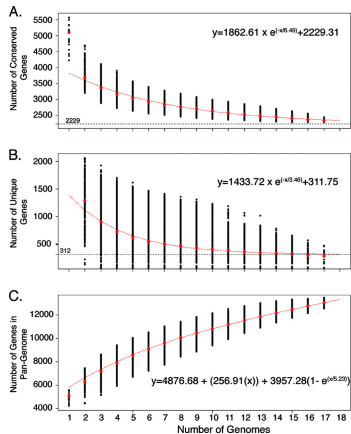


(Guimaraes et al., 2015)

Pan-Genomes



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(Rasko et al., 2008)

Rate Influencing Factors

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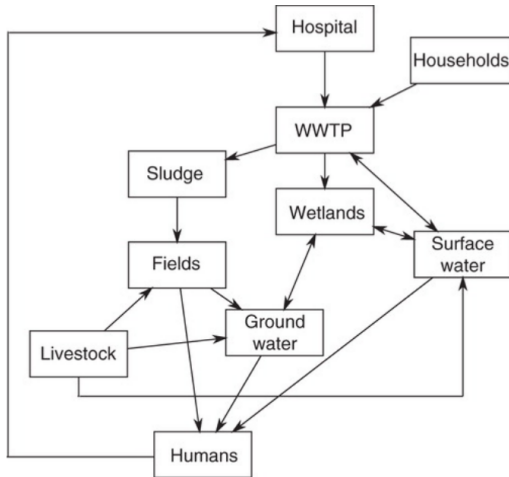
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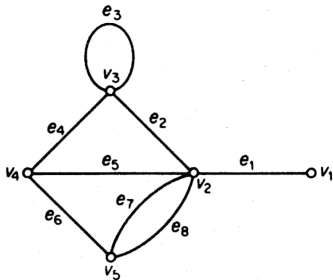


(Berglund, 2015)

Phylogenomic Networks

What is A Network?

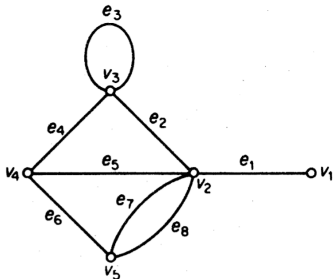
What is A Network?



- Useful mathematical abstraction of real world system

(Bondy and Murty, 2002)

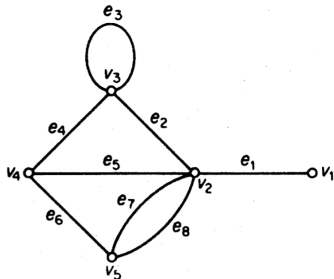
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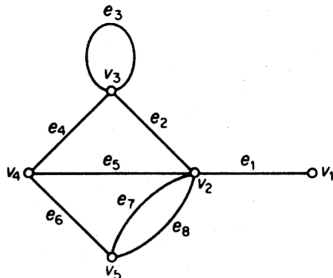
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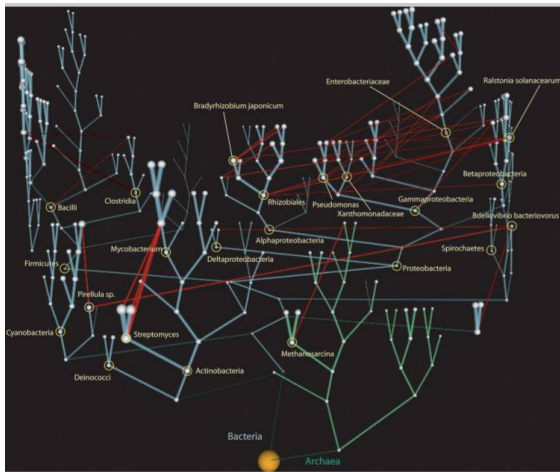


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- Directed or Undirected Edges
- Weighted or Unweighted Edges

(Bondy and Murty, 2002)

Prokaryotic “Net of Life”

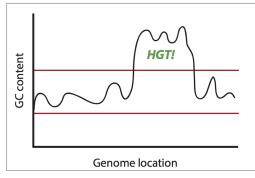
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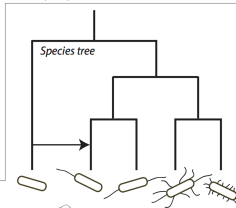
(Kunin et al., 2005)

Construction

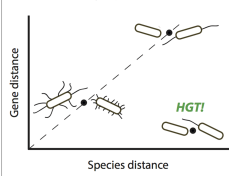
1. Parametric methods



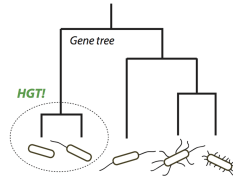
2. Phylogenetic methods



2a. Implicit phylogenetic methods



2b. Explicit phylogenetic methods



(Ravenhall et al., 2015)

Do CRISPR Systems Affect Horizontal Gene Transfer?

Yes

CRISPR Cost Complexity

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- Cost trade off factors:

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 - Prophage abundance (Watson, Staals, and Fineran, 2018)

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- CRISPR can enhance transduction-mediated HGT (Watson, Staals, and Fineran, 2018)

Previous Findings

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 - Can see inhibitory effects of CRISPR on HGT over short evolutionary time scales
 - Higher gene indel rates for CRISPR containing OTUs than non-CRISPR containing outgroups

My Project

Hypothesis

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Null Hypothesis

Bacterial strains or genera with known CRISPR systems will show no significant differences in network statistics compared to those strains or genera without known CRISPR systems

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Alternative Hypothesis

Bacterial strains or genera with known CRISPR systems will show a significant difference in at least 1 network statistic compared to those strains or genera without known CRISPR systems.

Objectives

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Within Network Comparisons

For genera with CRISPR containing OTUs, compare the node statistics of CRISPR containing OTUs to non-CRISPR containing OTUs.

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Gene Indel Rates vs. Network Statistics

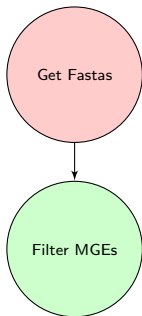
Compare gene Indel rates to node/network statistics for CRISPR containing and non-CRISPR containing OTUs

Workflow

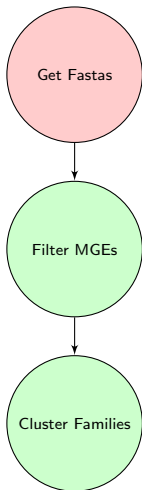


Get Fastas

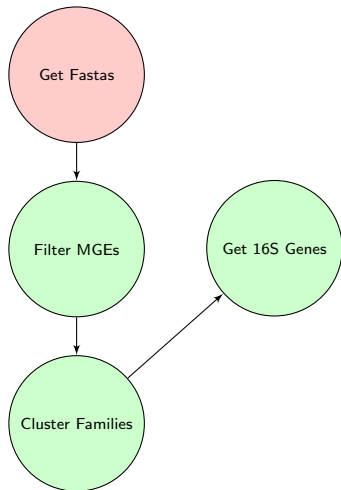
Workflow



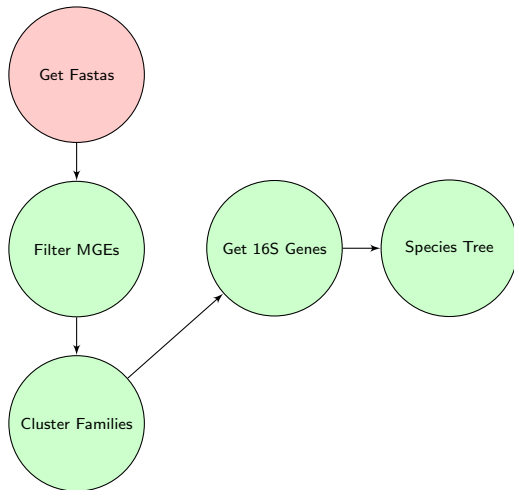
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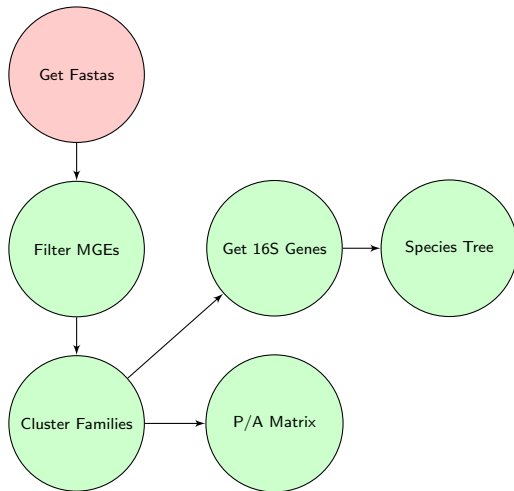
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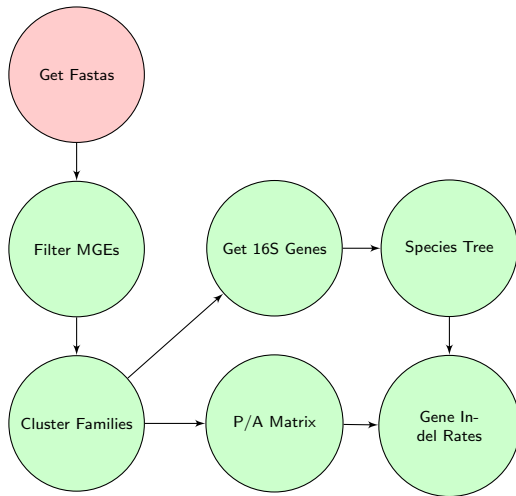
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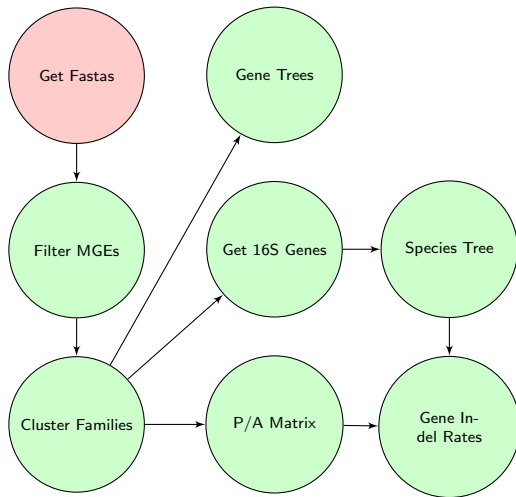
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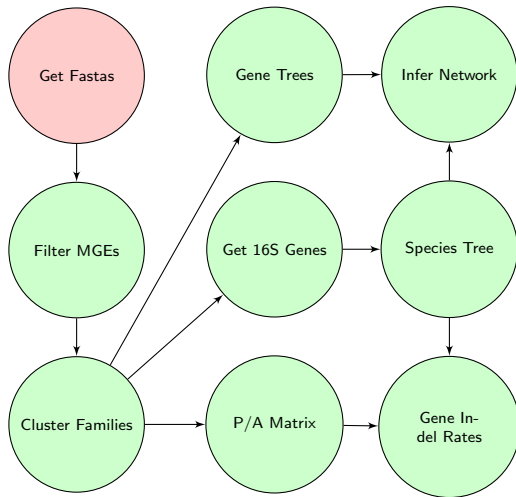
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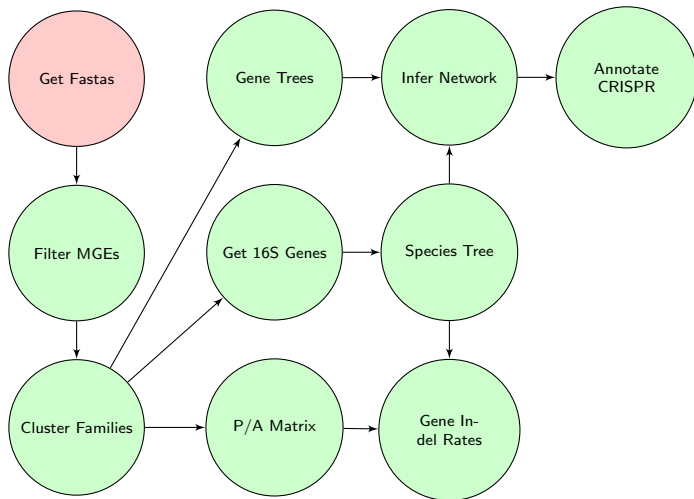
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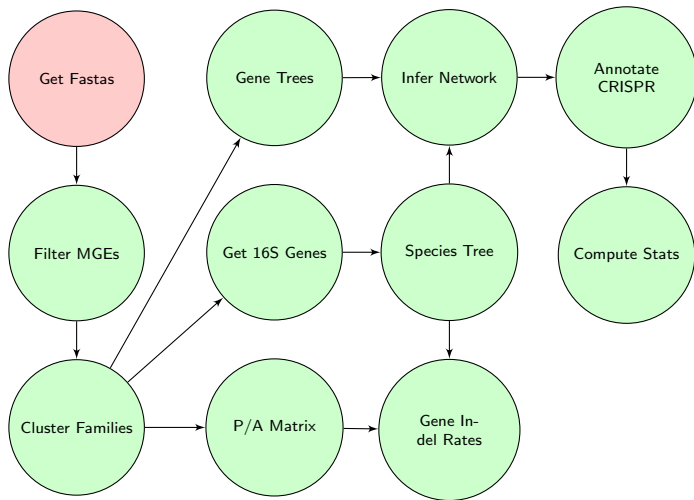
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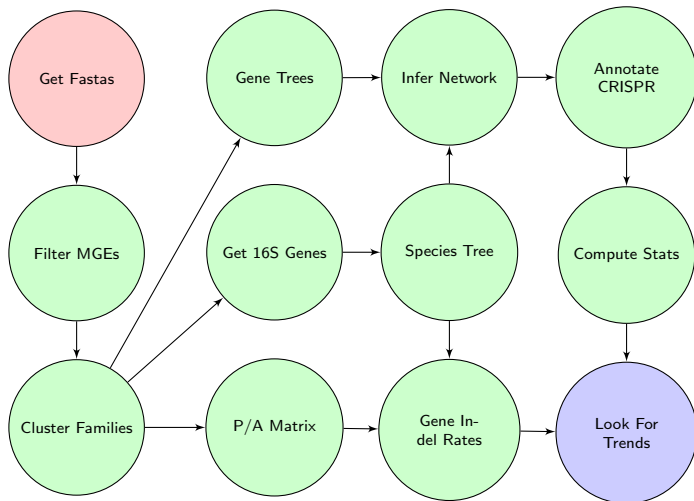
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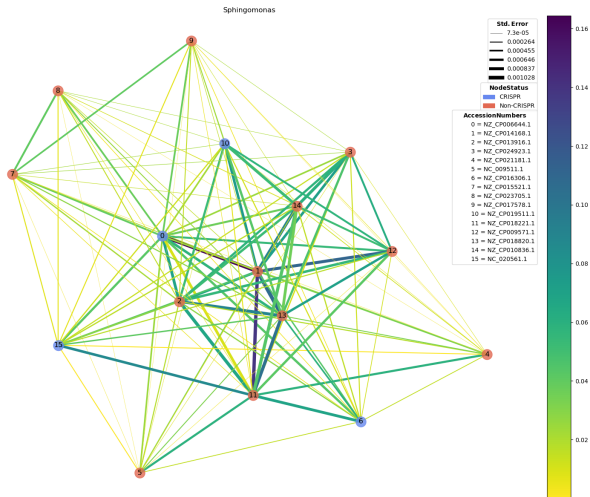
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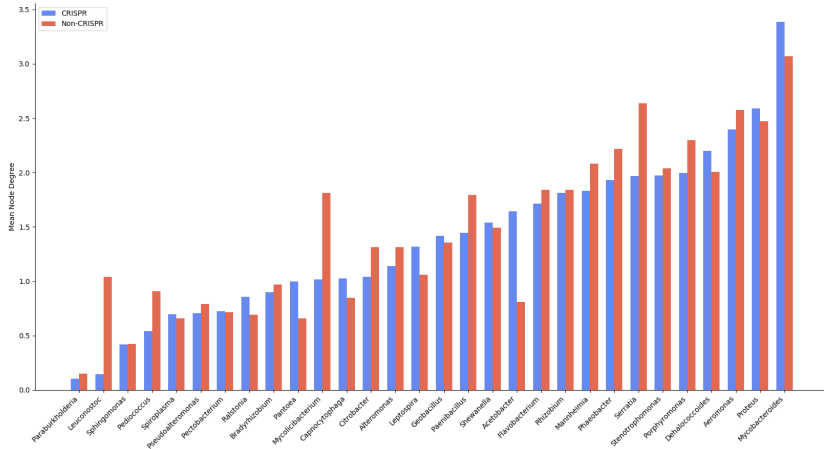
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- **Network Modularity:** $Q = \frac{1}{2m} \sum_{uv} [W_{uv} - \frac{k_u k_v}{2m}] \delta(u, v)$ where m is the total weight of all edges, k_u is the degree of u and $\delta(u, v)$ is 1 if u and v both have or do not have CRISPR systems and 0 otherwise. $Q \in [-1, 1]$ (Newman, 2004)

Results

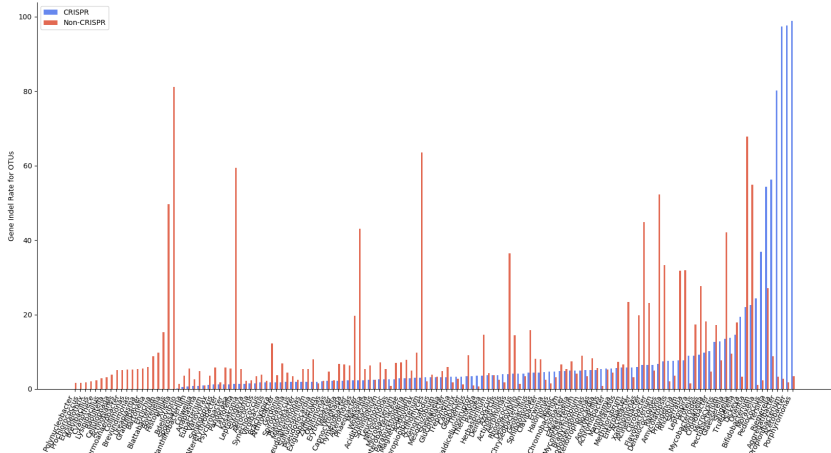
Example “Consensus” Network



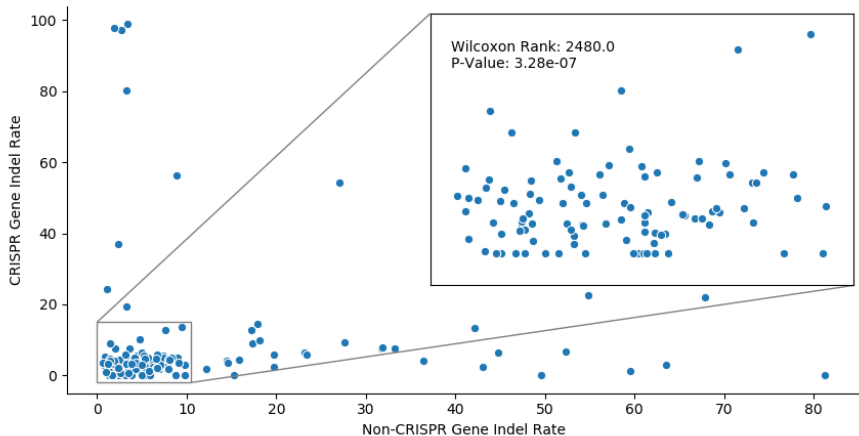
Mean Node Degree



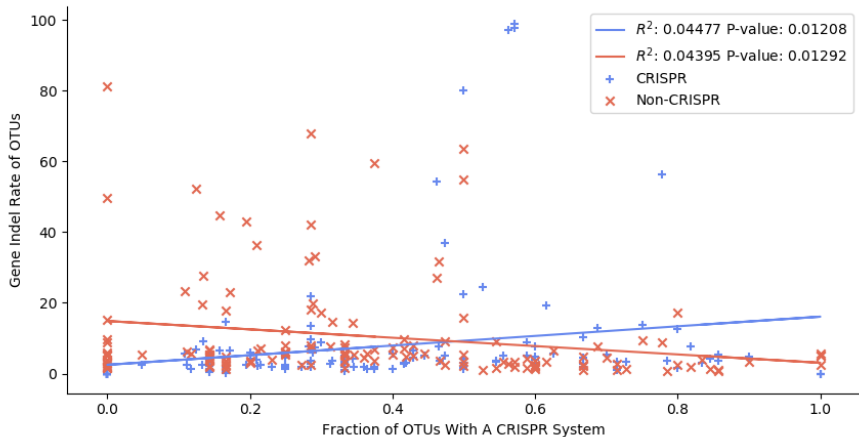
Gene Indel Rates



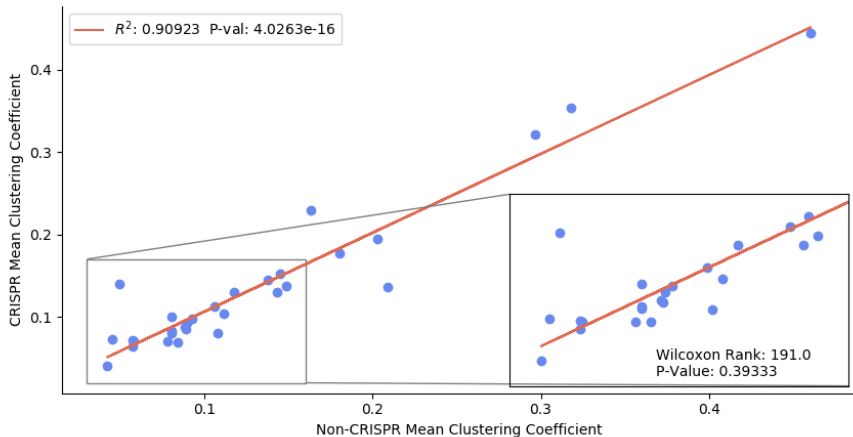
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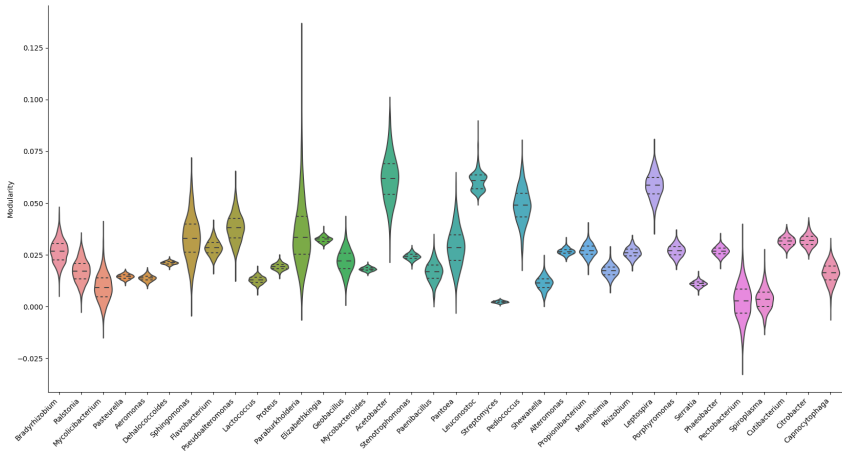
Gene Indel Rate Vs. Fraction of CRISPR OTUs



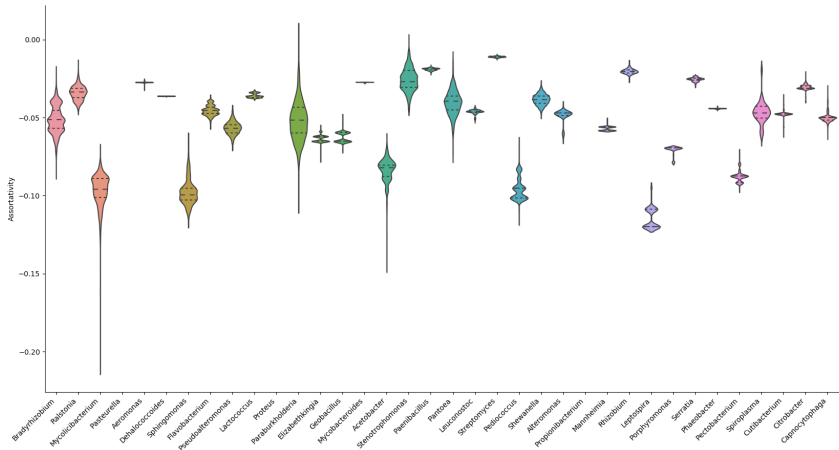
Mean Node Weighted Clustering Coefficient



Modularity Distributions



Assortativity Distributions



Conclusion

Findings

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- High mixing between CRISPR and non-CRISPR OTUs
- Population level effects of CRISPR-Cas systems may decrease HGT rates
- Interplay of CRISPR-Cas systems and HGT is complex and warrants further study

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- **Intergenic comparisons:** Combine any set of fasta files from OTUs for analyzing transfer dynamics
- **Considering bacterial ecology and environments:** Consider geographically close OTUs or differences between networks due to environmental factors

Is Sharing Caring?

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Yes, for researchers

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Yes, for researchers
Jury's still out for bacteria

Thanks

Thank you to

- Dr. G. Brian Golding
- Dr. Ben Evans
- The Golding lab
 - Caitlin Simopoulos
 - Daniella Lato
 - Zachery Dickson
 - Sam Long
 - Geoge Long
 - Lucy Zhang
 - Brianne Laverty
 - Nicole Zhang
- Everyone here for listening



All code used for this project is available at https://github.com/DJSiddharthVader/thesis_SidReed

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