Effects of Single Male Matings and Multiple Male Matings on Egg Survival in Erwin Strain Rainbow Trout

December 1993

Wes Orr, Bernie Shrable, Dan Brown
Ennis National Fish Hatchery, Montana

Introduction:

At Ennis NFH, MT paired matings (one female x one male) of rainbow trout to create future broodstocks have historically exhibited an 8 to 10 percent reduction in survival to the eyed stage as compared to multiple male matings. The purpose of this investigation was to determine what causes this problem.

Methods:

Milt was collected from 20 Erwin strain males into 20 individual test tubes. One ml of milt was removed from each tube and pooled into a separate container. To ensure viability any milt that was thick or watery or abnormal in appearance was discarded. The eggs from 20 Erwin strain females were air spawned into individual pans containing equal volumes of 0.75% saline solution. Only eggs that appeared viable were used in the test. The eggs from each container were equally divided into 2 containers, making a total of 40 containers, 2 from each female.

One container of eggs from each female was fertilized with 1 ml of precollected milt from 1 male. The other container of eggs from each female was fertilized with 1 ml of milt from the container of pooled milt representing 20 males.

After fertilization, eggs from each of the 40 containers were rinsed, and then water hardened in a 75 mg/l iodophor solution for 30 minutes. After water hardening, one-half of the eggs from each container were individually incubated in 1 liter upwelling jars. The remaining eggs in the 20 - 1 female x 1 male containers were pooled, mixed and then split into 4 replicates and incubated in 1 liter upwelling jars. Eggs remaining in each of the 20 - 1 female x 20 male matings were also pooled and divided into 4 - 1 liter upwelling incubators. All incubators were treated daily with 1200 mg/L of formalin for 15 minutes to prevent fungus. All eggs were mechanically shocked on day 14.

Each batch of eggs was picked mechanically, and an electronic egg picker was used to count normal eyed eggs, white eggs, dim eyed eggs, and blank eggs. A dim eyed egg is defined as an eyed egg which has very small eyes and dim eye pigmentation. A blank egg is defined as any post shock translucent egg with no visible cellular development.

Results:
In this test it appears that reduced eyeup can be attributed to a few individuals in the population. We can also surmise that even experienced spawn takers cannot always tell whether gametes are viable or not.

Statistically the study was broken into 2 parts; The first dealing with 20 single male treatments and 20 pooled male treatments, and the second part dealing with 4 replicates of single male and 4 replicates of pooled male treatments. In the first part there was a slight effect of female and male on percent eyeup, probably due to the wide variation between treatments. Also, there was a strong effect of female but no effect of male on blank eggs, and no effect of male or female on dim eyed eggs. In the second part of the study the effect of female was designed out and statistics confirmed there was a strong effect of male on the percent survival to the eyed stage.

Discussion:
If spawntakers could ascertain 100% of the time which eggs or sperm were not viable, the risk of losing genetic material would be greatly reduced. The problem of an occasional bad male can be overcome by using 2 males to fertilize one females eggs. If there are not enough males in the population, pool 2 males to fertilize the eggs of 2 females. Similarly, mating 2 females with 2 males insures that the gametes from 2 males is not lost because of 1 bad female. If future broodstock are derived from a parent population of several hundred fish, losing genetic material from a few individuals may not be important. However, when dealing with small populations, which is usually the case with threatened or endangered species, the loss of genetic material from a few individuals becomes paramount.

TABLE I

POOLED MILT
VS
SINGLE MALE MATINGS

Percent Percent Number
Number
Eyeup Eyeup Dim Eyed
Blank
Female Single Pooled Single Pooled
Single Pooled No. Male Males Male
Males Male Males

1	5.4	5.4	1	12	819	811
2	95.4	97.6	7	1	0	2
3	96.4	97.2	2	1	2	1
4	97.3	96.3	3	0	0	4
5	78.5	78.1	0	5	1	15
6	98.6	97.4	1	0	1	1
7	93.1	96.9	6	1	40	3
8	85.8	80.9	1	2	7	10
9	94.8	92.9	1	1	15	11
10	96.9	51.6	0	0	6	3
11	95.7	95.5	0	0	5	2
12	94.8	93.9	1	3	2	5
13	45.9	43.2	0	0	230	50
14	89.8	93.1	3	2	39	26
15	99.0	98.0	0	1	1	7
16	78.1	80.1	5	2	11	18
17	11.1	96.6	0	1	1	1
18	1.2	69.9	0	0	0	80
19	90.5	92.6	7	26	8	2
20	70.4	76.2	0	6	215	202
	AVERAGE	75.9	81.7			

TABLE 2

POOLED MILT
VS
SINGLE MALE MATINGS

SINGLE MALE MATINGS	PERCENT EYEUP	PERCENT DIM EYED	PERCENT BLANK
R1	67.8	2	7
R2	75.7	3	10
R3	74.6	2	12
R4	72.9	3	12
AVERAGE	72.8		

POOLED MALE MATINGS	PERCENT EYEUP	PERCENT DIM EYED	PERCENT BLANK
R1-P	83.2	2	7
R2-P	81.7	3	8
R3-P	82.9	1	7
R4-P	80.2	2	8
AVERAGE	82.0		