Using High Throughput Genomic Sequencing to Predict Ecological Impacts on Sea Turtle Populations

Lesley Anderson\textsuperscript{1,2} and Lisa Komoroske\textsuperscript{2}

\textsuperscript{1}California Polytechnic State University, San Luis Obispo
\textsuperscript{2}Southwest Fisheries Science Center, National Marine Fisheries Service, NOAA
Sea Turtle Life History

• Unique life history makes sea turtles difficult to study
Threats to Sea Turtle Populations

- Threats include:
  - Habitat loss
  - Overexploitation
  - Global warming
  - Disease
- All marine turtles in U.S. waters are protected
Research Goals

1. Impacts of pollution on sea turtle health
2. Identify SNPs to be used as biomarkers
   - Sex
   - Size
   - Migration
   - Foraging
High Throughput Genomic Sequencing

1. Field Sampling
2. RNA Extraction
3. Quality Control
4. Library Sequencing
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Field Sampling

- Capture turtles in the field
- Paxgene whole blood samples

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High Throughput Genomic Sequencing

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RNA Extraction

- Optimization of SOP
- Phenol-chloroform extraction method
RNA Extraction

• RNA is unstable
  – Temperature sensitive
  – Time sensitive
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Quality Control

- Nanodrop Spectrophotometry
  - Assessment of nucleic acid purity

Potential Protein Contamination
Potential Reagent Contamination
Quality Control

- Gel Electrophoresis
  - Agarose gel
  - Lighter fragments travel further
  - Bands represent concentrations of base pairs
Quality Control

- Gel Electrophoresis
  - Causes of degradation?

Ladder

Turtle 21
Hawaiian Sea Turtle Population

• Spring 2015: 11 turtles captured
  – 3 with known tumors (likely Fibropapillomatosis)
• Gel indicates one turtle with potential RNA degradation
  – Significance of degraded RNA in diseased turtles?
High Throughput Genomic Sequencing

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Library Sequencing

• High throughput sequencing

1. mRNA Isolation

2. Illumina Sequencing

3. Align Sequences against Genome

4. Generate Sequence Counts for all Genes in Genome

Gene A: $\frac{30}{10} = 3$ fold change
Gene B: $\frac{10}{5} = 2$ fold change
Potential Implications

1. Impacts of pollution on sea turtle health
   – Help conservationists and ecologists predict potential threats to habitat

2. Identify SNPs to be used as biomarkers
   – Provide useful genomic data to other researchers to support their studies
Acknowledgements

• Lisa Komoroske, SWFSC, NMFS-NOAA
• Special thanks to:
  – Camryn Allen
  – Gabriela Serra-Valente
  – Joel Schumacher
  – Billy Hilton

"This material is based upon work supported by the National Science Foundation through the Robert Noyce Teacher Scholarship Program under Grant No. (grantee must enter NSF grant number). Any opinions, findings, and conclusions or recommendations expressed in this material are those of the author(s) and do not necessarily reflect the views of the National Science Foundation."
Literature Cited

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Questions?