Why is there a difference in sex ratio between hatchery and wild steelhead?

Neil Thompson¹, Kassi Cole², Laura McMahon¹, Melanie Marine¹ and Michael Blouin¹

1 Department of Zoology, Oregon State University
2 Department of Zoology, University of Hawaii
Hood River steelhead

[Map showing the Hood River in Washington and Oregon]

[Hood River image]

[Steelhead fish image]
Hood River Sex Ratio

- Winter run
- Integrated broodstock program

<table>
<thead>
<tr>
<th>Brood Year</th>
<th>Hatchery Sex Ratio</th>
<th>Wild Sex Ratio</th>
</tr>
</thead>
<tbody>
<tr>
<td>1992</td>
<td>54.2</td>
<td>59.7</td>
</tr>
<tr>
<td>1993</td>
<td>43.8</td>
<td>59.9</td>
</tr>
<tr>
<td>1994</td>
<td>50.2</td>
<td>63.7</td>
</tr>
<tr>
<td>1995</td>
<td>53.0</td>
<td>67.9</td>
</tr>
<tr>
<td>1996</td>
<td>55.3</td>
<td>64.7</td>
</tr>
<tr>
<td>1997</td>
<td>53.0</td>
<td>60.6</td>
</tr>
<tr>
<td>1998</td>
<td>44.8</td>
<td>62.8</td>
</tr>
<tr>
<td>1999</td>
<td>47.3</td>
<td>60.0</td>
</tr>
<tr>
<td>2000</td>
<td>53.1</td>
<td>62.6</td>
</tr>
<tr>
<td>2001</td>
<td>51.9</td>
<td>58.9</td>
</tr>
<tr>
<td>2002</td>
<td>52.6</td>
<td>69.3</td>
</tr>
<tr>
<td>2003</td>
<td>48.4</td>
<td>63.0</td>
</tr>
<tr>
<td>2004</td>
<td>54.8</td>
<td>64.3</td>
</tr>
</tbody>
</table>

Mean SR: 50.9 (Hatchery), 62.9 (Wild)

*Sex ratio measured as % female*
Hood River Sex Ratio

• Wild population: ALL 13 years female biased
  Range: 58.9 – 69.3
• Hatchery population: 9 years female biased
  Range: 43.8 – 55.3

Difference in proportions Z-test, P <<0.001

Question: Why is there a difference in sex ratio between wild and hatchery populations?
Potential explanations

1. Sex reversal of chromosomally XX females into phenotypic males
2. Selection against hatchery females
3. Selection against wild males
4. Lower rates of residency in male hatchery fish
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1. Sex reversal of chromosomally XX females into phenotypic males
2. Selection against hatchery females
3. Selection against wild males
4. Lower rates of residency in male hatchery fish
Sex determination in *O. mykiss*

- Y chromosome = male
- No Y chromosome = female
Mechanism of Sex Reversal

• Aromatase enzyme
  – Converts testosterone to estrogen
• High temperature inhibits aromatase
Mechanism of Sex Reversal

- Aromatase enzyme
  - Converts testosterone to estrogen
- High temperature inhibits aromatase

XX females develop testes
Why Aromatase?

- Spawned Hood River Basin
- Reared at Oak Springs on Deschutes R.
- Deschutes warmer than Hood River
Research question

(1) Does the chromosomal sex match the phenotypic sex in Hood River steelhead?
   - % non-matching?
   - direction?
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   - % non-matching?
   - direction?

(2) Does the sex ratio in the hatchery population change from juvenile release to adult return?
Research question

(1) Does the chromosomal sex match the phenotypic sex in Hood River steelhead?
   – % non-matching?
   – direction?

(2) Does the sex ratio in the hatchery population change from juvenile release to adult return?

(3) Is the sex ratio of out-migrating wild smolts 50:50?
Methods – hatchery juveniles

- Hood River Production Program
- Sampled at 1 year of age
- Fin clip & floy tagged
- Bodies preserved in formalin
- Gonad inspected by stereomicroscopy
- Fish type, family, spawn date
Methods – wild juveniles

• Run year 2006
• 150 juveniles
• >= 140 millimeters fork length
• PIT tag
• Winter run offspring
Methods - Adults

- Adults sampled at Powerdale dam (1991-2009)
- Gender, origin, fin clip taken by ODFW
- Random 96 individuals per sex
- Total 196 adult samples
- Wild and Hatchery origin
Phenotypic sex

• Adults:
  – ODFW staff visually sexed fish @ Powerdale
Chromosomal sex

- OmyY1 marker - sex marker (Brunelli et al. 2008)
- 4% non-concordance rate
- Male band or female band
- Positive reaction band
Research question

(1) Does the chromosomal sex match the phenotypic sex?
   - % non-matching?

- Mismatch = putative sex reversal
- Test for non 50:50 sex ratio
  - Chi-square test
Results – hatchery juveniles

- 173 successful reactions
- 6 fish mismatched (3.5 % non-concordance)
- Male-to-female and female-to-male mismatch

<table>
<thead>
<tr>
<th>Juvenile ID</th>
<th>Spawn Date</th>
<th>Family ID</th>
<th>Fish Type</th>
</tr>
</thead>
<tbody>
<tr>
<td>2</td>
<td>4/20/2010</td>
<td>2821x2796</td>
<td>HH</td>
</tr>
<tr>
<td>3</td>
<td>5/24/2010</td>
<td>382x477</td>
<td>WW</td>
</tr>
<tr>
<td>29</td>
<td>5/10/2010</td>
<td>2764x2783</td>
<td>HH</td>
</tr>
<tr>
<td>44</td>
<td>5/10/2010</td>
<td>2812x2761</td>
<td>HH</td>
</tr>
<tr>
<td>62</td>
<td>5/17/2010</td>
<td>458x2804</td>
<td>WH</td>
</tr>
<tr>
<td>223</td>
<td>5/24/2010</td>
<td>2774x383</td>
<td>HW</td>
</tr>
</tbody>
</table>
Results - Adults

- 175 successful reactions
- 3 fish mismatched (1.7 % non-concordance)
- Male-to-female and female-to-male mismatch

<table>
<thead>
<tr>
<th>Run</th>
<th>Chromosomal Gender</th>
<th>% Non-concordance</th>
</tr>
</thead>
<tbody>
<tr>
<td>Winter</td>
<td>Male</td>
<td>2.27</td>
</tr>
<tr>
<td></td>
<td>Female</td>
<td>1.15</td>
</tr>
</tbody>
</table>
Sex Reversal?

• Juveniles
  – No evidence for sex reversal
  – Mismatches in both directions
  – Non-concordance rate below published rate
  – Mismatches from 6 different families, 4 spawn dates and 4 fish types
Sex Reversal?

- Adults
  - No evidence for sex reversal
  - Mismatches in both directions
  - Rate not above published non-concordance rate
Potential explanations

1. Sex reversal of chromosomally XX females into phenotypic males → NOT happening

2. Selection against hatchery females, in captivity or at sea
Juvenile sex ratio at release

<table>
<thead>
<tr>
<th></th>
<th># Male</th>
<th># Female</th>
</tr>
</thead>
<tbody>
<tr>
<td>Chromosomal</td>
<td>85</td>
<td>88</td>
</tr>
<tr>
<td>Phenotypic</td>
<td>87</td>
<td>86</td>
</tr>
</tbody>
</table>

*Not different from 50:50 ratio*
Chi-square p-values of 0.82 & 0.94

50:50 at release and at return to the dam
Potential explanations

1. Sex reversal of chromosomally XX females into phenotypic males → NOT happening

2. Selection against hatchery females, in captivity or at sea → NOT happening

3. Selection against wild males at sea
Wild smolt sex ratio

• 127 successful reactions
  – 64% female
  – 82 females, 45 males
• Ages 1-4
• 2 and 3 year olds – similar sex ratios
Potential explanations

1. Sex reversal of chromosomally XX females into phenotypic males \( \rightarrow \) NOT happening

2. Selection against hatchery females, in captivity or at sea \( \rightarrow \) NOT happening

3. Selection against wild males at sea \( \rightarrow \) NOT happening
Potential explanations

1. Sex reversal of chromosomally XX females into phenotypic males → NOT happening

2. Selection against hatchery females, in captivity or at sea → NOT happening

3. Selection against wild males at sea → NOT happening

Selection in freshwater? Rundio et al. 2012 → NOT likely
Potential explanations

4. Lower rates of residency in male hatchery fish

Christie et al. 2011
Potential explanations

1. Sex reversal of chromosomally XX females into phenotypic males → NOT happening

2. Selection against hatchery females, in captivity or at sea → NOT happening

3. Selection against wild males at sea → NOT happening

4. Higher residency in wild males than hatchery males → Most plausible explanation
Thank You!

- Jim Gidley & Albert Santos
  - Parkdale fish facility
- Lyle Curtis & staff
  - Oak Springs hatchery
- Blouin lab
Other research projects

• Noakes *et al.*. Temperature and Steelhead Sex: a threat from climate change?

• Thompson *et al.*. Sex biased survival in wild Alsea and Nehalem River smolts?