GREAT LAKES FISHERY COMMISSION

Project Completion Report[1]

Short-term Sterile Male Release Evaluation, 1995

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A Completion Report Submitted to the Great Lakes Fishery Commission

March 31, 1995

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Although the Commission is committed to a long-term field trial evaluating sterile male release in Lake Superior and the St. Marys River, the program is experimental and verification of the short-term success of our operational methods is needed. Previous research (Hanson and Manion 1978 and 1980) showed that, in the context of a study pairing sterile males with unsterilized controls from the same source, sterile males decrease production of viable eggs in proportion to the number of sterile males released. We needed to examine whether the operational techniques presently employed to sterilize and distribute about 20,000 animals to Lake Superior and 5,000 to the St. Marys River are achieving the same results.

Long-term effectiveness of the sterile-male-release technique in Lake Superior tributaries and on the St. Marys River, Lake Huron is to be evaluated through measurement of wounding rates on fish and the abundance of fish and adult sea lampreys (Hanson et al. 1990). Unfortunately, at least 6 to 10 years (one lamprey generation) will be required to realize any changes in these measures. We needed to measure effectiveness in the shorter-term, however, to ensure that our current methods are achieving results consistent with results of the published studies used to plan the program. The objectives of the short-term evaluations of sterile male release conducted in 1992-95 were to (1) determine if the ratios of sterile to untreated males observed on nests in selected streams are consistent with the predicted ratios based on estimated run size and numbers of sterile males released and (2) determine if reductions in the proportion of viable eggs are consistent with expected reductions based on the observed ratios of sterile to untreated males in each stream. (3) determine if competitiveness of sterile males changes with the ratio of sterile to untreated males.

Methods

The proposed procedures for the study (Bergstedt et al. 1994) were submitted for BOTE review on January 21, 1994 and approved on May 2, 1994. Amendments to the plan for the 1995 studies were submitted by letter on January 1, 1995 and approved on March 6, 1995. In brief, the study consisted of three components on each stream:

1. Estimating the number of native or "untreated" males (on the Misery and Rock Rivers, known numbers were placed above a barrier to upstream migration). These estimates would be used, in combination with the number of sterilized males released, to calculate expected ratios of sterile to untreated males on the nests.

2. Determining the actual ratio of sterile to untreated males on the nests. This ratio was t be determined from field observations on each stream following the release of sterile males. Any male building or occupying a nest was considered to be a nesting male and potentially competing for females.

3. Sampling nests with male parents in three classes ("untreated," "sterile," and "unobserved") to determine the mean proportions of viable eggs produced by untreated males, sterile males, and the combined population of males. The percent viability in nests with untreated males was intended to represent the likely success rate if no sterile males were present and the percent viability in nests where a male was not observed was intended to represent the overall success rate (including participation by sterile males).

Study Streams--Desired stream characteristics included: (1) high hatch success, (2) observable spawning areas, (3) spawning over a wide enough area that multiple usage of single nests is infrequent, and (4) a sufficient number of spawners. Lake Superior tributaries where studies were conducted in 1995 were the Wolf River (Ontario), the Misery River (Michigan), and the Rock River (Michigan). The St. Marys River (the outflow from Lake Superior to Lake Huron) was also selected as a study stream.

Planned analyses and comparisons

1. Comparison of the observed and expected ratios of nesting males with Chi-square. The expected ratio was to be based on the number of sterile males released and on mark-recapture estimates of the resident population. This tests the null hypothesis that the occurrence of sterile males on nests is in proportion to their presence in the stream population of males.

2. Comparison of the estimated percent viability of eggs in nests with untreated and unobserved male parents. This tests the null hypothesis that release of sterile males does not result in a reduction in egg viability.

3. Examining whether varying the ratio of sterile to untreated males produced a linear effect on the observed ratios on the nests. This tests the null hypothesis that there is a linear effect of varying the ratio of sterile to untreated males on the observed ratios.

Results

Observed versus Expected Ratios--Studies were conducted in two classes of streams--barrier streams where known populations were created above the barrier (Rock and Misery rivers) and streams without barriers where we had to rely on mark recapture estimates of the natural run for population information (Wolf and St. Marys rivers). We made reasonable numbers of observations of male lampreys on nests on all four streams. The fewest was on the St. Marys, where we observed 62. That was followed by the Rock River with 75, the Wolf River with 79, and the Misery River with 278.

In two barrier streams where we worked with known populations, the observed and expected ratios were significantly different but contradictory (Table 1). On the Misery River we observed more sterile males than expected and on the Rock River we observed less. In streams where we had to estimate the resident populations, we were successful on the St. Marys River but not on the Wolf River. On the St. Marys River, our estimate of the number of resident males was based on over 200 recoveries. Using that population estimate, the expected and observed ratios in the St. Marys were not significantly different (Table 1). The Wolf River had a very small run again in 1995, and recaptures of marked animals were not sufficient (two recaptures) for us to estimate the number of resident males. We do not plan on including this class of data from the Wolf River in subsequent analyses.

Table 1.--Estimated numbers of resident (untreated) males, numbers of sterile males released, and numbers of males observed on nests in four study streams during 1995.

	Misery	Rock	Wolf	St. Marys
Sterile males released	957	962	1195	4238
Number of resident males	196	389	-	5719
Sterile males on nests	256	41	74	27
Resident males on nests	22	34	5	36
Expected sterile:untreated ratio	4.9:1	2.5:1	-	0.74:1
Observed sterile:untreated ratio	11.6:1	1.2:1	14.8:1	0.75:1
Chi-square	16.3	10.0	-	0.002
	**	**	-	NS

<u>Relation of Expected to Observed Ratios</u>--Our results continue to support the conclusion that sterilized males generally appear on the nests in proportion to their relative abundance in the population of males. We now have eight cases where we have both an estimate of the ratio of sterile to untreated males on nests and an estimate of the numbers of both sterile and resident males in the stream to calculate the expected ratio

(Figure 1). For three of those cases, the St. Marys River in 1992 and the Misery and Rock rivers in 1995, the observed and expected ratios were significantly different--in the remaining five, the differences were not significant.

These data allowed us to examine whether there was a linear effect of the ratio of sterile to untreated males introduced on the ratio observed on the nests. Regression of the $\log_{10}$ of the observed ratio on the $\log_{10}$ of the expected ratio indicated a significant relation between the observed and expected ratios ($P = 0.0004$). The regression was consistent with the hypothesis of observed ratios being proportional to the expected ratios. The intercept was not significantly different than zero ($P = 0.66$), and the slope (0.83) was not significantly different than one ($P = 0.21$). Our results indicate that, on average, we can expect sterilized males that we introduce into streams to appear on the spawning grounds and construct nests.

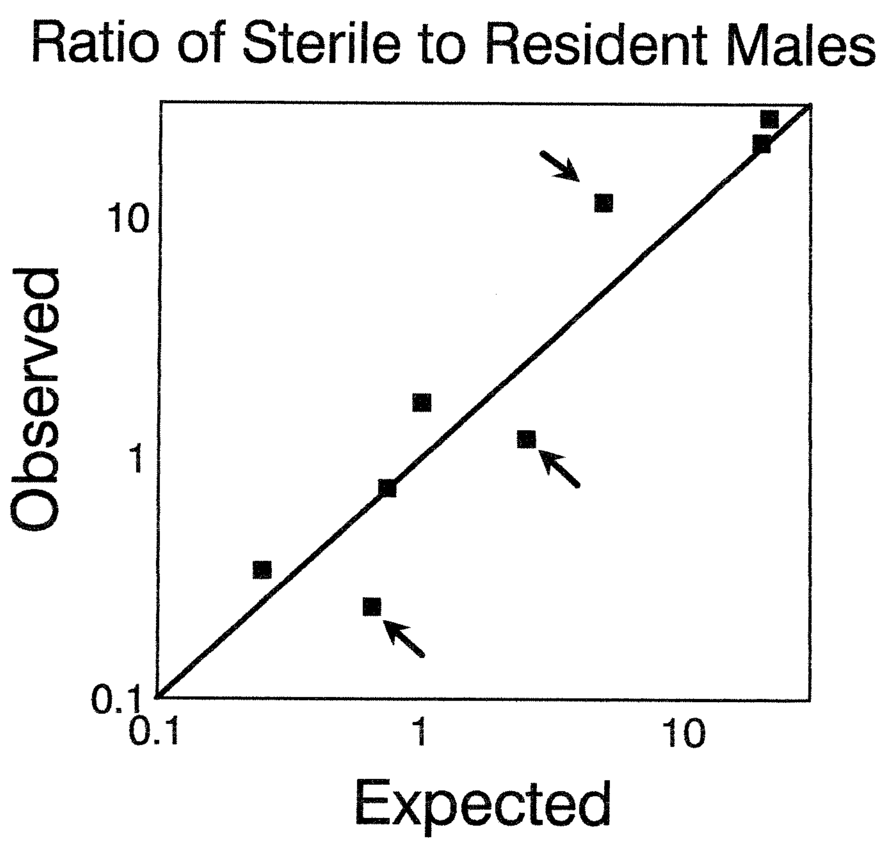

Figure 1.--Observed versus expected ratios of sterile:resident male sea lampreys observed on nests, 1992-95. The diagonal line indicates equal observed and expected ratios. Arrows denote observed ratios significantly different from expected.

Percent Egg Viability--It is also necessary to determine if sterile males attract and spawn with females, resulting in a decrease in viable prolarvae. Nests on which only an untreated or a sterilized male parent was observed, or on which no males were observed, were excavated and the percent viable eggs in each class was determined (Figures 2-5). The plan was to use the percent viable eggs in nests where only untreated males were observed to provide a baseline observation for typical nest success in that stream (had sterilized males not been released). The percent viable eggs in nests where males were not observed were taken as a measure of overall success with both untreated and sterilized males involved. If sterilized males were competitive in attracting females to their nests and spawning, then percent viability should be significantly reduced in nests where males were not observed compared to nests with untreated male parents.

Reductions in egg viability due to sterile male release were more difficult to demonstrate in 1995. Unfortunately, we could only evaluate reductions in egg viability in the Rock and the St. Marys rivers. In the Rock River, the distribution of egg viability for unobserved nests (as an indicator of overall success) seemed slightly more skewed in favor of low success compared to nests with untreated male parents (Figure 2), but the difference was not significant based on a Kolmogorov-Smirnov test (*P* near 1.0). The mean egg viability (medians in parentheses) were nearly identical at 10.7% (10.3%) for the nests with untreated male parents and 10.3% (7.7%) for nest where the male parent was unobserved. In the St. Marys River, the distribution of egg viability for unobserved nests was strongly skewed in favor of low success (Figure 3), when compared to nests where untreated male parents were observed. Again, the difference was not significant based on a Kolmogorov-Smirnov test ($P = 0.07$). The means (medians in parentheses) in this case were 49.3% (49.9%) for the untreated and 32.3% (22.3%) for the unobserved categories. While the lack of apparent effect in the Rock river is cause for concern, the observed decrease in the St. Marys is near the amount expected based on trap efficiency and it seems likely the non-significant result is due simply to low sample size. Analyses are not possible in the Misery (Figure 4) and Wolf (Figure 5) rivers, because of missing data.

Summary

Where we collected sufficient data during 1992-95 to make comparisons, the results were generally encouraging. Although there were shortcomings in producing complete data sets on all streams, our findings were supportive of the conclusions drawn by Hanson and Manion (1980). Sterilized males appeared to reach the spawning grounds and construct nests in the predicted numbers and the observed ratios appeared to be proportional to the expected ratios.

Overall, there is also evidence that sterile males mate with females and reduce the percentage of viable embryos at hatch. If all observations are simply combined, the data collected during 1992-95 suggest that females were attracted to nests with sterile males and that there was a demonstrable reduction in viable eggs (Figure 6) . This decrease was most likely due to participation by sterilized males in mating on the unobserved nests. The observed shift probably underestimates the size of the effect, making this a conservative finding. The underestimation is due to the distribution of percent egg viability in nests with untreated male parents (as the measure of baseline conditions) probably being shifted to the left by undocumented participation of sterilized males. There are, however, difficulties in combining data in this way, and Figure 6 is offered in this report only as a preliminary observation. Similarly, the data were combined for the three years where we sampled nests in the St. Marys River, 1993-95 (Figure 7). For the St. Marys River, the observed distributions of percent viable eggs in the untreated (top panel) and unobserved (gray bars in bottom panel) classes were also significantly different (Kolmogorov-Smirnov test, $P = 0.03$).

If we accept the distributions of egg viability in the untreated and sterile classes, we can also calculate a predicted distribution in the unobserved nests given a ratio of sterile to untreated males (Figures 6 and 7, black bars in lower panel). The ratio was selected by minimizing Chi-square comparing the observed and expected. This is not an exact calculation because we know there is at least some unobserved spawning by males of other classes in these nests. Nonetheless, the predicted distributions are close to the observed distributions, although still significantly different. In the St. Marys River, the ratio (0.58:1) is also close to the average ratio (0.66:1) used there 1993-95. Because of the extreme ratios applied to some streams, a similar comparison with the average ratio is not reasonable for the combined data.

Consistently accomplishing <u>all</u> the tasks needed to measure an effect of sterile male release in the field has proved difficult. However, the collective results from the work by Lee Hanson in the 1970s and the work conducted during 1992-95 provide reasonable evidence of the short-term effectiveness of sterile male release. We therefore have proposed studies to be conducted during 1996-99 that will move on to address the next question--whether decreases in reproduction realized at the point of hatch will persist through the larval stage. A manuscript containing the results from studies conducted 1992-95 is currently being prepared.

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Figure Captions

Figure 2.--Distributions of percent egg viability in sea lamprey nests of three classifications (untreated male parent, sterile male parent, and unobserved male parent) in the Misery River, Lake Superior, spring 1995.

Figure 3.--Distributions of percent egg viability in sea lamprey nests of three classifications (untreated male parent, sterile male parent, and unobserved male parent) in the Rock River, Lake Superior, spring 1995.

Figure 4.--Distributions of percent egg viability in sea lamprey nests of three classifications (untreated male parent, sterile male parent, and unobserved male parent) in the St. Marys River, Lake Huron, spring 1995.

Figure 5.--Distributions of percent egg viability in sea lamprey nests of three classifications (untreated male parent, sterile male parent, and unobserved male parent) in the Wolf River, Lake Superior, spring 1995.

Figure 6.--Distributions of percent egg viability in sea lamprey nests of three classifications (untreated male parent, sterile male parent, and unobserved male parent) for all study streams combined, 1992-95. In the bottom panel, the black bars show the predicted overall distribution given the observed distributions for untreated and sterile males. The ratio was selected by minimizing Chi-square comparing observed and predicted.

Figure 7.--Distributions of percent egg viability in sea lamprey nests of three classifications (untreated male parent, sterile male parent, and unobserved male parent) from the St. Marys River, 1993-95. In the bottom panel, the black bars show the predicted overall distribution given the observed distributions for untreated and sterile males. The ratio was selected by minimizing Chi-square comparing observed and predicted. The average expected ratio of sterile to untreated males over those years was 0.66.

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Egg Viability, Misery River, 1995

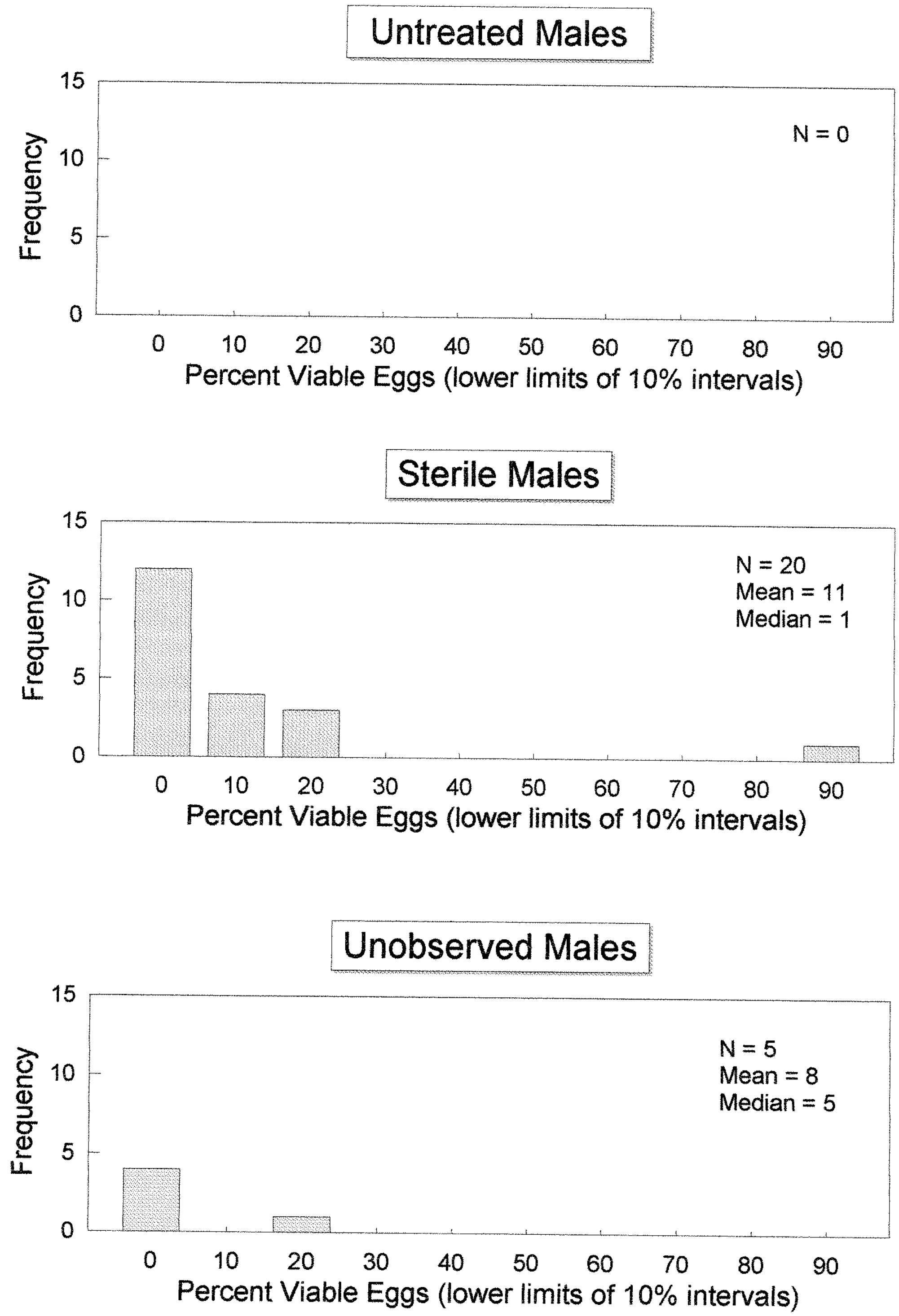


Figure 2

Egg Viability, Rock River, 1995

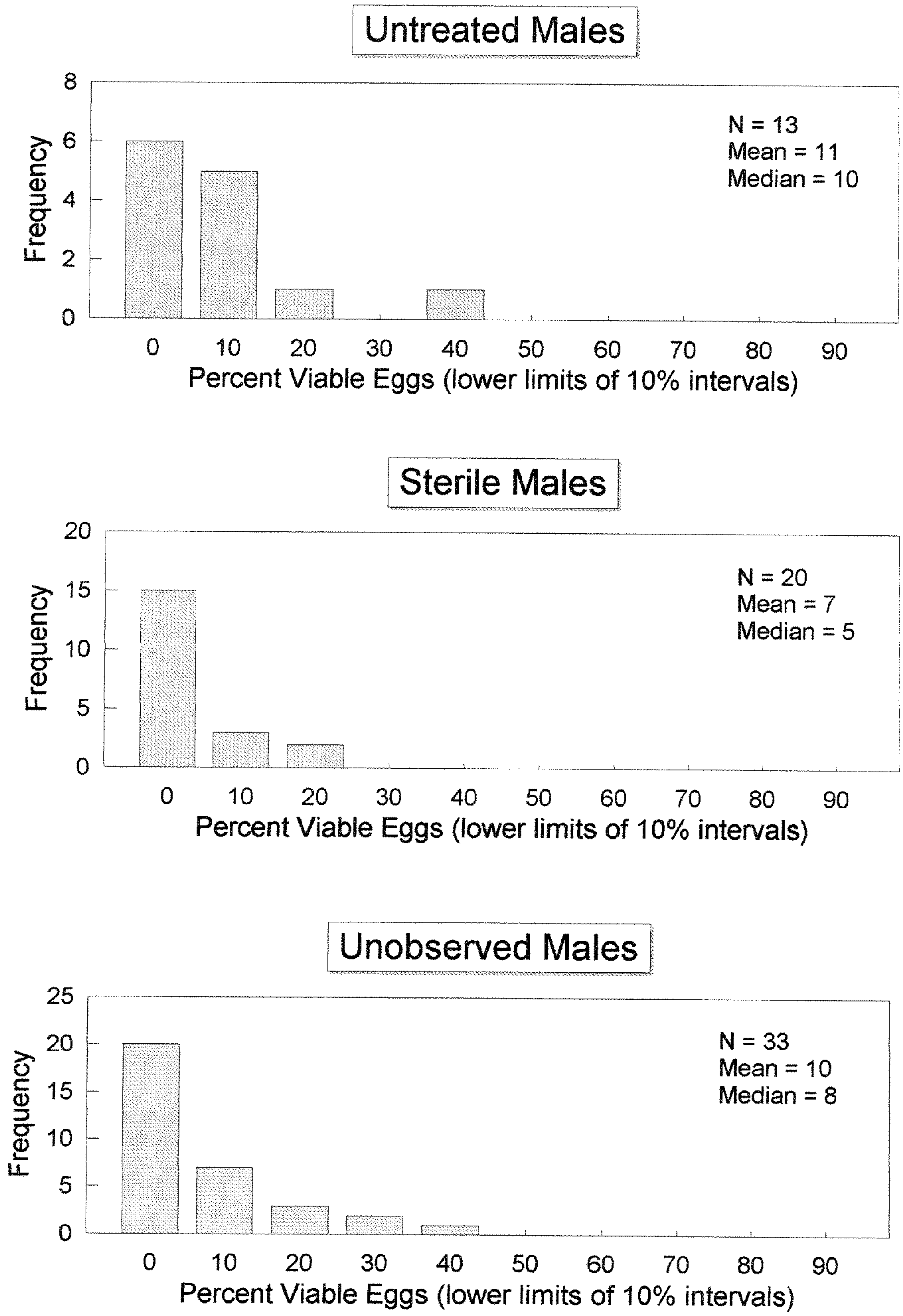


Figure 3

Egg Viability, St. Marys River, 1995

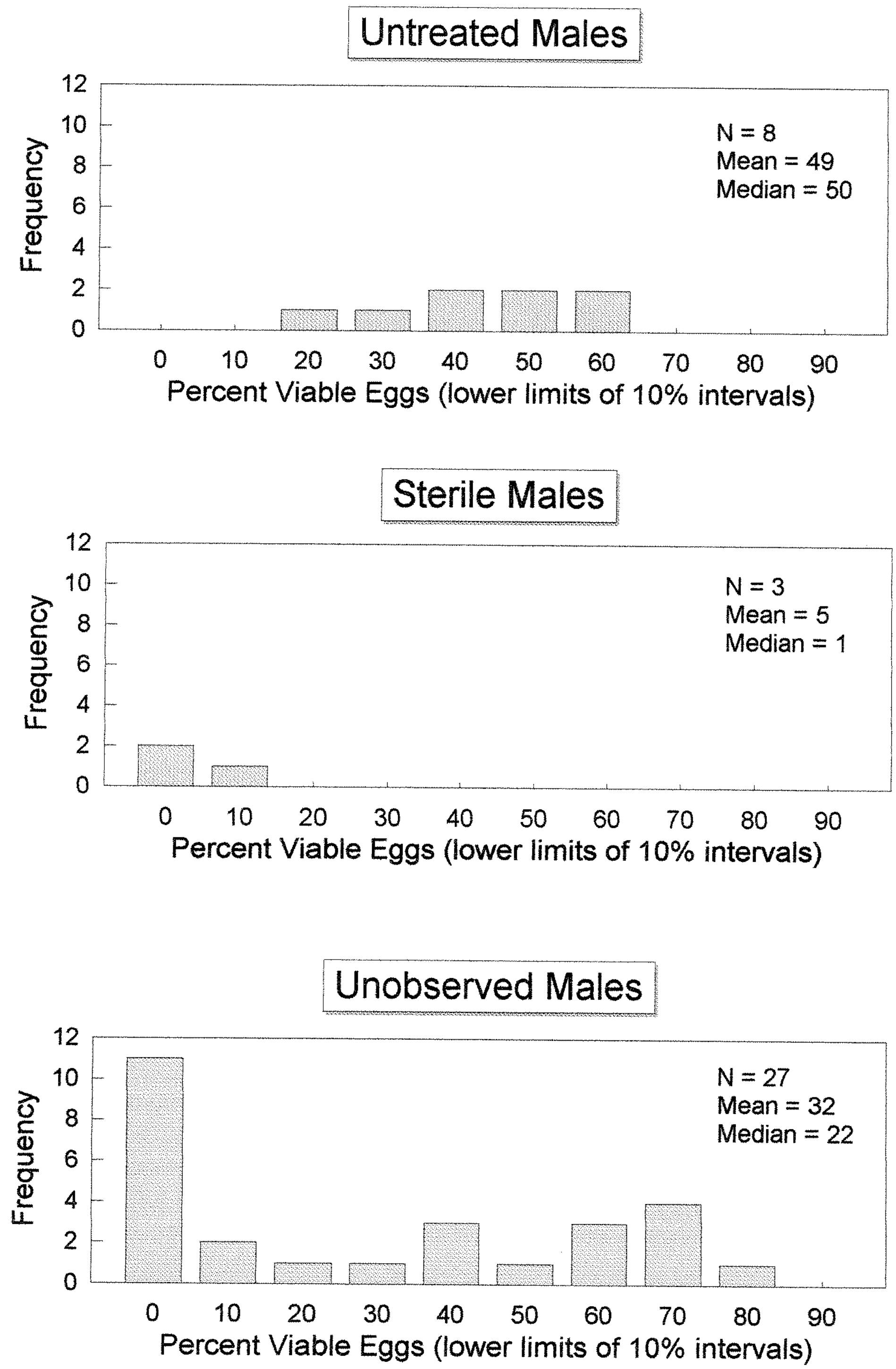


Figure 4

Egg Viability, Wolf River, 1995

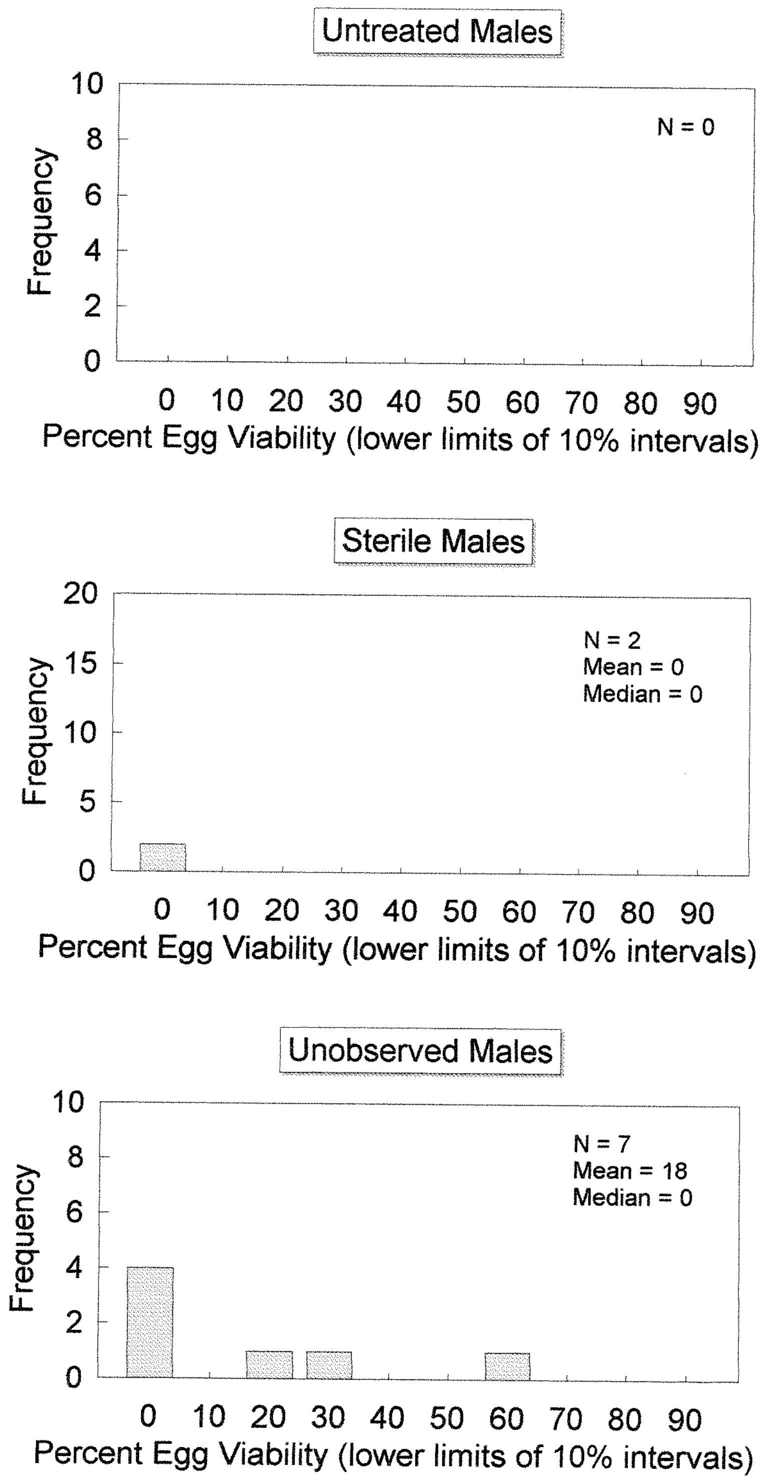
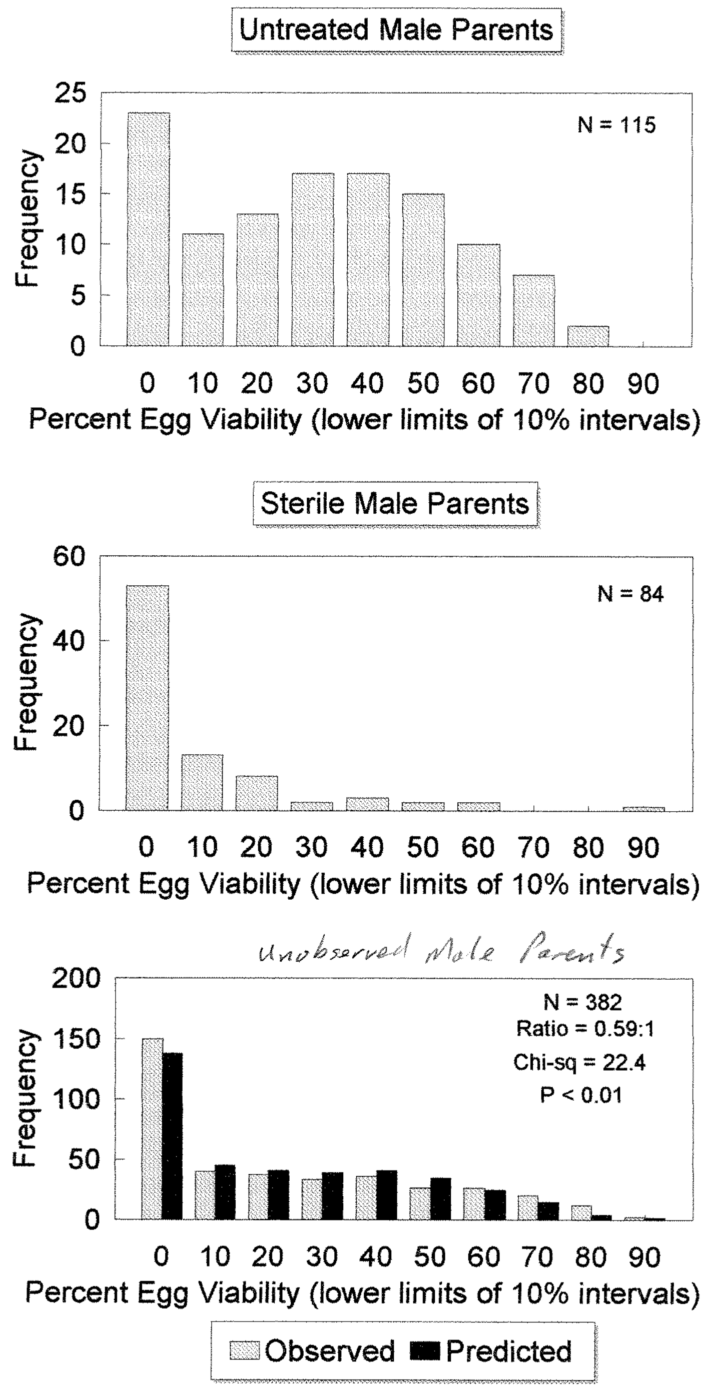


Figure 5

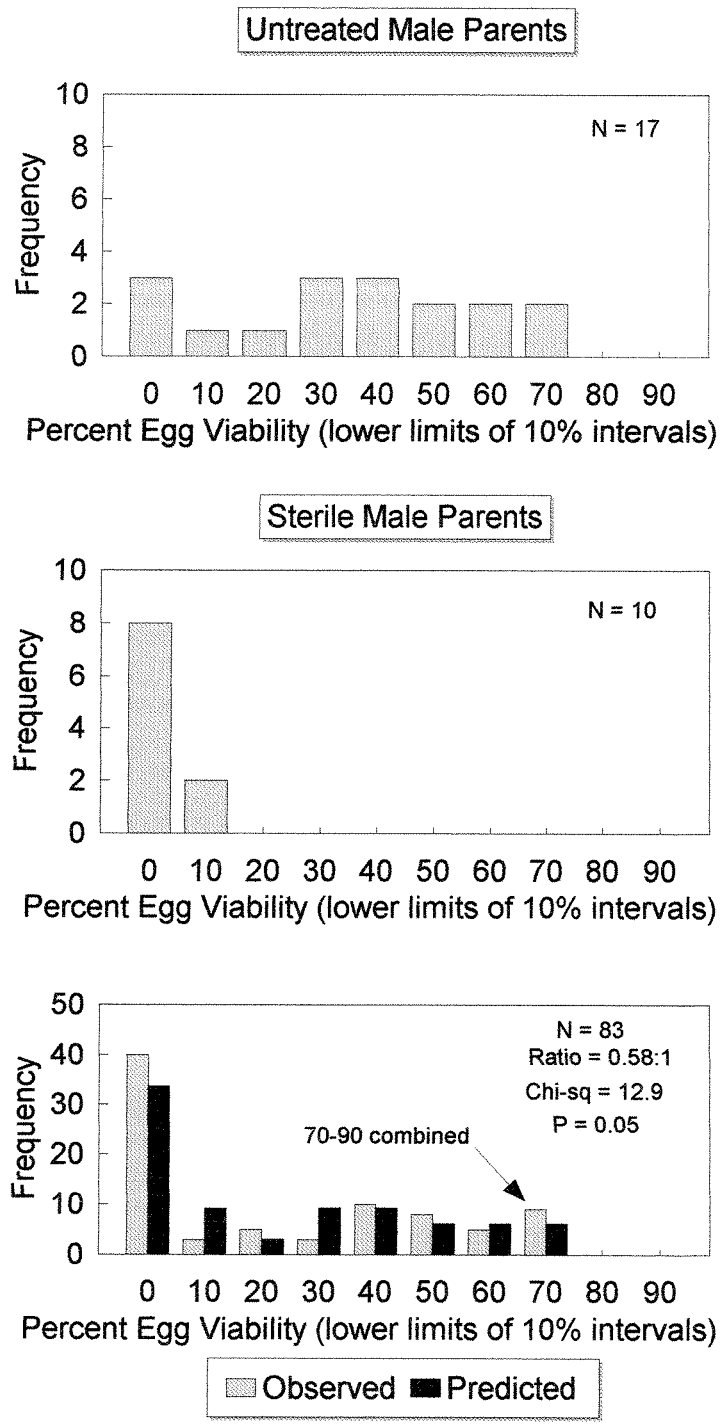
Egg Viability, All Streams Combined, 1992-95



	Resident	Sterile	Unobs.	Predicted	Ratio
0	23	53	150	137.4868	0.59
10	11	13	40	44.91784	
20	13	8	37	40.65872	Chi-sq
30	17	2	33	38.89041	22.40
40	17	3	36	40.57789	
50	15	2	26	34.71212	
60	10	2	26	24.2664	
70	7	0	20	14.62401	
80	2	0	12	4.178288	
90	0	1	2	1.687481	

Figure 6

Egg Viability, St. Marys River, 1993-95



	Resident	Sterile	Unobs.	Predicted	Ratio
0	3	8	40	33.64497	0.58
10	1	2	3	9.183768	
20	1	0	5	3.090097	Chi-sq
30	3	0	3	9.27029	12.89
40	3	0	10	9.27029	
50	2	0	8	6.180194	
60	2	0	5	6.180194	
70-90	2	0	9	6.180194	

Figure 7