

**A Statistical Review of Sampling
of Fishes in the Long Term Resource Monitoring Program**

by

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Preface

This report is a product of the Long Term Resource Monitoring Program (LTRMP) for the Upper Mississippi River System. The LTRMP was created in 1987 as a cooperative effort by the U.S. Army Corps of Engineers, the U.S. Fish and Wildlife Service, and resource management

and research agencies of the states of Illinois, Iowa, Minnesota, Missouri, and Wisconsin. The overall mission of the LTRMP is to provide decision makers, resource managers, and resource users with information needed to maintain the Upper Mississippi River System as a viable multiple-use ecosystem. This mission is undertaken using a combination of long-term trend monitoring and focused research into identified problem areas.

Data (factual record) and information (usable interpretation of data) are the primary products of the LTRMP. Data on water quality, vegetation, aquatic macroinvertebrates, and fish are collected using a network of six field stations on the Upper Mississippi and Illinois Rivers. Analysis, interpretation, and reporting of information are conducted at these field stations and at the Environmental Management Technical Center, the operational center of the LTRMP. Informational products of the LTRMP include professional presentations, reports, and publications in the open and peer-reviewed scientific literature.

This document summarizes a statistical evaluation of the sampling design used by the Fishery Component of the LTRMP. This report satisfies, for Fiscal Year 1993, Task 2.2.8.5, *Statistical Evaluation of Fish Data* under Goal 2, *Monitor and Evaluate the Condition of the Upper Mississippi River Ecosystem*, as specified in the Operating Plan for the Long Term Resource Monitoring Program (USFWS 1992). The purposes of this report are to (1) identify important components of variance in the fish sampling design and other statistical features of the fish monitoring design and (2) make any necessary recommendations for change to the LTRMP fish sampling design. The primary data summarized in this report are available from the Environmental Management Technical Center.

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Summary

Prior to 1993, the fish monitoring design of the Long Term Resource Monitoring Program (LTRMP) was based on fixed-site sampling, wherein non-randomly selected permanent stations within particular habitat types were sampled repeatedly through time. The LTRMP fish sampling sites were selected based on subjective judgments that particular sites were representative of their habitat type. Unfortunately, fixed-site sampling cannot produce data that can be used to verify whether specific sampling sites are truly representative of habitats. Because fixed-site sampling designs are not spatially randomized, they cannot support statistically valid conclusions about patterns or trends in the larger study area. Fixed-point sampling designs can only be used to make inferences about the specific set of sampling sites. For example, the LTRMP fish sampling design cannot be used to make statistically valid tests or trend estimates within whole habitat classes or study reaches. This reliance on subjectively chosen permanently fixed sampling sites was the major statistical deficiency of the LTRMP fish monitoring design.

Fixed-site sampling is often justified based on the supposition that, for example, fish catches are much more variable among different site locations than within any specific site. Although commonly accepted, this supposition has not been critically tested. To examine this supposition, I partitioned the 1990 LTRMP fish data into unique combinations of species, study reach, sampling gear, habitat class, and time period and estimated within- and among-site variance from each partition. Among-site variation was actually less than within-site variation in over 50% of the partitions. The hypothesis that among-site variation was less than or equal to within-site variation could not be rejected. Clearly, the 1990 LTRMP fish data do not support the supposition that within-site variation is less than among-site variation. Rather, the data support the conclusion that among- and within-site variation are approximately equal.

Stratified random sampling, where habitat classes serve as strata, is the statistically valid alternative to LTRMP fixed-site sampling. A test of logistics of random sampling in two areas of Pool 8 in which navigation is particularly difficult suggests stratified random sampling is practical, and most randomly selected sampling sites can be located to within 100 m using only a base map. Further, LTRMP experience with stratified random sampling of macroinvertebrates demonstrates feasibility for large rivers.

Given that (1) the current fixed-site fish sampling design is statistically invalid for trend monitoring within whole habitat classes and study reaches, (2) the rationale for fixed-site design use is not supported by LTRMP data, and (3) there do not appear to be any insurmountable logistical impediments to the statistically valid randomized sampling alternative, the LTRMP Fisheries Component implemented a stratified random sampling design beginning with the 1993 field season. The only viable reasons to retain fixed sites is to sample the tailwaters of locks and dams and to conduct specialized monitoring at those sites that are of intrinsic interest, for example, those where threatened or endangered species were previously detected.

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Introduction

From the inception of the Long Term Resource Monitoring Program (LTRMP), fish sampling has been conducted at subjectively chosen permanently fixed sampling sites. This use of subjectively chosen fixed sites was the major statistical limitation of the fish sampling component of the LTRMP because the ability to document trends is restricted to the limited set of fixed sites. The original LTRMP fish monitoring design could not provide valid information on trends or other patterns within whole habitat classes or study reaches. Other statistical issues, such as sample size and potential redundancy among sampling gears, are of minor importance compared to lack of design-based randomness.

Fixed-site sampling, wherein non-randomly selected permanent stations are sampled repeatedly through time, is commonly used to monitor freshwater fishes but is uncommon for other uses including monitoring estuarine and marine fishes. Fixed-site sampling ignores random spatial variation, instead treating the specific sampling sites as the only areas of interest. Statisticians and scientists generally consider fixed-site sampling inefficient or invalid for the lack of randomization ensuring unbiased results and validity of key statistical assumptions (Green 1979).

Fixed-site sampling is usually justified based on the supposition that among-site variation is much larger than within-site variation, even within a homogeneous habitat class. If this supposition were true, detection of trends within specific sites would require fewer samples than in the larger habitat class. However, this supposition also has been used to claim that high among-site variance makes estimation and testing in the larger habitat class impractical. Further, it is sometimes claimed that patterns and trends within a set of fixed sites can be used as "indices" of those in the larger habitat class (Johnson and Nielsen 1983). If taken together, these last two claims amount to circular and false logic. Data obtained from methods not supporting valid inference over a particular area do not attain validity to track trends in that area merely by calling them an index.

Sources of Variation in LTRMP Fish Data

Despite the fact that fixed-site sampling is inadequate for inference outside the set of fixed sites regardless of the relative sizes of among- and within-site variances of catch, testing the supposition that among-site variance is greater than within-site variance is still useful. Although commonly accepted, this supposition has not been critically examined in the primary scientific literature.

To examine this supposition, I partitioned the 1990 LTRMP fish data from Pools 8, 13, and 26 of the Mississippi River and LaGrange Pool of the Illinois River into unique combinations of sampling gear, study reach, habitat class, and time period. Each partition consisted of a unique combination of gear, study reach, habitat class, and time period. Only partitions containing at least two samples from each of at least two distinct sites were retained for further analysis. This selection ensured that both among- and within-site variance could be estimated. In the original LTRMP design, sites were vaguely defined areas containing two subareas, subjectively deemed similar, that were intended to serve as within-site replicates. For

each combination of study reach, sampling gear, and habitat class, I retained data from species having the three highest mean catch rates. Data for black crappie, channel catfish, sauger, and walleye also were included. These criteria ensured that species of high economic value were represented and that subsequent analyses were not unduly influenced by samples having catches of zero fish. This data partitioning and selection procedure produced 249 combinations of species and data partition.

I used 4'th roots of catch-per-unit-effort (C/f) as the response variable. This 4'th root transformation is a member of the Box-Cox family of power transformations given by $y^{0.25}$ (Box and Cox 1964). No single transformation is universally successful in yielding a symmetrically distributed variable from C/f derived from different species, populations, and times. However, the 4'th root transformation is fairly robust for highly skewed catch data.

I estimated variance components of the transformed C/f data from each combination of species and data partition using the minimum variance quadratic unbiased estimation (MINQUE) method (Rao 1973). I estimated variance due to the among-site and the within-site effects. The MINQUE method produces unbiased estimates of variance components, but sometimes produces negative estimates which contradict the principle that variance can never be less than zero. Although treating negative variance components as if they were zero is common practice, I took a more conservative approach and deleted all combinations of species and data partition yielding a negative variance estimate. A total of 103 of the 242 combinations yielded among-site variance component estimates less than zero. Elimination of these 103 combinations was conservative in that elimination actually favors the supposition that among-site variance was greater than within-site variance. All further analyses were based on the remaining 139 combinations of species and data partition yielding non-negative variance estimates.

I examined two null hypotheses of sources of sampling variation in the 1990 LTRMP fish data. First, I tested the null hypothesis that among-site variance is less than or equal to within-site variance in at least 50% of the combinations of species and data partitions. Expected among-site variance was less than expected within-site variance in at least 50% of the combinations of species and data partition (Fig. 1). I tested the first hypotheses using a one-sided nonparametric sign test (Daniel 1978) on the paired differences between among-site and within-site variance. The null hypothesis that among-site variance was less than or equal to within-site variance in at least 50% of the combinations of species and data partitions could not be rejected for any gear or for all gears combined (Table 1). No evidence in the 1990 LTRMP fish data exists that among-site variance is more frequently greater than within-site variance. Second, I tested the null hypothesis that medians of paired differences between among- and within-site variances were less than or equal to zero using the one-sided Wilcoxon matched-pairs signed-rank test (Daniel 1978). The null hypothesis could not be rejected in favor of the alternative that median differences between among- and within-site variances are greater than zero (Table 1). No evidence in the 1990 LTRMP fish data exists that among-site variance is greater than within-site variance.

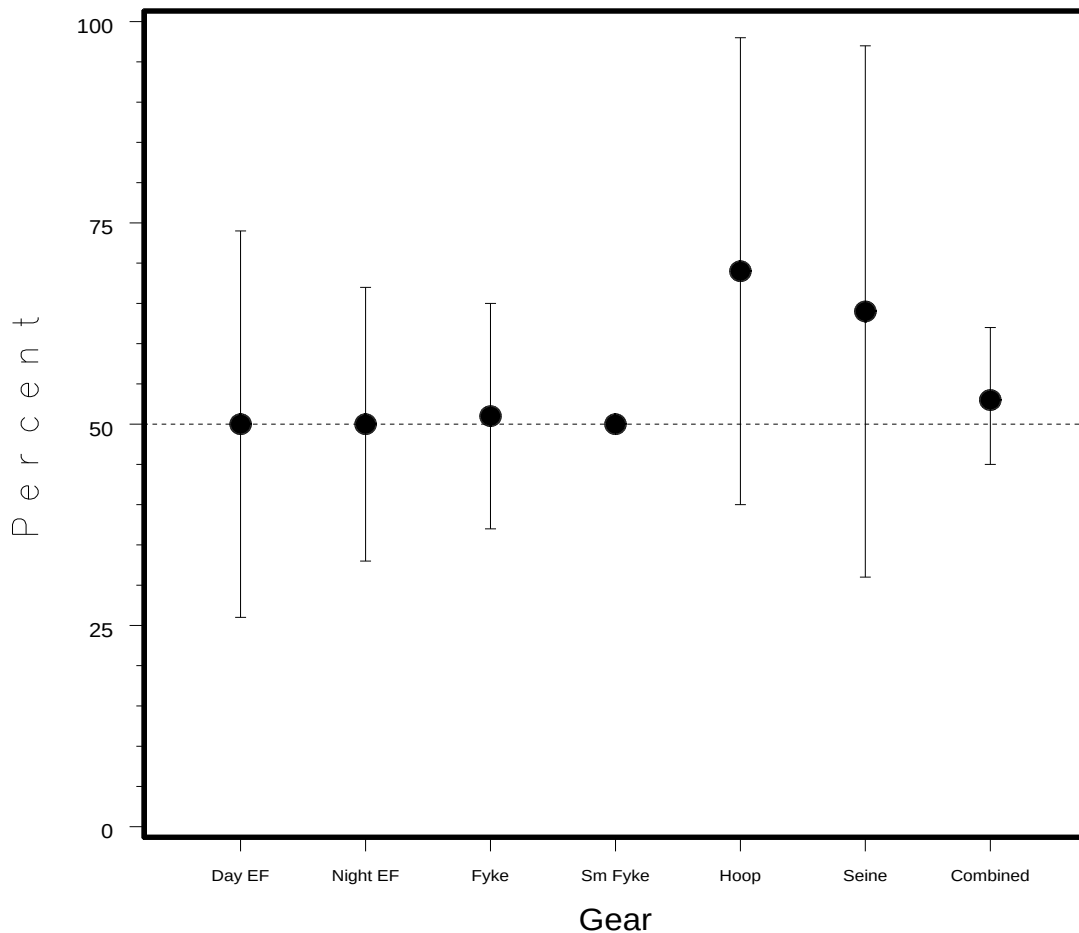


Figure 1. Percentages of combinations of species and data partitions, based on unique combinations of study reach, habitat class, and time period, in which among-site variation is less than or equal to within-site variation. The upper and lower bounds are 95% confidence intervals. Electrofishing is denoted EF; other gears are nets.

These analyses provide compelling evidence that the supposition that among-site variance is greater than within-site variance is not true in the LTRMP study reaches. Given that the statistically compromised fixed-site LTRMP fish sampling design is predicated on this suppositional truth, these results leave no reason, other than logistical constraints and perhaps monitoring threatened and endangered fishes, to continue monitoring under this design.

Table 1. Results of nonparametric sign and Wilcoxon matched-pairs signed-rank tests of differences between among- and within-site variance components in the 1990 LTRMP fish data. See text for explanation of tests .

Sampling gear	<i>P</i> -value		
	<i>N</i>	Sign	Wilcoxon
Day electrofishing	20	0.50	0.60
Night electrofishing	40	0.50	0.19
Fyke netting	53	0.39	0.07 ^a
Hoop netting	13	0.63	0.59
Seining	11	.77	0.95
All gears combined	139 ^b	0.87	0.60

^a Near significance due to one large negative difference, without which $P > 0.10$.

^b Includes two combinations from minnow fyke nets that could not produce meaningful tests because of small *N*.

The Stratified Random Sampling Alternative

Description

Stratified random sampling (Green 1976) is the statistically valid alternative to LTRMP fixed-site sampling. In these designs, the whole sampling unit is divided into relatively homogeneous classes called strata. Stratification reduces sampling variance whenever less variation (greater homogeneity) exists within strata than in the whole sampling unit. Each LTRMP study reach can be considered a whole spatial unit. However, each study reach contains several habitat types characterized by different fish communities. Therefore, habitat types are logical strata. In stratified random sampling, a randomly selected set of primary sampling units is sampled within each stratum. Random selection is conducted independently for each time period. In the LTRMP sampling program, spatially defined cells or sites can serve as primary sampling units. Randomization within strata ensures that, except for selectivity of sampling gear, results from every stratum are unbiased (representative of the stratum), enabling the monitoring of trends over whole strata and study reaches.

Logistical Considerations

Because stratified random sampling requires visiting many different sites during sampling operations, navigational limitations and other logistical constraints must be considered. Staff from the Environmental Management Technical Center (EMTC) and the Onalaska Field Station explored two areas of Pool 8 considered to be difficult site locations. We chose the Goose Island complex of backwaters and channels because it is probably the most complicated maze of islands and channels among the LTRMP study reaches. We chose the Stoddard stump field

because it is a large, open, almost featureless expanse in the lower pool. Other LTRMP study reaches may not contain areas where site location would be as difficult.

We attempted to find 12 randomly selected sites on June 10-11, 1992. The site locations in the Goose Island complex had been plotted on a base map showing land cover types. Site locations in the Stoddard stump field were plotted on a base map showing depth contours. We attempted to find each site by comparing physical features observed from the boat with representations on the base maps. When we believed we found a site, we anchored the boat and determined our true position, to within 100 m, using a Magellan Global Positioning System (GPS; Magellan Systems Corp., Monrovia, CA). Our attempts to find sites were defined as successful if the position obtained from the GPS was within 100 m of the location defined on the base maps. When an attempt was unsuccessful, we made a second attempt. If a third attempt was needed, we used the GPS in continuous mode to navigate to the site.

We found four of six sites in the Stoddard stump field on the first attempt. The remaining two sites required a second attempt; both attempts proved to be less than 200 m from our original guess. We successfully found three of four sites in the Goose Island complex. The fourth site required three attempts, but we also determined that one experienced crew member correctly identified this site during the initial approach to the general area.

Experienced river boaters should be able to locate, to within 100 m, most randomly selected sites using only base maps. For sites proving inaccessible, sampling the nearest possible location within the stratum and recording that secondary location as the sampling site should suffice. Randomized sampling may present some new challenges to the LTRMP sampling crews, but stratified random sampling has been shown to be practical in our exploratory work in Pool 8 and in the LTRMP invertebrate sampling program.

Recommended Sampling Plan

Given the inadequacy of the LTRMP fixed-site design and the necessity and practicality of the stratified random sampling alternative, it is imperative that the LTRMP implement stratified random fish sampling beginning with the 1993 field season. I recommend spatially randomized sampling from five strata: (1) main channel trough, (2) main channel border, (3) secondary and tertiary channel border, (4) contiguous backwaters, and (5) impounded areas. These strata are compatible with both the existing LTRMP habitat classes and the Cobb-Wilcox aquatic areas classification scheme (Wilcox 1993). Historically, the LTRMP has not routinely sampled fish in secondary and tertiary channels, so this stratum is new. Also, the LTRMP has traditionally distinguished vegetated versus unvegetated backwaters and impounded areas. The original designation of vegetation status was made early in the program. Vegetation has proven to be a transient feature; some fixed sites that were vegetated in 1989 no longer are, and some that were open in 1989 are now vegetated. Status of vegetation seems best recorded at the time of sampling. Decisions regarding sampling vegetated and open sites should be agreed upon by EMTC and field station staff.

Only two reasons exist for retaining some fixed sampling sites. First, the LTRMP has been successful in detecting threatened or endangered fishes at a few sites. Given the difficulty

in detecting rare fishes, occurrence at a specific site is evidence that those sites may provide critical habitat. Therefore, stratified random sampling should be augmented with specialized sampling at sites in which LTRMP sampling previously documented the occurrence of threatened or endangered fishes. Second, the LTRMP should sample fishes at fixed sites in the tailwaters of locks and dams because tailwaters are small, unique habitat types.

A stratified random sampling design, augmented with relatively few fixed sites in tailwaters and where rare species have been encountered, was implemented at the start of the 1993 field season. This design is described in Gutreuter et al. (1993).

Sampling Intensity and Statistical Power

Sampling intensity (sample size) is an important statistical issue, but far less so than is randomization. Traditionally, an estimate of adequate sample size for some particular random variable is computed from a preliminary estimate of its variance. Two limitations to the meaningful application of this approach occur in the LTRMP Fisheries Component.

First, the LTRMP Fisheries Component is a broad-spectrum effort in which many species are monitored. For example, during 1990, the LTRMP collected 76 species from six study reaches (Gutreuter 1992). Further, several attributes of fish populations and communities are monitored. For all species, relative abundance is monitored, as are length distributions when possible. Further, aspects of fish community structure such as species richness are also of interest. Therefore, the LTRMP is responsible for monitoring not one or a few random variables, but upwards of 100 of them. A (likely different) estimate of adequate sample size could be computed for each of these random variables. The ultimate sampling intensity choice would rest on a subjective decision of which of these many random variables is most important to the LTRMP, a task for which consensus-building would be difficult.

Second, sample size estimation assumes existence of a statistically valid sampling design and an accurate estimate of variance. Historical (prior to 1993) data were collected under a fixed-site sampling design that did not support valid statistical inference, including sample size estimation, over entire habitat classes or study reaches. Sample size estimation is very sensitive to the estimates of among-site variances within sampling strata. Although the above analyses of variance components suggest randomization will not produce estimates of among-site variance greater than within-site variance estimates from the fixed-site sampling program, it is prudent to defer any estimation of adequate sample size until variance estimates are obtained from the statistically valid randomized sampling initiated in 1993.

Despite these limitations of traditional sample size estimation, the LTRMP has developed information useful in assessing the adequacy of the historical intensity of fish sampling. Systemic analyses of the 1990 LTRMP fish data (Gutreuter 1992) demonstrate that historical levels of sampling intensity are adequate in detecting among-habitat and among-reach difference in the relative abundances and length distributions of important species, as well as the overall community structure of relatively common species. Barring distortions resulting from extreme serial (temporal) correlation, these preliminary analyses, coupled with the results of the variance component analyses discussed above, suggest that the historical sampling intensity is sufficient to detect trend effects at least as large as the spatial differences observed in the 1990 data. That

is, current available information suggests that the LTRMP will be effective in detecting trends in the relative abundance, length distribution, and community structure of fishes in the Upper Mississippi River System. Data from several years following the change to stratified random sampling will be needed to confirm this hypothesis.

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