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A MODIFIED FOLSOM PLANKTON SPLITTER FOR ANALYSIS OF METER NET SAMPLES[1]

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ABSTRACT

The large number of meter net samples needed to determine the spawning success of various species of fish in Canton Reservoir, Oklahoma, made subsampling advantageous. The basic Folsom plankton splitter was enlarged and modified so that meter net samples with volumes up to 4,000 ml. could be split into 10 approximately equal subsamples.

The splitter was constructed from a 12 inch diameter Plexiglas[2] cylinder. Construction was accomplished using common shop tools.

Chi-square tests (0.05 level) showed that there were no significant differences between the observed subsample counts and the expected counts. A nonparametric sign test showed that each chamber did not consistently have higher or lower counts than any other chamber.

The minimum total number of organisms per sample that could be subsampled yielding estimates of the total sample number with less than a 10 percent error 95 percent of the time were determined for larval gizzard shad and larval Chaoborinae. Determination of the minimum number of organisms per sample needed for subsampling other organisms can be completed as necessary.

INTRODUCTION

The use of meter net samples to determine the 1968 spawning success of various species of fish in Canton Reservoir, Oklahoma, required an estimated 1,200 manhours to sort and enumerate. The average volume of a standard 5 minute haul was 800 ml. and over 300,000 ml. were collected. Therefore, a subsampling method was desirable that would save time while producing similar results.

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[2]Registered trademark.

Several of the standard subsampling methods and separation techniques did not seem applicable to meter net samples. Flotation as described by Anderson (1959) and Whitehouse (1966) would not reduce enumeration time as flotation is not a means of subsampling. Separation of meter net samples by flotation would be difficult. Zooplankters make up the bulk of most samples and they would float along with the larval fish and insects. Small, vertical tube splitters as that described by Cushing (1961) would not handle the larger meter net samples and seemed difficult to enlarge. The rotating splitter described by Waters (1969) had merit but the zooplankters probably would have clogged the screen. Any screen small enough to stop the larval fish would also stop zooplankters.

The Folsom plankton splitter was enlarged and modified to split up to a 4,000 ml. sample into 10 subsamples. The reliability of the original Folsom plankton splitter was proven by McEwen (1954) and it was assumed from the work done by Scarola (1968) with a four chambered sampler that a ten chambered splitter would be reliable. Scarola found the distributional differences in organism counts among the four chambers to be non-significant at the 0.05 level using a chi-square test.

An evaluation of this 10-chambered Folsom plankton splitter was made to determine the reliability of the splitter on some of the more common organisms found in the meter net samples: larval gizzard shad *Dorosoma cepedianum* (Lesueur) and larval and pupal Chaoborinae. All larval fish except gizzard shad occurred in such low numbers that they could not be subsampled and therefore not used in this evaluation.

Determinations were made of the minimum total number of organisms that could be reliably subsampled at 10%, 20% and 50% subsampling levels. Reliable subsampling for use here meant an estimate of the total number of organisms with less than a 10 percent error 95 percent of the time. This was done for each individual organism when it became advantageous to subsample that particular organism.

The samples used to evaluate the splitter were actual meter net samples collected during 1969 at Canton Reservoir, Oklahoma. These samples covered the range of expected variations in sample volumes and compositions. It was therefore possible to determine if the subsampler would work for all possible meter net samples.

MATERIALS AND METHODS

The splitter shown in Figure 1 was constructed from a Plexiglas cylinder, 12 inches in diameter and 11 ⅜ inches long. The basic measurements are shown in Figure 2. Construction was completed using common shop tools.

All Plexiglas used in the construction of the splitting unit was ⅛ inch thick. Care was taken in fitting and cementing the chamber partitions to the inside surface of the cylinder to insure that they would be exactly one inch apart. The internal seams were sealed with a clear silicone rubber to prevent leakage. The leading edge of each partition was filed to a 45° point to facilitate splitting. Individual subsamples were removed through drains of ⅝ inch I. D. Plexiglas tubes mounted on the bottom of the cylinder. The tubes were sealed with corks.

The center support tube (0.750 O. D. and 0.375 I. D.) was added to strengthen the internal partitions and take some of the weight off the end plates. A 5/16 inch dowel rod was used as the axle. To prevent the cylinder from rotating when in use a sliding safety pin was added.

The splitter was mounted on a ¼ inch Plexiglas frame which gave clearance for removal of subsamples. This Plexiglas frame was then attached to a ½ inch, plywood base with leveling screws.

The splitter was statistically evaluated to determine the reliability of sub-

sampling. Chi-square tests were used to determine if observed subsample counts were significantly different from expected. A nonparametric sign test was used to determine if any one chamber had a consistently higher or lower count than any other chamber. A nonparametric statistic was used because the great variation in sample sizes would have masked any significant differences with analysis by parametric statistics.

To determine the minimum total number of organisms per sample that could be reliably subsampled it was assumed that at each of the subsampling percentages (10%, 20%, and 50%) there was an error caused by the subsampling process alone. It was assumed that this error was caused by the randomness of splitting and was not dependent on the total number of organisms with set ranges. This error was found by calculating all the actual possible errors that would have been made if each subsample within a set range of total number of organisms had been used to estimate the total number of organisms per sample. A standard

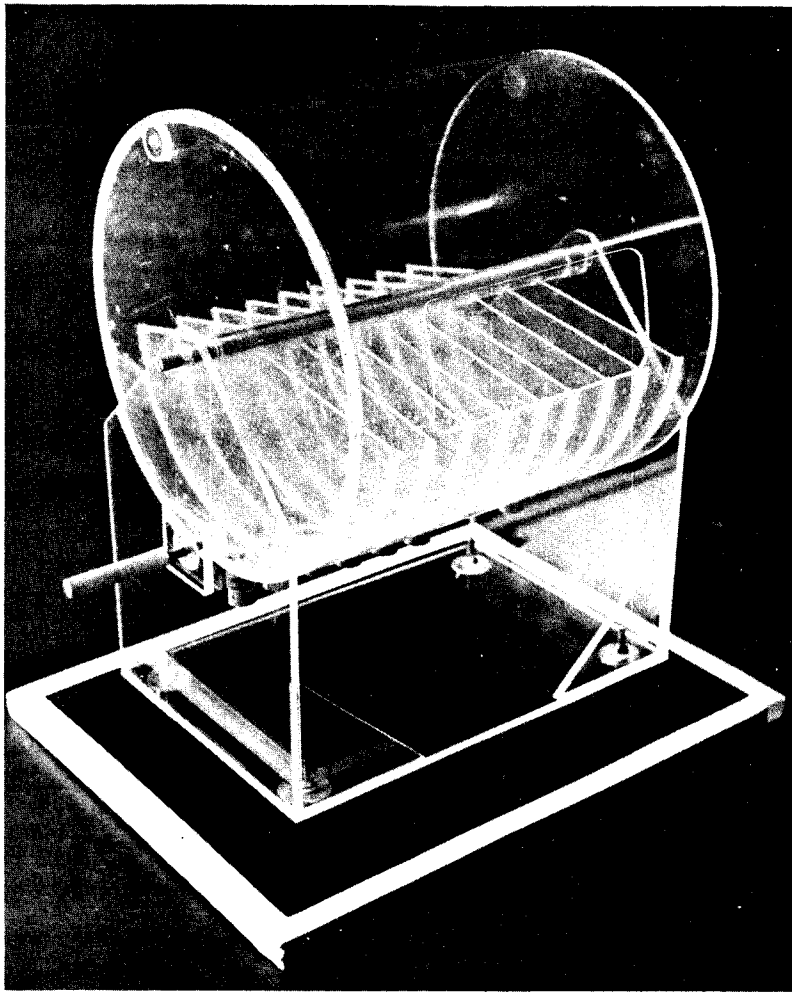

FIGURE 1. The modified Folsom plankton splitter showing the 10 chambers.

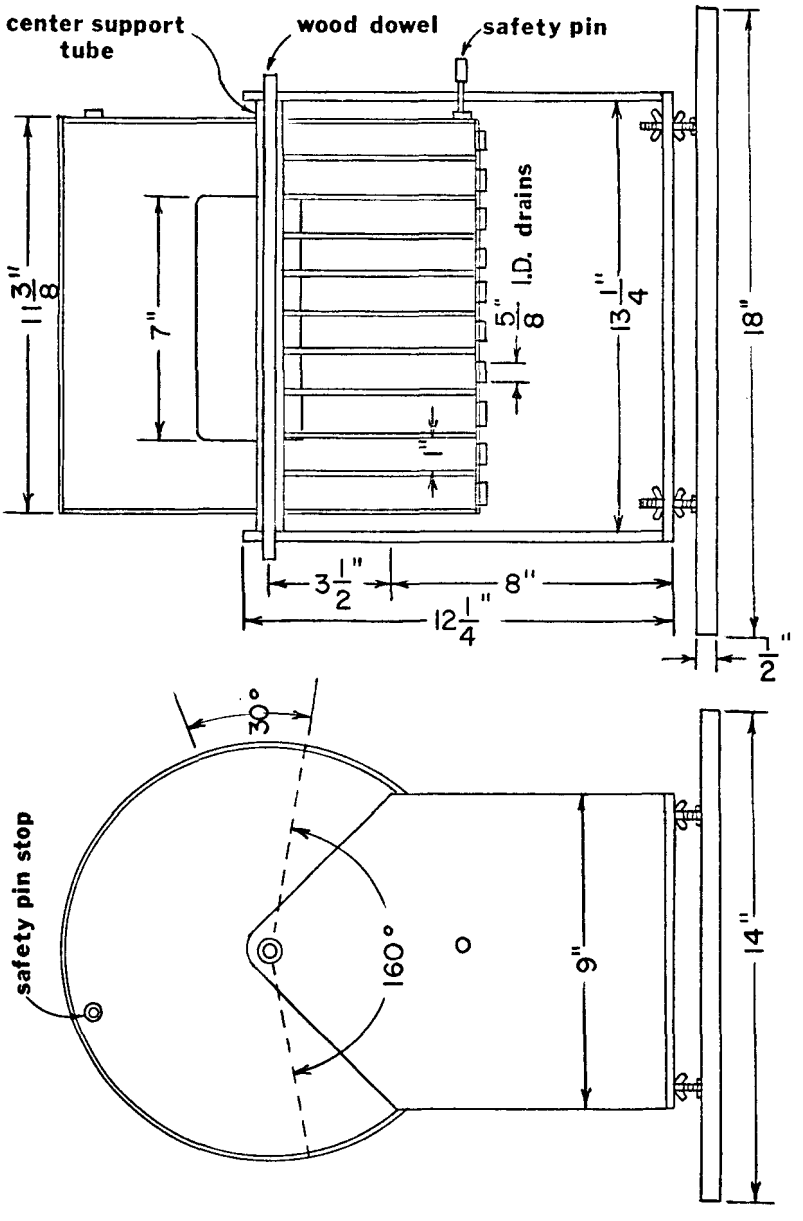

FIGURE 2. Detail drawings of modified Folsom plankton splitter with basic measurements, end view, and logitudinal section through the center.

deviation was then calculated and a 95 percent confidence interval for expected errors determined for that range of total number of organisms. If the upper limit of this confidence interval, using a one sided probability, was just less than 10 percent of the smallest total number of organisms within the set range it was assumed for our puposes that the lower limit of this set range was the minimum total number of organisms that could be subsampled. The range limits of total number of organisms were changed until the above relationships were found for each of the subsampling percentages and for each organism. Had there been an adequate number of samples with approximately the same total number of organisms standard deviations and confidence limits could have been placed on the estimated total numbers and not the errors. The total number of organisms per samples used here ran between 200 and 10,000. For simplicity of analysis at the 20% and 50% subsampling level adjacent chambers were used in place of all the possible combinations.

RESULTS

Preliminary evaluation of the splitter showed that the reliability of subsampling was adversely affected by the small settled volumes, larval fish over ¾ of an inch long, and lack of agitation prior to splitting. When the settled volume was less than 500 ml. it was necessary to increase the volume by the addition of zooplankters to obtain reliable samples. The zooplankters which were added came from previously worked samples and contained no countable organisms. With the removal of larval fish over ¾ of an inch, reliable estimates of the total number of organisms were obtained. Prior to splitting vigorous agitation of the samples with a glass rod in a noncircular pattern increased the subsampling reliability.

Chi-square values for the expected number of larval shad per subsample and the actual counts showed no significant differences at any of the subsampling levels. The total number of larval shad per sample ranged between 3,120 and 242. At the 10% subsample level chi-square values ranged between 0.788 and 4.852 with 9 degrees of freedom. At the 20% subsample level chi-square values ranged between 0.309 and 2.164 with 4 degrees of freedom. At the 50% subsample level chi-square values ranged between 0.001 and 0.226 with 1 degree of freedom. Larval and pupal Chaoborinae had similar nonsignificant chi-square value ranges at the 10% subsample level, 0.125 to 15.739 and 1.988 to 12,885 respectively. The total number of organisms per sample ranged from 10,360 to 220 for the larvae and 3,923 to 217 for the pupae. The nonsignificantly different chi-square values show that the subsamples are approximately equal and approach the 1 : 1 : . . . 1 ratio.

Nonparametric sign tests of the larval gizzard shad at the three subsampling levels showed that no one chamber had continuously higher or lower counts than any other chamber. Probabilities ranged between 0.14 and 0.67 at the 10% subsampling level, 0.090 and 70.212 at the 20% level and 70.133 at the 50% level. Nonparametric sign tests of the larval and pupal Chaoborinae counts at the 10% level showed no significantly higher or lower counts for any one chamber. These nonparametric sign tests showed that the variations between subsample counts were random.

The standard deviation of the error encountered when taking a 10% subsample of larval gizzard shad was 30.17 for samples with a total number of 1000 or more. The upper confidence limit using a one sided probability was 94.24. This indicates that the minimum total number of organisms that can reliably be subsampled at the 10% level is 1000. At this sample size estimates would not vary more than 9.4% from the actual value with a 95% reliability. The minimum total number of organisms that could be subsampled with a 20% or 50% subsample were 500 and 200 respectively. At a minimum of 500 for the 20% subsample level estimates of the total number would not vary more than

7.7% with a 95% reliability. At the 200 minimum for 50% subsamples the estimated totals would not vary more than 4.4% from the actual total with a 95% reliability. This meant that if the first chamber counted contained 100 or more shad it was a reliable estimate of the total number when multiplied by 10. If there was less than 100 but more than 50 one additional subsample was needed to gain a reliable estimate. If there was less than 50 but more than 20 only half the subsamples had to be counted. If there was less than 20 all the subsamples had to be counted.

For larval Chaoborinae a minimum total number of 1000 was needed for subsampling at the 10% level. Estimates of the total number of organisms at this minimum varied less than 6.8% from the actual number with a 95% reliability. The minimum total number of larva that can be subsampled at the 20% and 50% subsampling levels will be determined as needed. Minimums will also be set for the Chaoborinae pupae as needed.

CONCLUSIONS

Modifying the Folsom plankton splitter so that it would hold 4,000 ml. meter net samples and split them into 10 approximately equal subsamples did not reduce its subsampling reliability. Chi-square tests and nonparametric sign tests have shown that subsample counts are random and not significantly different.

In order to obtain reliable subsamples with this subsampler the settled volume had to be over 500 ml. The removal of larval fish over ¾ of an inch and noncircular agitation were also required for reliable subsampling.

Subsampling at the different percentage levels with set minimums permitted reliable estimates to be obtained with the least amount of effort. If the number of larval shad or larval Chaoborinae is over 100 in the first subsample they do not have to be counted in any of the remaining subsamples. If the number of shad ran between 99 and 50 then only one more subsample had to be counted. If the number of shad ran between 49 and 20 four more subsamples were needed to obtain an accurate estimate. With the subsampling percentages and subsampling minimum numbers being changeable it will be possible to reliably subsample other organisms as needed.

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