

Sex reversal in hatchery steelhead



Neil F. Thompson
Department of Zoology
Oregon State University

Presentation Outline

- Mechanisms of sex change
- Hood River Steelhead
- Study question
- Methods
- Results
- Summary

Changing gender in hatchery fish

- Addition of sex hormone

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Functional Masculinization of Genetic Females in Rainbow Trout

Houji OKADA^{*1}, Haruyoshi MATUMOTO^{*1}, and Fumio YAMAZAKI^{*2}

(Received October 23, 1978)

The present study was carried out to accomplish functional sex reversal by oral treatment of methyltestosterone to get XX male which sire all female progenies. The fry were fed with diet supplemented with methyltestosterone at the concentration of 1 ppm, 5 ppm and 10 ppm for 58 days starting from the swim up stage, after which they were fed normal diet. No female having normal ovaries was observed at the end of the treatment. On the other hand, percentage of males ranged from 60-88 depending on the concentration of the hormone at the termination of the experiment. High mortality was recorded during the hormone treatment.

Twenty fish of 1 ppm group were reared to check the sex ratio of F₁ generation. Eleven males and 7 females survived to maturity and were fertilized individually. In progeny test, 2 males produced nearly all female progenies and the others produced 1:1 sex ratios. This fact indicates that these two were XX males functionally sex reversed from genetic females.

All produced females were normal in reproductive function and produced 1:1 sex ratio in F₂ generation. The present data strongly suggest that rainbow trout is female homogamety (XX) and functionally sex reversed XX male produces all female progenies with normal reproductive function.

Changing gender in hatchery fish

- Environmental toxins/chemicals
- Elevated or depressed temperature
 - Testosterone and Estradiol --> Aromatase Enzyme



More males



Hood River steelhead

- Spawned at Parkdale fish facility over approximately 3 months (April-June)
- Transferred to Oak Springs hatchery for rearing
- Oak Springs hatchery varies water temperature to hatch all eggs synchronously (36-53 degrees F)
- Hood River – cold and flashy
 - Snowmelt begins in April



Study question

- Does the genetic sex of hatchery steelhead match their phenotypic sex at time of release from the hatchery?

If reversal happening:

male skewed sex ratio

genetic females phenotyped as males

Methods

- Fish spawned in late spring 2009
- Pure hatchery crosses and pure wild crosses
- Juvenile stage in early August (exogenous feeding)
- Transferred out of hatch house to a circular tank in Oct.
- Underwent typical hatchery culture for entire lifespan



Methods

- Fish spawned in April and May 2009
- Pure hatchery crosses and pure wild crosses
- Juvenile stage in early August (exogenous feeding)
- Transferred out of hatch house to a circular tank in Oct.
- Underwent typical hatchery culture for entire lifespan
- **Design mimicked production culture as close as possible**
 - Lower density than production
 - Poned outside at a later date

Methods

- End of May 2010
 - Measured (fork length)
 - Fin clip for genetic analysis
 - Floy tagged
 - Kept on ice and brought to Corvallis
- At OSU fish were given a phenotypic score (1-4) and phenotypically sexed
 - Anterior gonad



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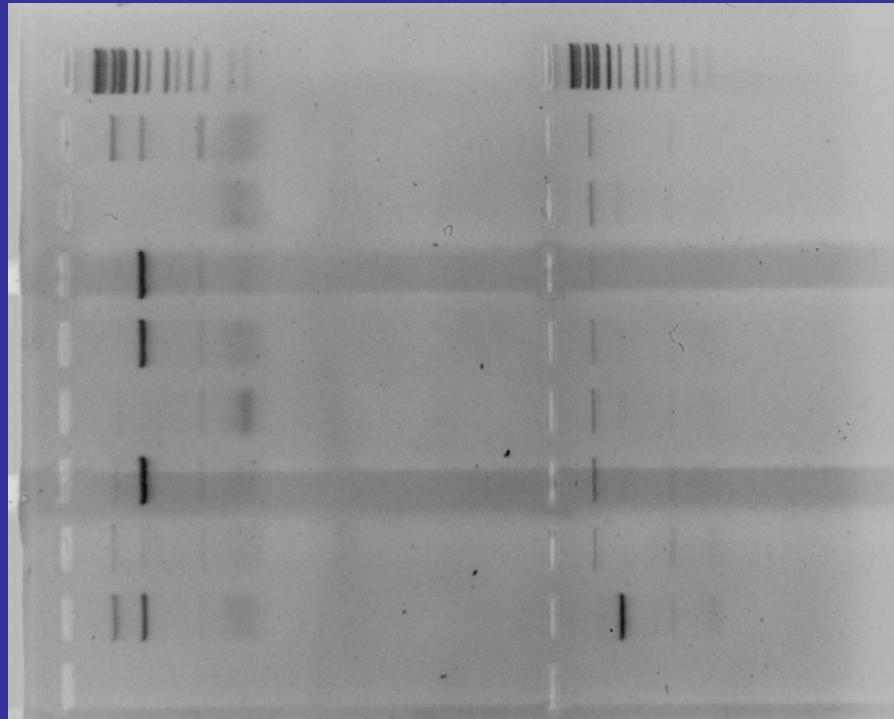
Phenotype Key

- | | |
|---|--|
| 1 | Smolt, Strong silver coloration |
| 2 | Transitioning to smolt, predominately silver with some pink striping |
| 3 | Strong pink stripe, dark vertical bands, some spotting |
| 4 | Rainbow trout, dark body with vibrant pink stripe, blue banding and heavy spotting |



Methods

- Sub-sample of total population (96 fish)
- DNA extracted and PCR'd with Omy Y1 primer
- Run on agarose gel and scored by eye



Results – Preliminary!

49 Wild 42 Hatchery 5 TBD

Phenotype	% reversed
1	30 (3/10)
2	50 (7/14)
3	7 (3/41)
4	22.5 (7/31)

ALL genetic males
reversed to phenotypic
females

Phenotype Key

- 1 Smolt, Strong silver coloration
Transitioning to smolt, predominately silver with
- 2 some pink striping
- 3 Strong pink stripe, dark vertical bands, some spotting
Rainbow trout, dark body with vibrant pink stripe.
- 4 blue banding and heavy spotting

Results – Preliminary!

Phenotype	% reversed	# H or W of reversed	# of families
1	30 (3/10)	2 Wild 1 Hatchery	3 W 5 H
2	50 (7/14)	6 Wild 0 Hatchery 1 TBD	5 W 2 H
3	7 (3/41)	1 Wild 2 Hatchery	14 W 13 H
4	22.5 (7/31)	3 Wild 3 Hatchery 1 TBD	12 W 6 H

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Phenotype	% of Population
1	11
2	15
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Is there a family affect of what phenotype juveniles express?

Coming soon!

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Results – Preliminary!

**Phenotypic
Sex Ratio F:M**

62:38

**Genetic
Sex Ratio F:M**

40:60

1 Mature Female
164 Immature Females
76 Immature Males
25 Mature Males



25% of males mature!



Results – Preliminary!

Phenotypic Sex Ratio F:M

62:38

1 Mature Female
164 Immature Females
76 Immature Males
25 Mature Males

Genetic Sex Ratio F:M

40:60

Harder to explain.

33% of males mature!

Carl Schreck and Co.
found a 10% error rate in
sexing 1 year old trout.

Summary

- 20 individuals sex reversed (approx 20%)
 - All genetic males to phenotypic females
 - No obvious hatchery/wild or family affect
- Sex ratios slightly skewed
 - High % of mature males

Future Work

- Genetic sex of 96 more individuals
 - Complete sample of smolt and pink stripe phenotypes
- Look for family or hatchery/wild affect of sex reversal
- Look for family or hatchery/wild affect of phenotype
- Compare sex ratios to those seen in the wild (at Hood River dam) and at swim up in hatchery

Thank you!

- Jim Gidley and staff at Parkdale fish facility
- Lyle Curtis and Oak Springs hatchery staff
- Blouin lab members

Questions?