RECLAMATION Managing Water in the West

## Sequencing the quagga mussel genome

A resource for developing biocontrols

Yale Passamaneck Bureau of Reclamation

## Mussel Impacts

- Ecological
- Economic
- Operational
- Recreational







## Control and eradication

Physical and chemical treatments cannot be economically scaled for large reservoirs



UV treatment – Parker Dam



Potash - Millbrook Quarry, VA

#### Genetic Biocontrols

#### **CRISPR-Cas9**

- Targeted changes to genome
- Adaptable to novel organisms



**UT** Austin

## Genetic Biocontrols

#### **Gene drive**

- Propagation of deleterious changes
- Potential to eradicate a population



Esvelt & Gemmell 2017

## Need for genome sequencing

- Identify genes of interest
- Determine gene structure and copy number
- Characterize variation
- Target site uniqueness



#### Quagga Mussel Genome

• Estimate 1.6 Gbp size



- PacBio Sequel long but noisy
- Illumina HiSeq short but accurate
- 100x coverage for each

#### Quagga Mussel Genome

- Collection of adults from Davis Dam
- DNA extraction from males
- Both PacBio and Illumina sequencing from a single individual





#### Genome Assembly

Canu – long reads only

N50 = 740,653

Assembly size = 2.7 Gbp (!?)



- MaSuRCA hybrid of long and short reads Analysis in process
- RNASeq for gene prediction in process
- Collaboration with Dr. Kevin Kocot, U Alabama

#### Transmissible Disseminated Neoplasia

- Collaboration with BiomiLab
- Cancerous hemocytes infect neighboring bivalves
- Can cause high mortality
- Described from multiple species
- Inducible in the lab?



#### p53 – the (anti)cancer gene

- Wild-type forms have anti-cancer roles
- Mutant forms linked to many human cancers
- *p53* associated with bivalve neoplasia
- Homolog present in quagga genome, with conservation at key sites
- Associated genes (*mdm2*, *p21*) also in the genome



Surf clam p53 protein structure Holbrook et al 2009

# *TERT* – Cell imortality

- Telomeres at the end of chromosomes
- Protect chromosome form degradation
- Shortening of telomeres can lead to cell death
- TERT maintains telomere length





Telomeres at tips of chromosomes

NIH

## Changes to sex ratios or fertility

- Make all transgenic offspring male
- As the proportion of males increases, the population decreases
- Potential eradication



James et al 2018

# Sex determination in dreissenids

- No sex chromosomes (like other bivalves)
- Rare hermaphrodites in zebra mussels
- Genetic and environmental influence?
- Mechanism is unknown
- Sequencing female quagga for comparison



Zebra mussel karyotype Boron et al 2004

#### Do mussels get DUIs?

- Doubly uniparental inheritance (DUI) of mitochondrial DNA
- Unique to bivalves and widespread
- Not present in zebra or quagga mussels?



Breton et al 2014



Conserved sex determination / differentiation genes

- Males: SoxH, Dsx, GATA4, WT1
- Females: FoxL2, WNT4, ß-catenin
- Conserved roles in sex determination?
- Many of these genes have multiple roles in development



Zhang et al 2014

# Challenges for genetic manipulation of mussels

- Culturing mussels
- Transgenesis
- Identification of promoters to drive expression
- Cryopreservation





# Future considerations for genetic biocontrol

- Efficacy
- Genetic resistance
- Safety & environmental impacts
- Risk assessment
- Regulatory approvals
- Bioethics



#### Yale Passamaneck ypassamaneck@usbr.gov 303-445-2480

