

**GREAT LAKES FISHERY COMMISSION**  
*Research Completion Report<sup>1</sup>*

**Short-term Evaluation of Sterile Male Release in Selected  
Lake Superior Tributaries and the St. Marys River**

by

Roger Bergstedt  
National Fisheries Research Center--La Crosse  
Hammond Bay Biological Station  
11188 Ray Road  
Millersburg, Michigan 49759

Robert Young  
Department of Fisheries and Oceans  
Sea Lamprey Control Centre  
1 Canal Drive  
Sault Saint Marie, Ontario P6A6Wa

John Heinrich  
Marquette Biological Station  
1924 Industrial Parkway  
Marquette, Michigan 49855

Rod McDonald  
Department of Fisheries and Oceans  
Sea Lamprey Control Centre  
1 Canal Drive  
Sault Saint Marie, Ontario P6A 6W4

Kasia Mullet  
Marquette Biological Station  
1924 Industrial Parkway  
Marquette, Michigan 49855

March 21, 1995

---

<sup>1</sup>Project completion reports of Commission-sponsored general research are made available to the Commission's cooperators in the interest of rapid dissemination of information that may be useful in Great Lakes fishery management, research, or administration. The reader should be aware that project completion reports have not been through a peer review process and that sponsorship of the project by the Commission does not necessarily imply that the findings or conclusions are endorsed by the Commission.

Bergstedt, R., R. Young, R. McDonald, J. Heinrich and K. Mullet. 1995. Short-term Sterile Male Release Evaluation, 1994. Completion Report Submitted to the Great Lakes Fishery Commission, March 31, 1995.

Executive Summary--Where we collected sufficient data in 1994 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. In the Misery, Rock, and St. Marys rivers (the three streams where both population estimates to calculate expected ratios and observations of ratios of sterile males on nests were available) sterilized males were present on nests at slightly above the expected ratios. The observed and expected ratios were not significantly different, supporting the hypothesis that sterilized males nest in proportion to their abundance in the population.

Where we had sufficient data, measures of egg viability also suggested that females were attracted to nests with sterile males and there was a demonstrable reduction in viable eggs. There was a significant decrease in egg viability in nests with unobserved male parents compared to nests with untreated male parents in the Amnicon, Bad, and Wolf rivers (the three rivers where data were available). 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) being shifted to the left by undocumented participation of sterilized males.

We could not address the question of whether the competitiveness of sterile males changed with the ratio of sterilized to untreated males because streams where we failed to make estimates of the number of untreated males included all the intermediate ratios. Data were only available at ratios of 1:1 (one stream) and about 20:1 (two streams). The fact that expected and observed ratios were similar and not significantly different across that substantial range in ratio does suggest that competitiveness does not change with ratio.

Accomplishing the tasks needed to measure an effect of sterile male release in the field continued to prove difficult. The largest shortcoming of the 1994 field work was the inability to reliably produce solid estimates of the number of untreated males present in the study streams. Acting on that observation, field work proposed for 1995 is concentrated on streams where we are confident that the numbers of males in the natural run will be known.

**Short-term Sterile Male Release Evaluation, 1994**

A Completion Report Submitted to the Great Lakes Fishery Commission

March 21, 1995

by

Roger Bergstedt  
National Biological Service, Hammond Bay Biological Station  
11188 Ray Road, Millersburg, Michigan 49759

Robert Young and Rod McDonald  
Department of Fisheries and Oceans, Sea Lamprey Control Centre  
1 Canal Drive, Sault Ste. Marie, Ontario P6A 6W4

John Heinrich and Kasia Mullet  
U.S. Fish and Wildlife Service, Marquette Biological Station  
1924 Industrial Parkway, Marquette, Michigan 49855

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 1994 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. 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). 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 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.

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 which met those criteria and were selected as study streams are the Pancake River (Ontario), the Wolf River (Ontario), the Amnicon River (Wisconsin), the Bad River (Wisconsin), the Misery River (Michigan), and the Rock River (Michigan). By distributing the effort over six streams varying widely in size, the risk of not achieving satisfactory results because of water conditions was reduced. 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 male on the observed ratios.

## **Results**

Observed versus Expected Ratios--Of the six streams studied on Lake Superior (Figure 1) the largest number of observations of males on nests (Table 1) was on

the Amnicon River (185 total, 54 sterile to 131 untreated) followed by the Bad River (157, 143 to 14), the Misery Rivers (108, 103 to 5), the Rock River (54, 52 to 2), the Wolf River (17, 5 to 12), and the Pancake River (5, 1 to 4). In three of the six streams, we reached our goal of 100 observations of males on nests. In one other (the Rock River, 54 observations), we also did reasonably well and would have come close if the natural run had been typical; the runs were generally much smaller than normal in Lake Huron and Superior tributaries in spring 1994. The very low numbers of males observed in the north shore streams (Wolf and Pancake Rivers) are difficult to explain. Those rivers were picked in part because of the observability of the spawning grounds. However, the number of males observed there were much lower than along the south shore. This was particularly obvious with the observations of sterilized males. The percentage of the sterilized males placed in south shore streams that were subsequently observed on nests ranged from 4 to 8%. The corresponding percentages in the north shore streams were 0.1% (Pancake River) and 0.4% (Wolf River). Since those streams were picked for observability and the work was done according to the same protocol, the implication is that behavior there was somehow different. This could involve either emigration from the stream or timing of spawning activity. There is no direct evidence in our observations to say what actually happened.

The available data suggested that sterile males were constructing and occupying nests in proportion to their numbers in the stream. Comparisons of observed and expected ratios of sterile to untreated males on nests were possible in three streams. Of the six Lake Superior streams, population estimates of untreated males (necessary to determine an expected ratio of sterile to untreated males) were only available for two. We had estimates only on the Misery and Rock Rivers, where known populations were created by passing animals above barriers. The failure to make population estimates was in part due to small runs of sea lampreys that resulted in small trap catches, and therefore, low numbers marked and released and few recaptures. In some cases there were also errors made by personnel contracted to conduct the mark-recapture studies. In addition to the two Lake Superior streams, an estimate of the expected ratio of sterile to untreated males was also available for the St. Marys River, the outflow from Lake Superior to Lake Huron. In the Misery, Rock, and St. Marys rivers, the observed ratios were all higher than the expected ratios but the differences were not significant. All three comparisons therefore suggested that the sterile males were competitive as far as building and occupying nests. The subset of streams we used for comparison were selected only on the basis of data availability.

Percent Egg Viability--Sterile males also appeared to 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 2-8). 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. As with the observations of males on nests, data were deemed sufficient to make that comparison on only three streams--in this case, the Amnicon River (N=137, Figure 2), the Bad River (N=20, Figure 3), and the Wolf River (N=22, Figure 8). On the Misery and Rock rivers, the spawning run was very small and there were simply not enough females in the rivers to collect enough egg samples. On the St. Marys river, we observed and sampled as many nests as could be safely accessed. The low numbers of nests sampled in the Pancake River was due to the very small number of males observed. In all three streams where we compared the percent egg viability among nests with untreated and unobserved male parents, the egg-viability distribution for nests with unobserved male parents was shifted significantly (Kolmogorov-Smirnov test,  $P < 0.05$ ) to the left compared to those with untreated male parents. This suggests that sterile males do in fact attract and mate with females in high enough numbers to produce a measurable effect.

Given an observed ratio of sterile to untreated males, the size of the reduction in percent viability can be predicted. Predicted percent viabilities in the unobserved nests calculated assuming total sterility of sterilized males, using the observed sterile:untreated ratios (Table 1), and using percent viability in nests with untreated males (Figures 2, 3, and 8) as a baseline were: lower than measured values in the nests with unobserved male parents in the Bad River (predicted viability 3%, measured 20%), close to measured values in the Amnicon River ( predicted 28%, measured 29%), and greater than measured values in the Wolf River (predicted 19%, measured 7% ).

Comparison of the results of the 1994 quality control study to the distribution of egg viability in nests on the Amnicon River with sterile male parents (Figure 2) suggested that nests could not be reliably assumed to have just one male involved and raised questions about an underlying assumption of our approach. The quality control study (Fredricks 1994) suggested that although some (9%) of the males might not receive a proper dose of sterilant, most spawning should result in near zero percent viability. Because the males in that study were ripened naturally over a period of weeks, we felt it would be unlikely that results such as on the Amnicon are due to incomplete sterilization. It seems more likely that there was some degree of movement of males among nests between the daily observations.

To examine whether the egg viability data from the Amnicon River was consistent with the above explanation we attempted to reproduce the distribution of percent

viability in the nests with unobserved males. This required first making an informed guess about the actual distribution of percent viability in nests with sterile and untreated male parents if the nests had been accurately classified. This was done in two ways. We moved observations between the first and second panels in Figure 2 to more closely match the quality control results (Figure 9). We also specified a mean and standard deviation for the distributions to accomplish the same task (Figure 10). By specifying the distributions of percent viability for the two classes of males and their ratio in the population we could then calculate a predicted distribution of percent viability in the nests with unobserved male parents. These could then be compared to the actual distribution observed in the Amnicon with Chi-square. In both Figure 9 and 10, we adjusted the ratio to minimize Chi-square and achieve the best fit to the observed data. Given the distributions in Figure 9, a ratio of 0.8:1 produces the distribution most similar to the observed and Figure 10 suggests a ratio of about 0.6:1. Both are not far from the observed ratio of 0.4:1 and indicate that a ratio of less than 1:1 might produce the shift we observed on the Amnicon River. Because we are stipulating the distributions and not deriving them from the data, this analysis does not prove that the decreased viability of eggs in the unobserved nests was due to the release of sterile males. It is, however, encouraging that the distribution observed is close to that expected given the apparent ratio and the presumed degree of sterility.

Relation of Expected to Observed Ratios--The expected ratios in the three streams where we had data were 1.0:1 (St. Marys River), 19.6:1 (Misery river), and 21.0:1 (Rock River). Because of the small number of points and the distribution of independent variables (1.0, 19.6, and 21.0) we did not attempt a regression analysis. The regression would have been fit between the point at 1:1 and the two points near 20:1, giving a trivial result. None of the observed ratios, however, were significantly different than expected across this substantial range, and it seems likely that the null hypothesis of a linear effect will eventually be accepted. We will have data at more intermediate ratios following the 1995 studies and hope to complete this analysis then.

## Summary

Where we collected sufficient data in 1994 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. In the Misery, Rock, and St. Marys rivers (the three streams where both population estimates to calculate expected ratios and observations of



ratios of sterile males on nests were available) sterilized males were present on nests at slightly above the expected ratios. The observed and expected ratios were not significantly different, supporting the hypothesis that sterilized males nest in proportion to their abundance in the population.

Where we had sufficient data, measures of egg viability also suggested that females were attracted to nests with sterile males and there was a demonstrable reduction in viable eggs. There was a significant decrease in egg viability in nests with unobserved male parents compared to nests with untreated male parents in the Amnicon, Bad, and Wolf rivers (the three rivers where data were available). 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) being shifted to the left by undocumented participation of sterilized males.

We could not address the question of whether the competitiveness of sterile males changed with the ratio of sterilized to untreated males because streams where we failed to make estimates of the number of untreated males included all the intermediate ratios. Data were only available at ratios of 1:1 (one stream) and about 20:1 (two streams). The fact that expected and observed ratios were similar and not significantly different across that substantial range in ratio does suggest that competitiveness does not change with ratio.

Accomplishing the tasks needed to measure an effect of sterile male release in the field continued to prove difficult. The largest shortcoming of the 1994 field work was the inability to reliably produce solid estimates of the number of untreated males present in the study streams. Acting on that observation, field work proposed for 1995 is concentrated on streams where we are reasonably confident that the numbers of males in the natural run will be known.

**References**

- Bergstedt, R. A., R. Young, J. Heinrich, R. McDonald, and K. Mullet. 1994. Short-term Evaluation of Sterile-Male Release in Six Lake Superior Tributaries. Research Proposal Submitted to the Great Lakes Fishery Commission, January, 1994.
- Fredricks, K. T. 1994. Evaluation of the current male sea lamprey sterilization process. Completion Report HB-94-00075-01, Lake Huron Biological Station.
- Hanson, L. H. and P. J. Manion. 1978. Chemosterilization of the sea lamprey (Petromyzon marinus). Great Lakes Fishery Commission, Technical Report No. 29, 15p.
- Hanson, L. H. and P. J. Manion. 1980. Sterility method of pest control and its potential role in an integrated sea lamprey (Petromyzon marinus) control program. Canadian Journal of Fisheries and Aquatic Sciences 37:2108-2117.
- Hanson, L. H., A. K. Lamsa, P. J. Manion, J. G. Seelye, and J. J. Tibbles. 1986a. Potential sites for the implementation of the sterile-male-release technique in sea lamprey control. Presented to the Sea Lamprey Committee of the Great Lakes Fishery Commission, April 1986, Agenda Item 3.0. 40 p.
- Hanson, L. H., W. E. Daugherty, A. K. Lamsa, P. J. Manion, Rodney B. McDonald, and J. G. Seelye. 1986b. Progress report of the sterile-male-release technique (SMRT) task force. Presented to the Great Lakes Fishery Commission at the Interim Meeting, December 1986. 28 p.
- Hanson, L. H., W. E. Daugherty, J. H. Heinrich, A. K. Lamsa, P. J. Manion, J. G. Seelye, and J. G. Weise. 1987. A preliminary field trial to simulate implementation of the sterile-male-release technique: Release of tagged, normal lampreys into Lake Superior. Great Lakes Fishery Commission Interim Meeting, December 1986. 15 p.
- Hanson, L. H., W. E. Daugherty, J. H. Heinrich, A. K. Lamsa, R. J. Schuldt, J. G. Seelye, and J. G. Weise. 1988. Progress report of the sterile-male-release technique (SMRT) task force with recommendations for future studies and schedule for implementing the technique. Presented to the Sea Lamprey Committee of the Great Lakes Fishery Commission, March 1988, 52 p.
- Hanson, L. H., J. W. Slade, and J. H. Heinrich. 1989. Dispersal and spawning behavior of male sea lampreys captured in Lakes Huron and Michigan

streams and released in three Lake Superior tributaries Presented to the Sea Lamprey Committee of the Great Lakes Fishery Commission, April 1989.

Hanson, L. H., J. H. Heinrich, G. T. Klar, A. K. Lamsa, J. Powpowski, J. G. Seelye, and J. G. Weise. 1990. Integration of the sterile-male-release technique in the sea lamprey control program in Lake Superior. Presented to the Lake Superior Technical Committee, February, 1990.

Table 1.--Estimated and observed ratios of sterile to untreated male sea lampreys in seven study streams during the 1994 field season.

	Amnicon	Bad	Misery	Rock	Wolf	Pancake	St. Marys
Target ratio (sterile:untreated)	1:1	2:1	10:1	4:1	6:1	8:1	-
Estimated number of untreated males	-	-	67	37	-	-	2,667
Number of sterile males	954	3,500	1,313	777	1,350	900	2,667
Estimated ratio (sterile:untreated)	-	-	19.6:1	21.0:1	-	-	1.0:1
Untreated males observed on nests	131	14	5	2	12	4	10
Sterile males observed on nests	54	143	103	52	5	1	17
Observed ratio (Sterile:Untreated)	0.4:1	10.2:1	20.6:1	26.0:1	0.4:1	0.25:1	1.7:1
95% Confidence limit for observed ratio	0.3:1 to 0.6:1	6.5:1 to 21.4:1	10.6:1 to 100+:1	10.4:1 to 100+:1	0.1:1 to 1.0:1	0.0:1 to 1.2:1	0.8:1 to 4.3:1
Chi-square for estimated vs. observed ratios	-	-	0.01 (ns)	0.09 (ns)	-	-	1.81 (ns)

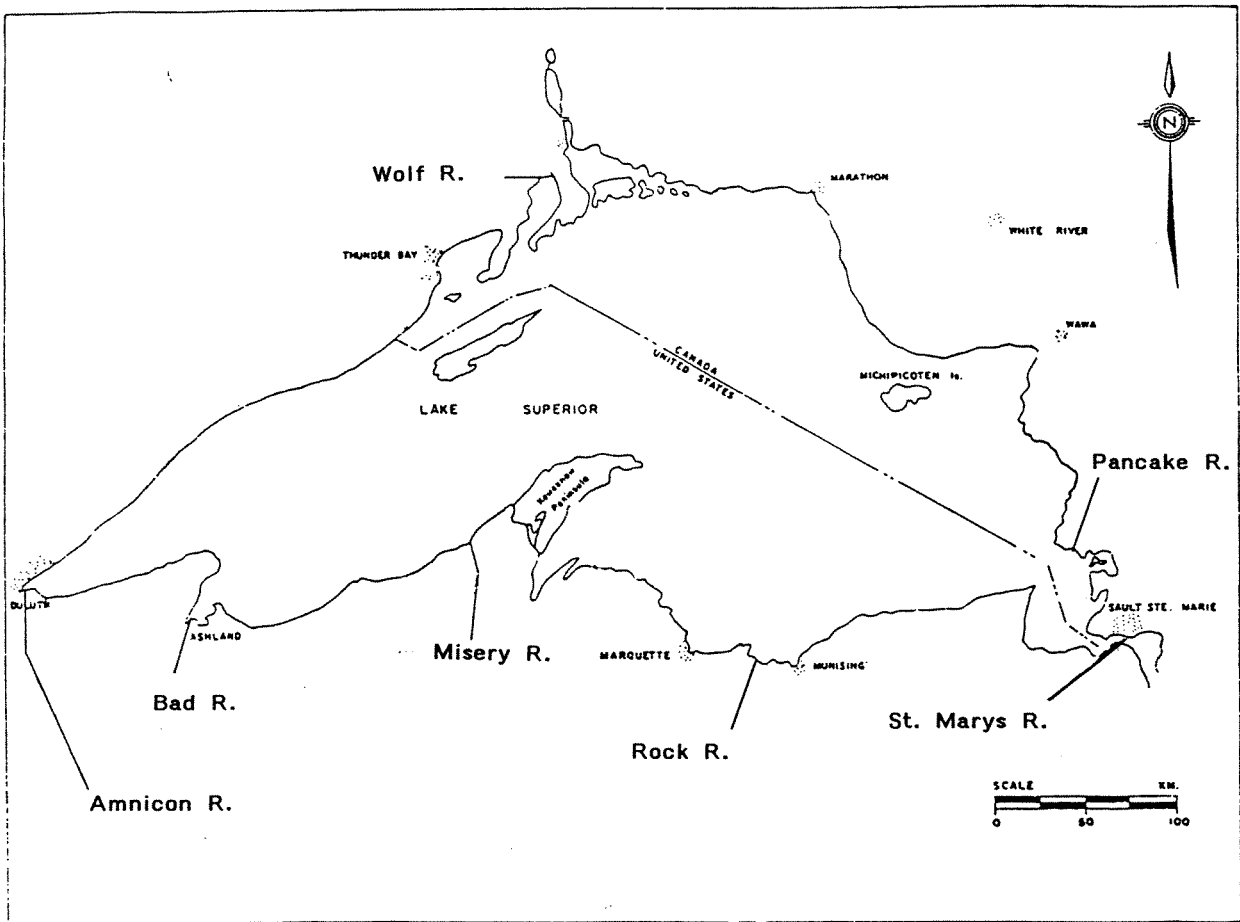


Figure 1.--Lake Superior, showing locations of the study streams.

## Egg Viability, Amnicon River, 1994

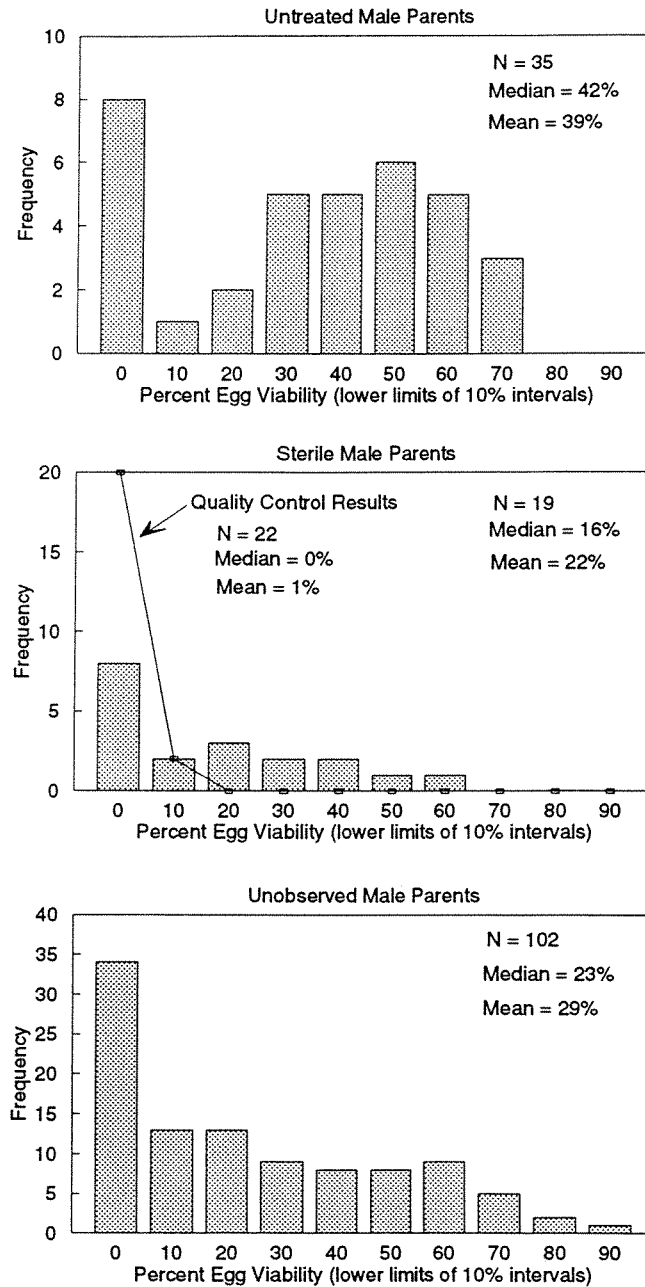


Figure 2.--Distributions of percent egg viability in sea lamprey nests of three classifications (untreated male parent, sterile male parent, and unobserved male parent) in the Amnicon River, Lake Superior, spring 1994. The line in the center panel shows results of the 1994 quality control study.

# Egg Viability, Bad River, 1994

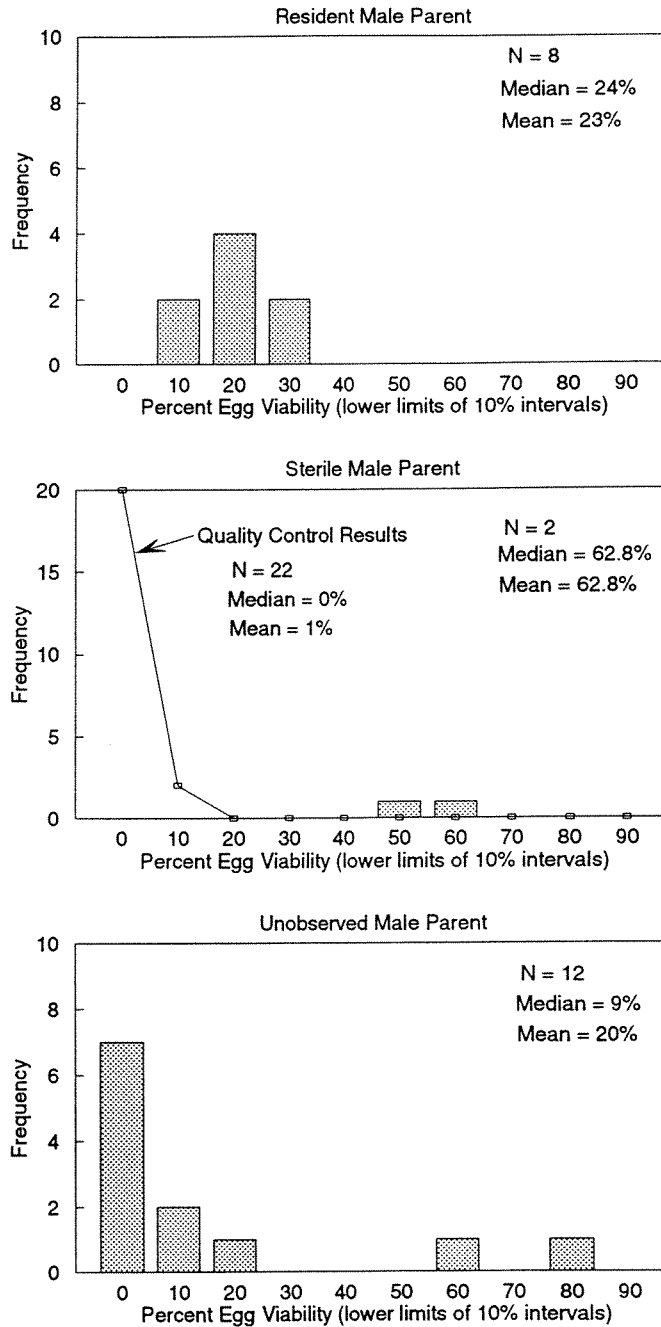


Figure 3.--Distributions of percent egg viability in sea lamprey nests of three classifications (untreated male parent, sterile male parent, and unobserved male parent) in the Bad River, Lake Superior, spring 1994. The line in the center panel shows results of the 1994 quality control study.

## Egg Viability, Misery River, 1994

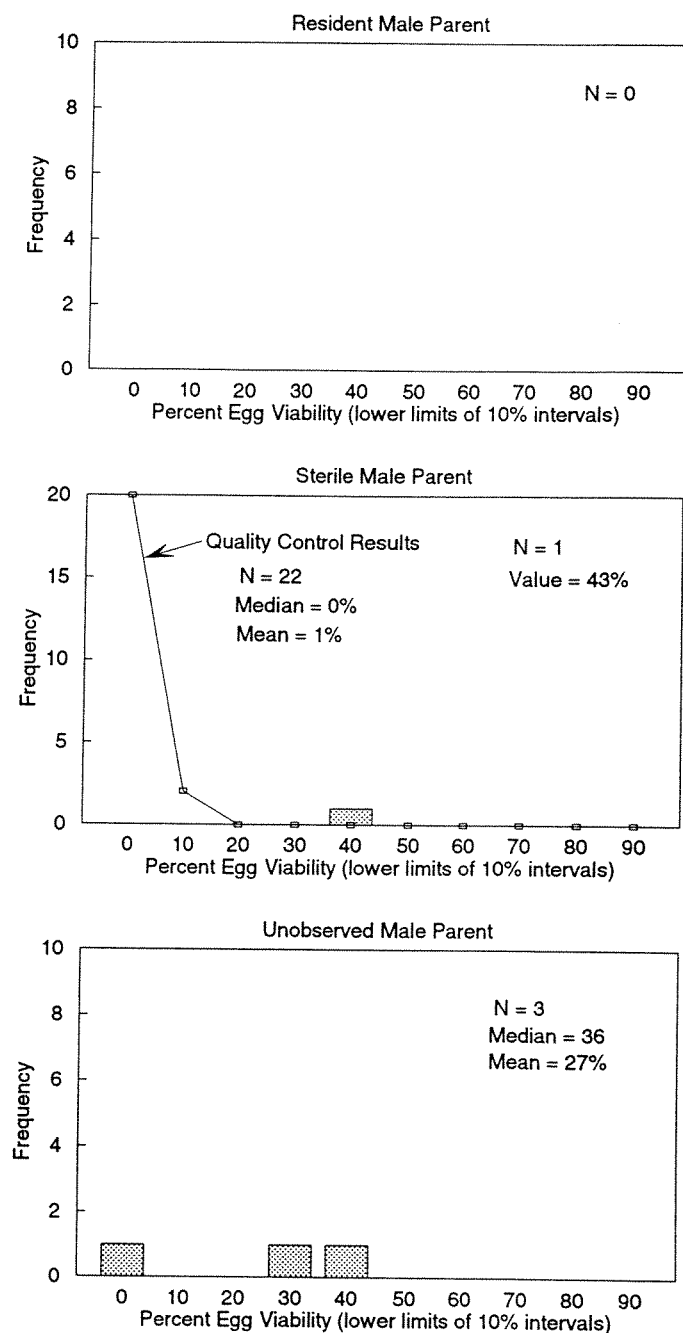


Figure 4.--Distributions of percent egg viability in sea lamprey nests of three classifications (untreated male parent, sterile male parent, and unobserved male parent) in the Misery River, Lake Superior, spring 1994. The line in the center panel shows results of the 1994 quality control study.



# Egg Viability, Rock River, 1994

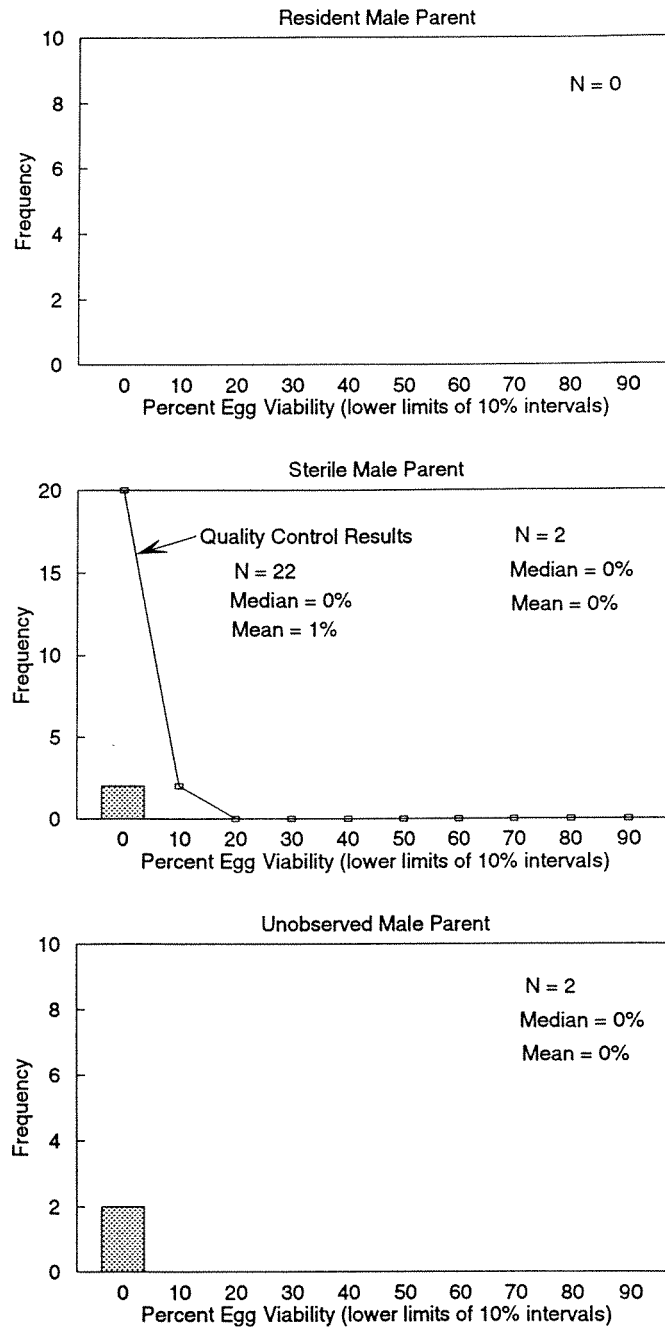


Figure 5.--Distributions of percent egg viability in sea lamprey nests of three classifications (untreated male parent, sterile male parent, and unobserved male parent) in the Rock River, Lake Superior, spring 1994. The line in the center panel shows results of the 1994 quality control study.

# Egg Viability, St. Marys River, 1994

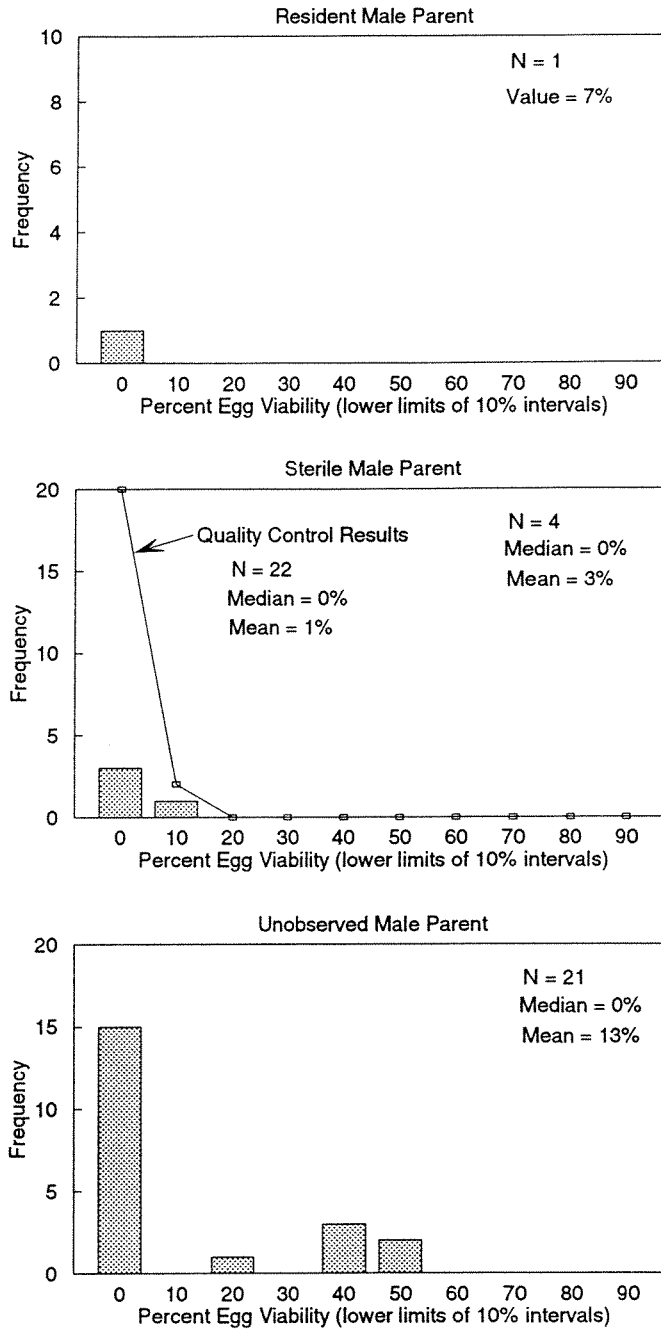


Figure 6.--Distributions of percent egg viability in sea lamprey nests of three classifications (untreated male parent, sterile male parent, and unobserved male parent) in the St. Marys River, Lake Huron, spring 1994. The line in the center panel shows results of the 1994 quality control study.

# Egg Viability, Pancake River, 1994

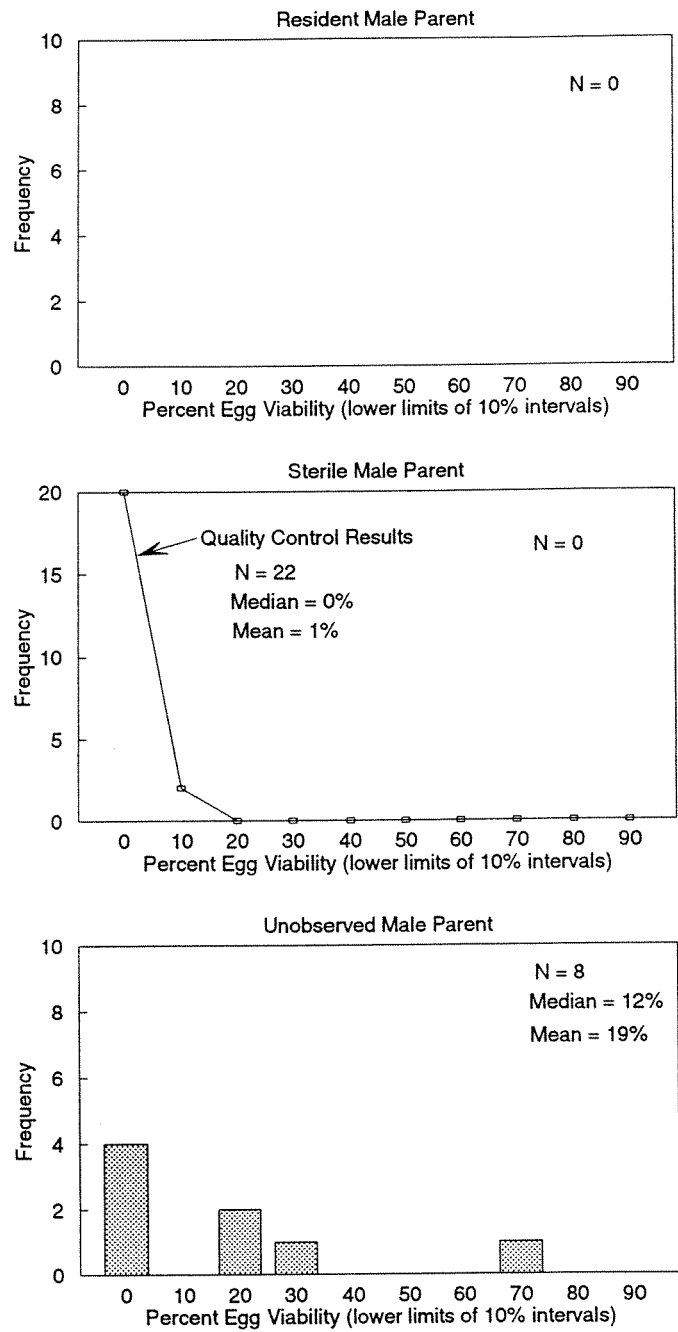


Figure 7.--Distributions of percent egg viability in sea lamprey nests of three classifications (untreated male parent, sterile male parent, and unobserved male parent) in the Pancake River, Lake Superior, spring 1994. The line in the center panel shows results of the 1994 quality control study.

18  
Egg Viability, Wolf River, 1994

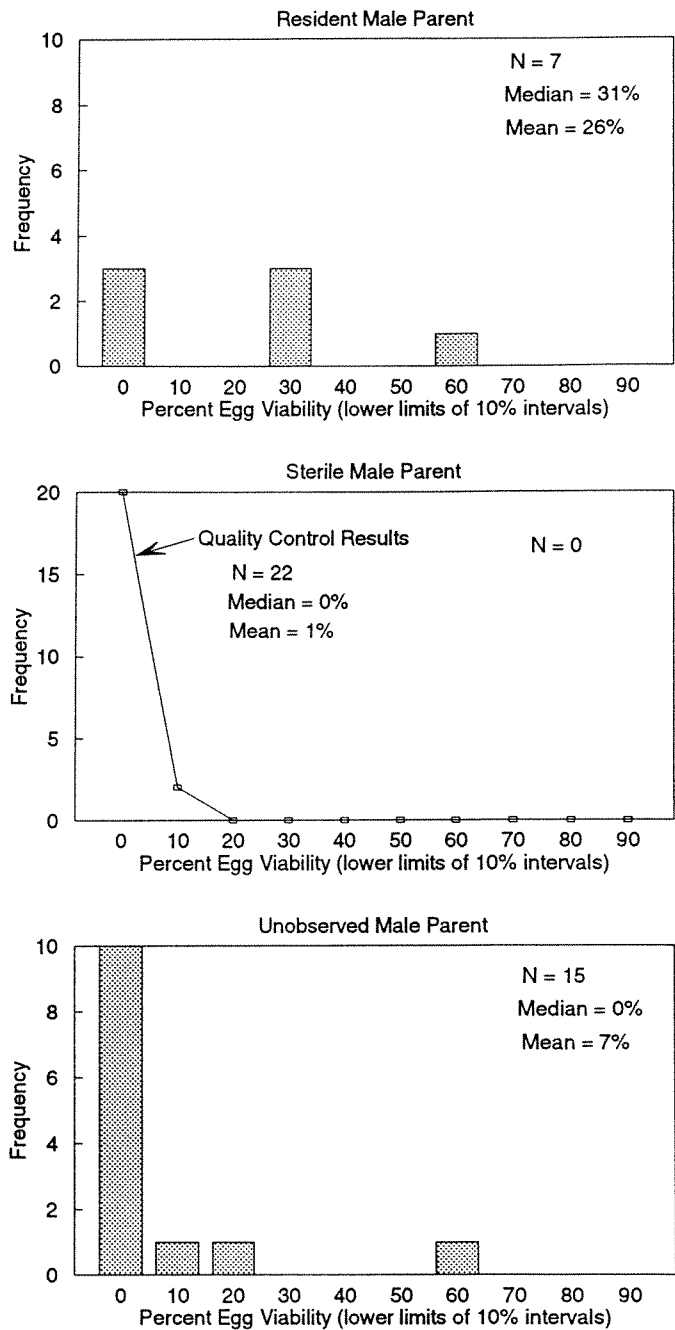
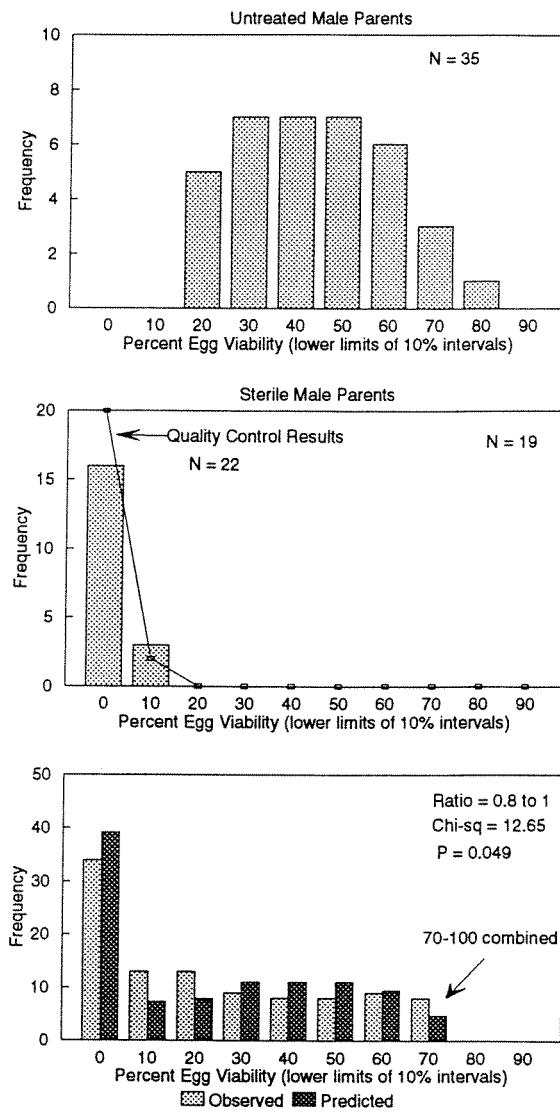


Figure 8.--Distributions of percent egg viability in sea lamprey nests of three classifications (untreated male parent, sterile male parent, and unobserved male parent) in the Wolf River, Lake Superior, spring 1994. The line in the center panel shows results of the 1994 quality control study.

# 19 Egg Viability, Amnicon River, 1994



	Resident	Sterile	Unobs.	Predicted	Ratio
0	0	16	34	39.2	0.84
10	0	3	13	7.35	
20	5	0	13	7.92	Chi-sq
30	7	0	9	11.09	12.65
40	7	0	8	11.09	
50	7	0	8	11.09	
60	6	0	9	9.05	
70-100	3	0	8	4.75	

Figure 9.--Hypothesized distributions of percent egg viability in sea lamprey nests with untreated male parent and sterile male parent, and the predicted and observed distributions in nests with unobserved male parents in the Amnicon River, Lake Superior, spring 1994. The line in the center panel shows results of the 1994 quality control study. The hypothesized distributions were adjusted by eye from Figure 2, using the quality control study results as a guide.

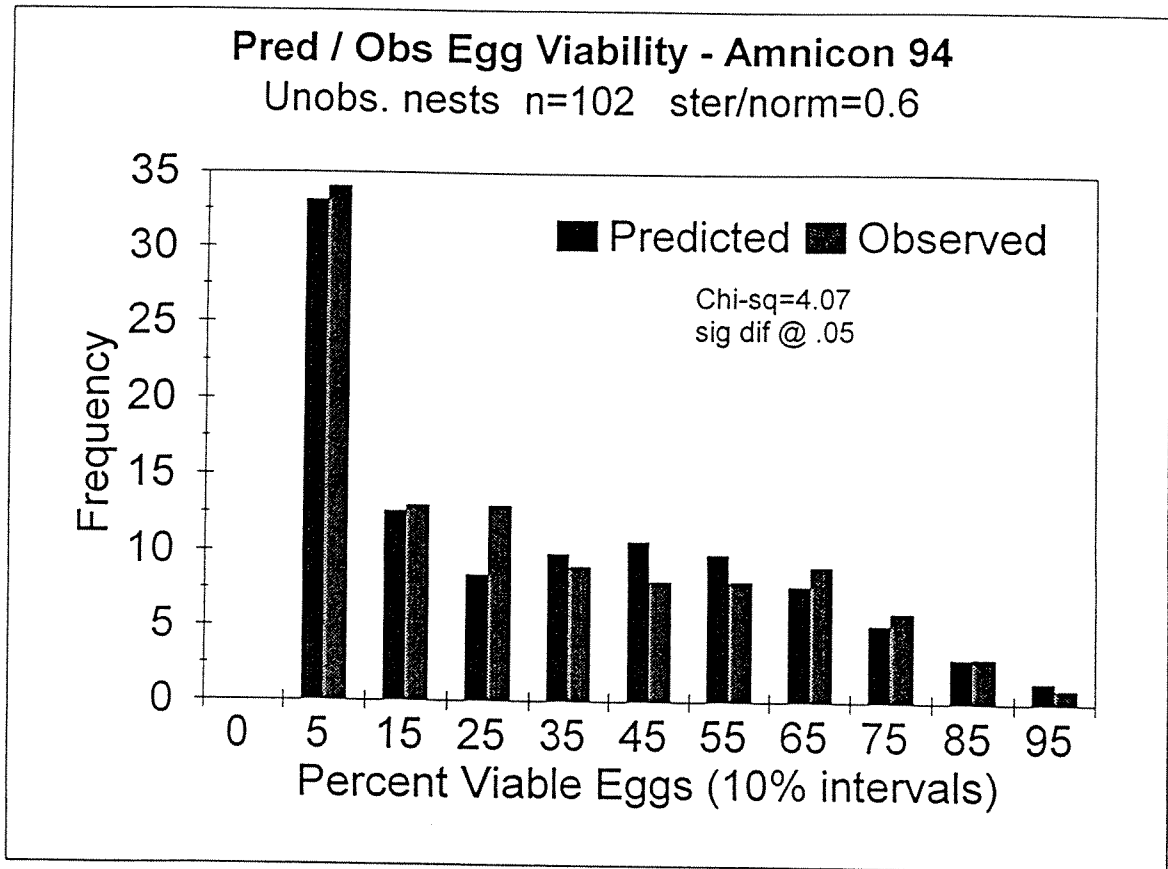


Figure 10.--Hypothesized distributions of percent egg viability in sea lamprey nests with untreated male parent and sterile male parent, and the predicted and observed distributions in nests with unobserved male parents in the Amnicon River, Lake Superior, spring 1994. The line in the center panel shows results of the 1994 quality control study. The hypothesized distributions were generated using means and standard deviations selected using the observed distributions in Figure 2 and the quality control study results as a guide. Figure 10 by G. Christie.