Bacterial community structure and composition – does it correlate with pond productivity?

Sandra Infante-Villamil, Roger Huerlimann, Kelly Condon, Gregory Maes and Dean Jerry

Centre for Sustainable Tropical Fisheries and Aquaculture, James Cook University
Challenges to shrimp farming

- Diseases
  - Trade barriers
  - Global shrimp industry
  - International market prices
  - Feed costs

Wang et al., 2000
Tran et al., 2013

Introduction
Diseases

Bacteria
• Traditionally
Causative agents of disease

• Recently
Bacteria ↔ Host = Well-being
Gut microbiome plays an important role in maintaining a host’s health (Fernandez et al., 2014).

- Development of the immune system
- Protection against pathogens
- Synthesis of aa and vitamins
- Modulation of metabolism
- Caloric extraction from indigestible comp of diet
Bacterial community

Culture methods

Metagenomics

99%
Next generation Sequencing technology (NGS)

Next-Generation Sequencing of Microbial Communities in the Athabasca River and Its Tributaries in Relation to Oil Sands Mining Activities

Etienne Yergeau,a John R. Lawrence,b Sylvie Sanschagrin,a Marley J. Waiser,b Darren R. Korber,b and Charles W. Greer*a
National Research Council Canada, Montreal, Quebec, Canada; Environment Canada, Saskatoon, Saskatchewan, Canada; and University of Saskatchewan, Saskatoon, Saskatchewan, Canada

Ultradeep 16S rRNA Sequencing Analysis of Geographically Similar but Diverse Unexplored Marine Samples Reveal Varied Bacterial Community Composition

Chairmandurai Aravindraja*a, Dharmaprakash Viszwapriya*a, Shunmugiah Karutha Pandian*a
Department of Biotechnology, Alagappa University, Karaikudi, Tamil Nadu, India

Next-Generation Sequencing Reveals Significant Bacterial Diversity of Botrytized Wine

Nicholas A. Bokulicha,b, C. M. Lucy Joseph1, Greg Allen3, Andrew K. Benson4, David A. Mills1,a
1Department of Viticulture and Enology, University of California Davis, Davis, California, United States of America, 2Department of Food Science and Technology, University of California Davis, Davis, California, United States of America, 3Dolce Winery, Oakville, California, United States of America, 4Department of Food Science, University of Nebraska, Lincoln, Nebraska, United States of America
Bacterial community

Aquaculture sector
From single species detection to whole communities

1. Collect an environmental sample
2. DNA extraction from environmental sample
3. Amplify DNA markers
4. High-throughput sequencing
5. Bioinformatic processing
6. Species identification
7. Ecological analysis

Introduction
NGS in prawn culture

<table>
<thead>
<tr>
<th>Location</th>
<th>Number of samples</th>
<th>OTU</th>
</tr>
</thead>
<tbody>
<tr>
<td>Bali bay (non-polluted)</td>
<td>7</td>
<td>295</td>
</tr>
<tr>
<td>Bali aquaculture</td>
<td>10</td>
<td>269</td>
</tr>
<tr>
<td>Jakarta (polluted)</td>
<td>7</td>
<td>122</td>
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</table>

Introduction
Pond productivity

- Pond size
- Stocking date (genetic stock)
- Density of PL
- PL from same hatchery
Research question

- Are bacterial communities in the prawn (GUT and Gastric mill) and the environment (sediments and water) different in ponds with different productivity?
**Sampling design**

**Methods**

**Sampling design**

<table>
<thead>
<tr>
<th>Ponds</th>
<th>P1(HP1)</th>
<th>P2(HP2)</th>
<th>P3(LP1)</th>
<th>P4(LP2)</th>
</tr>
</thead>
<tbody>
<tr>
<td>High productive (HP)</td>
<td>Sediment</td>
<td>Intestine (GUT) and Gastric mill (GM)</td>
<td>Water</td>
<td></td>
</tr>
<tr>
<td>Low productive (LP)</td>
<td>1</td>
<td>4</td>
<td>3</td>
<td>1</td>
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**Replicate**

<table>
<thead>
<tr>
<th>Ponds</th>
<th>P1(HP1)</th>
<th>P2(HP2)</th>
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<tr>
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<th>n = 3</th>
<th>n = 9</th>
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</table>
Pond productivity (HP)

- Standardized to most productive pond as determined by yield.

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<tr>
<th>Pond</th>
<th>Yield (%)</th>
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<tbody>
<tr>
<td>HP1</td>
<td>98</td>
</tr>
<tr>
<td>HP2</td>
<td>100</td>
</tr>
<tr>
<td>LP1</td>
<td>42</td>
</tr>
<tr>
<td>LP2</td>
<td>43</td>
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</tbody>
</table>

- Pond size
- Genetic stock
- Density of PL
- PL from same hatchery
Bacterial richness (OTU)

Results
Bacterial community

![Diagram showing bacterial community distribution in Water, Sediment, Gut and GM, with labels for LP and HP](image-url)
Bacterial community

Results

GUT

GM

LP

HP
## Relative abundance

<table>
<thead>
<tr>
<th>Genus</th>
<th>High Productive</th>
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<tbody>
<tr>
<td></td>
<td>GM</td>
<td>GUT</td>
</tr>
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<td><strong>Photobacterium</strong></td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td><strong>Vibrio</strong></td>
<td>X</td>
<td>X</td>
</tr>
<tr>
<td><strong>Candidatus bacilloplasma</strong></td>
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<tr>
<td><strong>Fusibacter</strong></td>
<td></td>
<td></td>
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<tr>
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<tr>
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Relative abundance

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Pearson’s correlation

Abundance of Vibrio associated with prawn weight

Results
Insights

High productive

• Gut and GM
  $\uparrow$ *Photobacterium* and *Candidatus bacilloplasma*
  $\downarrow$ *Vibrio*

• Sediment
  $\uparrow$ *Desulfovoccus*

Low productive

• Gut and GM
  $\downarrow$ OTU richness
  $\uparrow$ *Vibrio*, *Fusibacter* and *Propionigenium*

• GUT
  $\uparrow$ *Ferrimonas*
Insights

- **Bacterial communities**
  
  Water ≠ Sediment ≠ GUT and GM (selective pressure)

  Chaiyapechara *et al.* (2012), and by Cardona *et al.* (2016) in water and GUT.

- **Vibrio**
  
  - > 50% bacterial community in GUT and GM in LP. AND
  
  - Abundance ↓ prawn weight
Insights and future

• NGS technology + bioinformatics
  ✓ Enable the understanding of the microbial diversity thriving in aquaculture ponds.

  ✓ Associations among microbial communities and prawn genotype or phenotype + environmental factors?

  ✓ Selection of potential probiotic candidates? (e.g. Candidatus bacilloplasma). Effect in survival, growth, water quality.
Acknowledgements

• Commercial partner

• JCU – Aquaculture Genetics Group
Questions?

“What do you mean ‘extreme’? We love it here!”