WGS in Florida
Impacts in the Food Lab

S. Brian Caudle, MS

Division of Food Safety
Florida Department of Agriculture and Consumer Services
GenomeTrakr 2018
Stanley.Caudle@freshfromflorida.com
Getting Started

- The Early Days
- Wet-Work
- Starting Analysis
- Dedicated Resources
- Protocols

- MiSeq Purchased in 2015
- Attend Training Programs
  - GenomeTrakr MiSeq Training
  - PulseNet MiSeq Training
- Rearrange Lab to Install Sequencer
Getting Started

- The Early Days
- Wet-Work
- Starting Analysis
- Dedicated Resources
- Protocols

- Follow GenomeTrakr Protocols
- Optimize for Local Conditions
- Purchase Additional Equipment
- Start Working on Internal SOP
Getting Started

- The Early Days
- Wet-Work
- Starting Analysis
- Dedicated Resources
- Protocols

- Start Off Basic
- Test Software for Usefulness
- Develop Workflow/Pipeline
Getting Started

- The Early Days
- Wet-Work
- Starting Analysis
- Dedicated Resources
- Protocols
- Adequate Space for Equipment
- Dedicated or Shared Benches?
- Specialized Analysts/Cross Training
- Analytical Requirements
Getting Started

- The Early Days
- Wet-Work
- Starting Analysis
- Dedicated Resources
- Protocols
- Data Collection – FDA GenomeTrakr
- QA/QC – Follow Network
- Data Analysis – Internal Pipelines
- National Comparisons – NCBI
Supporting Cast

- Bureau of Inspections
- Rapid Response Team
- Division Leadership
- Primary Customer
- State Sampling Provider
- Enforcement Responsibilities
- Requirement – Easily Understood Reports
Supporting Cast

- Bureau of Inspections
- Rapid Response Team
- Division Leadership

- Investment in Rapid Methods
- Funds for Supplemental Supplies
- Provide for Computational Requirements
Supporting Cast

- Bureau of Inspections
- Rapid Response Team
- Division Leadership

- Supporting Program Expansion
- Integration of WGS into Regulatory Decisions
- Expanding Understanding of Capabilities/Limitations
Laboratory Worksheets

- Bench Worksheets
- *de novo* Reports
- Additional Information

- Covers Bench-Work Requirements
- From Isolate Through Sequence
- Modeled After PFGE Workflow
Laboratory Worksheets

- Bench Worksheets
- *de novo* Reports
- Additional Information

- Relevant QC Metrics
- Basic Metagenomics Report
- Easy Review of Run
Laboratory Worksheets

- Bench Worksheets
- *de novo* Reports
- Additional Information

- Metagenomics Profiles
- Assembly Graphs
- Specialized Info Upon Request
### de novo Report

<table>
<thead>
<tr>
<th>Assembly</th>
<th>de novo #_contigs</th>
<th>WGS Total Length</th>
<th>Pipeline Ave_Insert_Si</th>
<th>Metrics Ave_Coverage</th>
<th>01_29_2018 Pct_Lm</th>
<th>Analyst:</th>
<th>Pct_Slm</th>
<th>Pct_Ec</th>
<th>Pct_Other</th>
</tr>
</thead>
<tbody>
<tr>
<td>FLAG-XXXXX</td>
<td>33</td>
<td>4718263</td>
<td>403.24</td>
<td>134.68</td>
<td>0</td>
<td>95.85522</td>
<td>0</td>
<td>4.14478</td>
<td></td>
</tr>
<tr>
<td>FLAG-XXXXX</td>
<td>116</td>
<td>4708331</td>
<td>388.06</td>
<td>151.12</td>
<td>0</td>
<td>95.92731</td>
<td>0</td>
<td>4.07269</td>
<td></td>
</tr>
<tr>
<td>FLAG-XXXXX</td>
<td>47</td>
<td>5003027</td>
<td>395.46</td>
<td>121.46</td>
<td>0</td>
<td>96.06801</td>
<td>0</td>
<td>3.93199</td>
<td></td>
</tr>
<tr>
<td>FLAG-XXXXX</td>
<td>51</td>
<td>4663740</td>
<td>391.58</td>
<td>145.95</td>
<td>0</td>
<td>96.61282</td>
<td>0</td>
<td>3.38718</td>
<td></td>
</tr>
<tr>
<td>FLAG-XXXXX</td>
<td>32</td>
<td>4642433</td>
<td>394.06</td>
<td>141.14</td>
<td>0</td>
<td>100</td>
<td>0</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>FLAG-XXXXX</td>
<td>51</td>
<td>4663459</td>
<td>353.85</td>
<td>166.36</td>
<td>0.02688</td>
<td>93.47261</td>
<td>0</td>
<td>6.5005</td>
<td></td>
</tr>
<tr>
<td>FLAG-XXXXX</td>
<td>55</td>
<td>4655048</td>
<td>385.53</td>
<td>132.12</td>
<td>0.02704</td>
<td>94.76515</td>
<td>0</td>
<td>5.20781</td>
<td></td>
</tr>
<tr>
<td>FLAG-XXXXX</td>
<td>40</td>
<td>3086346</td>
<td>368.89</td>
<td>197.43</td>
<td>99.37338</td>
<td>0</td>
<td>0</td>
<td>0.62662</td>
<td></td>
</tr>
<tr>
<td>FLAG-XXXXX</td>
<td>137</td>
<td>3070740</td>
<td>364.95</td>
<td>186.3</td>
<td>99.78331</td>
<td>0</td>
<td>0</td>
<td>0.21662</td>
<td></td>
</tr>
<tr>
<td>FLAG-XXXXX</td>
<td>22</td>
<td>3067106</td>
<td>376.63</td>
<td>187.96</td>
<td>99.84636</td>
<td>0</td>
<td>0</td>
<td>0.15364</td>
<td></td>
</tr>
<tr>
<td>FLAG-XXXXX</td>
<td>24</td>
<td>2959006</td>
<td>387.52</td>
<td>200.1</td>
<td>100</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>FLAG-XXXXX</td>
<td>24</td>
<td>3037051</td>
<td>370.04</td>
<td>187.12</td>
<td>100</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>FLAG-XXXXX</td>
<td>25</td>
<td>3027332</td>
<td>361.96</td>
<td>195.61</td>
<td>100</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>FLAG-XXXXX</td>
<td>18</td>
<td>3005378</td>
<td>331.06</td>
<td>193.44</td>
<td>100</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>FLAG-XXXXX</td>
<td>16</td>
<td>2924238</td>
<td>353.42</td>
<td>205.54</td>
<td>100</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td></td>
</tr>
<tr>
<td>FLAG-XXXXX</td>
<td>19</td>
<td>2885431</td>
<td>384.4</td>
<td>196.83</td>
<td>100</td>
<td>0</td>
<td>0</td>
<td>0</td>
<td></td>
</tr>
</tbody>
</table>
Local Databases

- Organism Specific
  - Rough Pass Analysis
  - Cluster Detection
  - *Listeria monocytogenes*
  - *Escherichia coli*
  - *Salmonella*
  - More as Lab Testing Expands
Local Databases

- Organism Specific
- Rough Pass Analysis
- Cluster Detection
- k-mer based
- Reference-Free
- Clusters for SNP analysis
Local Databases

- Organism Specific
- Rough Pass Analysis
- Cluster Detection
- Search Tree for Isolate
- Identify relevant clade
- Prepare Input for SNP analysis
SNP reports

- CFSAN SNP pipeline
- Report Generation
- Interpretation

- Prepare Input File
- Reference Determination
- Run on Samples of Interest
SNP reports

- CFSAN SNP pipeline
- Report Generation
- Interpretation

- Phylogeny
- Annotate Tree with Sample Information
- Image Editing – Overlay Info
SNP reports

- CFSAN SNP pipeline
- Report Generation
- Interpretation

- Sample of Interest Highlighted
- Similarity Stated Clearly
- Easy to Understand
Early Reports

- Early Reports Were Hard To Read
- Customer Feedback
- Improvement Through Iteration
Current Reports

- Easy to Read: Bold Colors
- Sample Metadata Included
- Plain Language SNP Matching

Group A: 13 SNPs
Group B: >1,000 SNPs
Group C: >50 SNPs
Group D: >90 SNPs
Group E: >1900 SNPs
FLAG-29962 DOES NOT MATCH any samples in the local database. Nearest related sample is >40 SNP different.
FLAG-41742 MATCHES three historical isolates. ≤ 9 SNP different. (No National Matches)
Example 3

Group A: 13 SNPs
Group B: >1,000 SNPs
Group C: >50 SNPs
Group D: >90 SNPs
Group E: >1900 SNPs
Streamlined Analyses

- Decrease de novo Analysis Time
- Decrease Database Comparison Time
- Increase Trained Analyst Numbers
Future Analytics

- AMR
- Biochemicals
- Serotyping
- Virulence Typing
- Metagenomics (?)
Benchwork Replacement

- Microbial Identification
- Biochemical Analyses
- Serology
The Florida Crew