

## Evaluating Sampling Strategies for Larval Cisco (*Coregonus artedi*)

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**ABSTRACT.** To improve our ability to assess larval cisco (*Coregonus artedi*) populations in Lake Superior, we conducted a study to compare several sampling strategies. First, we compared density estimates of larval cisco concurrently captured in surface waters with a 2 × 1-m paired neuston net and a 0.5-m (diameter) conical net. Density estimates obtained from the two gear types were not significantly different, suggesting that the conical net is a reasonable alternative to the more cumbersome and costly neuston net. Next, we assessed the effect of tow pattern (sinusoidal versus straight tows) to examine if propeller wash affected larval density. We found no effect of propeller wash on the catchability of larval cisco. Given the availability of global positioning systems, we recommend sampling larval cisco using straight tows to simplify protocols and facilitate straightforward measurements of volume filtered. Finally, we investigated potential trends in larval cisco density estimates by sampling four time periods during the light period of a day at individual sites. Our results indicate no significant trends in larval density estimates during the day. We conclude estimates of larval cisco density across space are not confounded by time at a daily timescale. Well-designed, cost effective surveys of larval cisco abundance will help to further our understanding of this important Great Lakes forage species.

**INDEX WORDS:** Cisco, *Coregonus artedi*, recruitment, survey, gear comparison, Lake Superior.

### INTRODUCTION

The larval stage of cisco (*Coregonus artedi*) development is characterized by a pelagic existence (Anderson and Smith 1971, Selgeby *et al.* 1978, Hatch and Underhill 1988, Oyadomari and Auer 2004), where a host of physical and biotic influences may act to limit recruitment (Roughgarden *et al.* 1988, Heath 1992). Research has shown that er-

atic fluctuations in adult abundance of many fish species, like those observed for Lake Superior cisco (Yule *et al.* 2008), are often the result of extremely high mortality rates and variable survivorship during the larval phase (Ricker 1954, Houde 1987). Many mechanisms have been proposed to explain variations in recruitment, but the relative importance of alternative mechanisms remains poorly understood (Cushing 1996). Development of a sampling program that effectively captures the larval stage is a promising avenue for estimating important population metrics and investigating hypotheses concerning recruitment (Sammons and

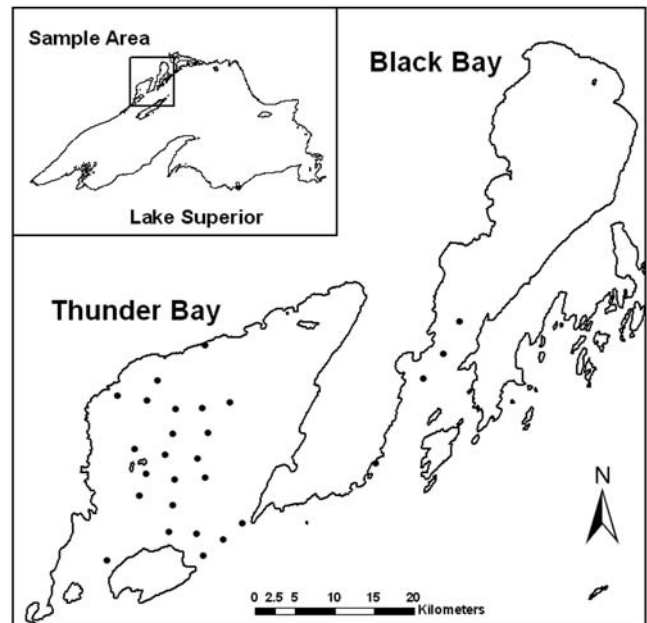
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Bettoli 1998, Castro and Hernandez 2000, Karjalainen *et al.* 2000).

Reliable assessment techniques are critical so that attempts to understand complex ecological processes are not undermined by poor survey design. Net avoidance is a serious consideration when attempting to measure the abundance of agile planktonic organisms (Barkley 1972). If cisco larvae exhibit an escape response, nets with a smaller effective sampling area would be subject to conservative estimates of density compared to more encompassing gears. Also, it would be expected that smaller nets would be more sensitive to the patchiness of larval cisco (Oyadomari and Auer 2004) while larger framed nets would reduce variability caused by fine-scale patchiness. Recent studies have suggested that larval fish may react to the turbulence induced by a vessel's propeller (Claramunt *et al.* 2005, Overton and Rulifson 2007). This creates a potential for differences in estimated density due to the pattern in which the gear is towed and the time spent directly in the boat wash. An added consideration is that samples within a survey are confounded by the inherent limitation of sampling over both space and time simultaneously (Levin 1992). Researchers have found that coregonid larvae in Finnish lakes are aggregated near the surface during daytime, with maximum catches occurring at approximately 1100–1400 hours (Viljanen *et al.* 1995). In Lake Superior it has been demonstrated that larval cisco density is generally highest in the surface stratum (Selgeby *et al.* 1978, Hatch and Underhill 1988) during daytime (Oyadomari and Auer 2004), yet no study has investigated the repeatability of estimates through this period.

In this study we compare the performance of two gears for estimating larval cisco abundance and make recommendations for survey tactics for this important Great Lakes species. The present work was part of a larger study to investigate the impact of rainbow smelt predation on larval cisco in Thunder Bay and Black Bay, Ontario. Sampling of both bays in a timely fashion required us to collect larval cisco using two vessels equipped with different gears. We assessed the performance of the two gear types (a  $2 \times 1$ -m paired neuston net and a 0.5-m [diameter] conical net) to determine whether density and mean length estimates from the smaller conical net were comparable to those estimated from the neuston net. We also investigated whether propeller wash influenced density and mean length estimates by comparing samples collected off the stern of vessels using sinusoidal and straight tow patterns.



**FIG. 1.** Stations used to evaluate sampling strategies for larval cisco.

Finally, we examined potential trends in density estimates from early morning to late afternoon to examine if time of day could be ignored when generating mean density estimates across sites within a day.

## METHODS

Data for this study were collected in Thunder Bay and Black Bay, Ontario (Fig. 1) during 2–25 May 2006. Two vessels were used to collect samples. The first vessel was a 7.9-m Bertram with twin, 3.6 liter, stern-driven, Mercruiser engines and was equipped with a hydraulic winch. Larval fishes were sampled from this vessel with a  $2 \times 1$ -m paired neuston net (hereafter referred to as “neuston net”) towed in the surface stratum. The second vessel was a 6-m Boston Whaler with twin 150 horsepower Evinrude E-TEC outboard motors. Larval fishes were sampled with a 0.5-m (diameter) conical net (hereafter referred to as “conical net”) towed in the upper 1 m of the water column from this vessel. Both gears were equipped with 500- $\mu$ m mesh nets.

A single sample consisted of a 5-min tow in the 0–1 m stratum. Nets were deployed behind each vessel with approximately 30 m of warp line out. During sampling, each vessel weaved in a sinusoidal pattern to minimize towing time in the re-

spective vessels' wash. The exception to this protocol was when testing for the effects of propeller wash, in which one sample was towed in a sinusoidal manner while the other paired sample (e.g., from the same vessel) was collected using a straight pull. To maintain consistency, all tows were pulled with the wind on the stern at a speed of 3.2 to 3.9 km/hr. The volume of water filtered was determined using General Oceanics model 2030 flowmeters. Both collections from the paired neuston net were pooled for a single sample. All specimens were preserved in 95% ethyl alcohol.

Larval lake whitefish (*Coregonus clupeaformis*) were distinguishable from other larval coregonids based on size (Hinrichs 1979) and melanophore patterns across the dorsum (Hinrichs 1979, Auer 1982). Differentiating larval cisco from bloater (*Coregonus hoyi*) and kiyi (*Coregonus kiyi*) is not possible with visual observation (Anderson and Smith 1971, Hinrichs 1979). Because bloater are believed to spawn later than cisco (Hinrichs 1979, Auer 1982), we assumed that emergence of cisco was separated temporally from the emergence of bloater. Kiyi are not known to spawn in Thunder Bay and Black Bay (Ken Cullis, Ontario Ministry of Natural Resources, personal communication). Additionally, the maximum bathymetric depth in Thunder Bay, the deeper of the two bays, is 91 m and the spawning habitat of kiyi is reported to be 91 to 168 m depth (Scott and Crossman 1998). Thus, all emergent coregonids collected in May 2006 were identified as cisco or lake whitefish.

All larval coregonids were counted for each sample. When samples contained  $\leq 100$  larval fish all specimens were identified as cisco or lake whitefish and photographed using a Leica S6D dissecting scope equipped with a Panasonic digital camera. When samples contained  $>100$  individuals a random sample of 50 specimens was identified as cisco or lake whitefish and photographed. Photographs were used to digitally measure total length of each fish to the nearest 0.1 mm. Density of larval cisco (#/1,000 m<sup>3</sup> of water) in each sample was calculated by dividing number caught by volume of water filtered. We assumed net efficiency was 100%. All statistical tests were performed using the statistical software SAS (SAS Institute Inc. 2003). Our significance level for all analyses was  $\alpha = 0.05$ .

### Gear Comparison

To compare density estimates and length measurements between the two gears, we collected

samples using side-by-side tows with the two vessels. The vessels were separated by approximately 50 m. Coordination by way of VHF radio allowed for synchrony in gear deployment and tow duration. To satisfy the assumption of normally distributed errors, density estimates were log<sub>10</sub>-transformed prior to analysis. A paired *t*-test was used to test the null hypothesis that there was no difference between density estimates from the two gears. For length comparisons we used a Kolmogorov-Smirnov test to test the null hypothesis that there was no difference between the distributions of preserved lengths from the two gears.

### Tow Pattern

To examine the effect of turbulence caused by the propellers on density estimates and average preserved length, paired samples were collected successively at various sites. One sample of each pair was collected using a sinusoidal pattern and the other sample was collected using a straight pull. A single gear was used for sampling a specific site yet both vessels (and the respective gears of each vessel) were used within this analysis. The sequence of collections at a specific site was determined randomly. Both tows began at approximately the same starting position yet the respective transects were not overlapped. Collection of both samples took approximately 25 minutes. To satisfy assumptions of normality we applied a log<sub>10</sub>-transformation, with a small constant added to the one zero observation (Johnson and Rauser 1971). Mosteller and Tukey (1977) recommended adding a constant that is 1/6 the minimum observable value, leading us to use a constant of 3 (i.e., with a single larvae captured by the conical net the minimum density estimate was 18 larval cisco / 1,000 m<sup>3</sup>). A paired *t*-test was used to assess the null hypothesis of no differences between density estimates derived from sinusoidal and straight tow patterns. A Kolmogorov-Smirnov test was used to test for differences in the length frequency distributions resulting from the pattern in which the net was towed.

### Daytime Trends

To investigate potential patterns in larval cisco density estimates during light hours, we sampled a site during four time periods over the course of a day (approximately 0600, 1000, 1400, 1800). Within each time period, three consecutive samples were collected using the neuston net. This strategy

**TABLE 1.** The number of samples collected, number of coregonid larvae captured, and the mean density (#/1,000 m<sup>3</sup>) of cisco larvae sampled for the evaluation of sampling strategies in Thunder Bay and Black Bay, Ontario, during 2–25 May 2006.

	Gear Comparison		Tow Pattern		Daytime Trends
	Neuston	Conical	Sinusoidal	Straight	Neuston
No. Samples	28	28	20	20	66
No. Whitefish Larvae	2	0	5	0	17
No. Cisco Larvae	8,088	1,089	5,145	242	18,013
Mean Cisco Density ( $\pm$ SE)	474 (98)	672 (212)	299 (75)	331 (110)	453 (69)

was replicated for a total of 6 days (six different sites) throughout the study period. A log<sub>10</sub>-transformation was applied to the density estimates prior to analysis to stabilize variances and better meet assumptions of normality. A repeated measures ANOVA was used to test the null hypothesis that there were no temporal trends within the light period of an individual day. Because the model is incapable of handling missing data we performed the analysis twice. In the first analysis we used only the days in which a complete set of 12 samples were collected ( $n = 3$ ). In the second analysis we used only the first sample from each time period, thereby increasing the sample size ( $n = 5$ ). On 5 May 2006 rough seas prevented the collection of all late afternoon samples, causing us to exclude this day from both analyses.

## RESULTS

A total of 162 ichthyoplankton samples were collected to meet the objectives of this study. We collected 114 samples using the neuston net and 48 samples using the conical net. We captured 31,246 and 1,331 cisco larvae with the neuston and conical nets, respectively. Very few lake whitefish larvae were identified during the study. Rainbow smelt larvae were observed in the samples but were not counted. The number of samples collected and coregonid larvae counted for the three respective analyses are summarized in Table 1.

### Gear Comparison

A total of 28 side-by-side tows were made with the two gears. Density estimates ranged between 50 and 2,317 larvae/1,000 m<sup>3</sup> for the neuston net and 33 and 4,651 larvae/1,000 m<sup>3</sup> for the conical net. Density estimates were not significantly different between the two gears ( $t$ -value =  $-0.473$ ,  $P = 0.64$ ,  $DF = 27$ ; Fig. 2A). Given the observed variance for

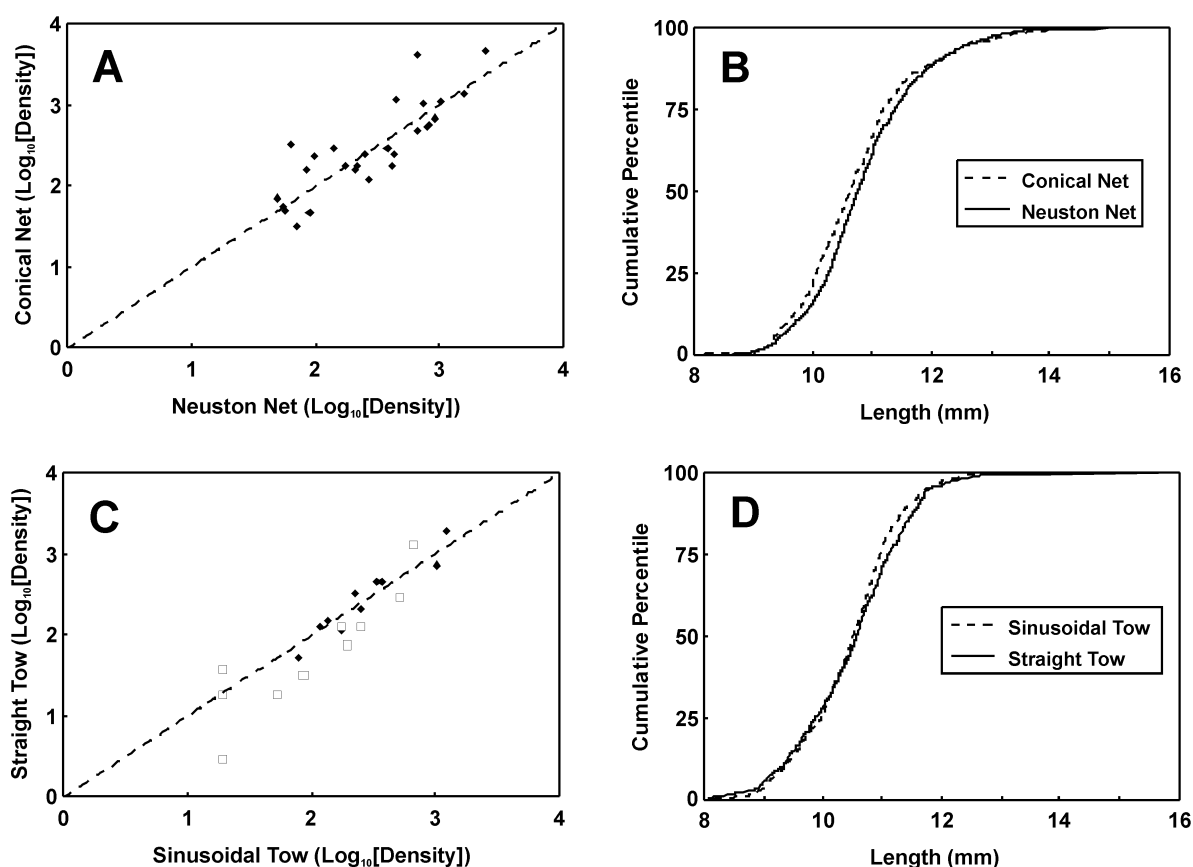
the 28 paired collections we would have been able to detect an effect size of 1.46 with 80% power (Lenth 2006). Because we are ultimately interested in mechanisms that cause orders of magnitude differences in abundance, we feel that a difference between gears of less than 50% is not meaningful for larval cisco assessments and our sampling effort was appropriate for the objectives of this investigation. Cisco larvae caught using the neuston net had a mean length of 10.85 mm (SE = 0.026,  $n = 1,389$ ) and those caught by the conical net had a mean length of 10.73 mm (SE = 0.042,  $n = 548$ ). The distribution of larval cisco lengths were significantly different between the two respective gears (K-S test:  $D = 0.094$ ,  $P < 0.005$ ) although the differences in lengths captured were very small (Fig. 2B).

### Tow Pattern

We collected 10 paired samples using each respective net for a total of 20 comparisons. Because we found no difference between the two gears we combined data from both nets for one analysis examining the effect of propeller wash. We found no significant difference in density estimates based on tow pattern ( $t$ -value = 1.715,  $P = 0.104$ ,  $DF = 19$ ; Fig. 2C). Cisco larvae captured using the sinusoidal tow pattern had a mean length of 10.45 mm (SE = 0.031,  $n = 688$ ) and those caught using a straight tow pattern had a mean length of 10.50 mm (SE = 0.035,  $n = 677$ ). The Kolmogorov-Smirnov test revealed a significant difference between the length frequency distributions resulting from the alternative tow patterns ( $D = 0.078$ ,  $P < 0.05$ ), despite the fact that the cumulative length frequency distributions were remarkably similar (Fig. 2D).

### Daytime Trends

Despite the variability in density estimates within a single day at some sites (Fig. 3), on average there



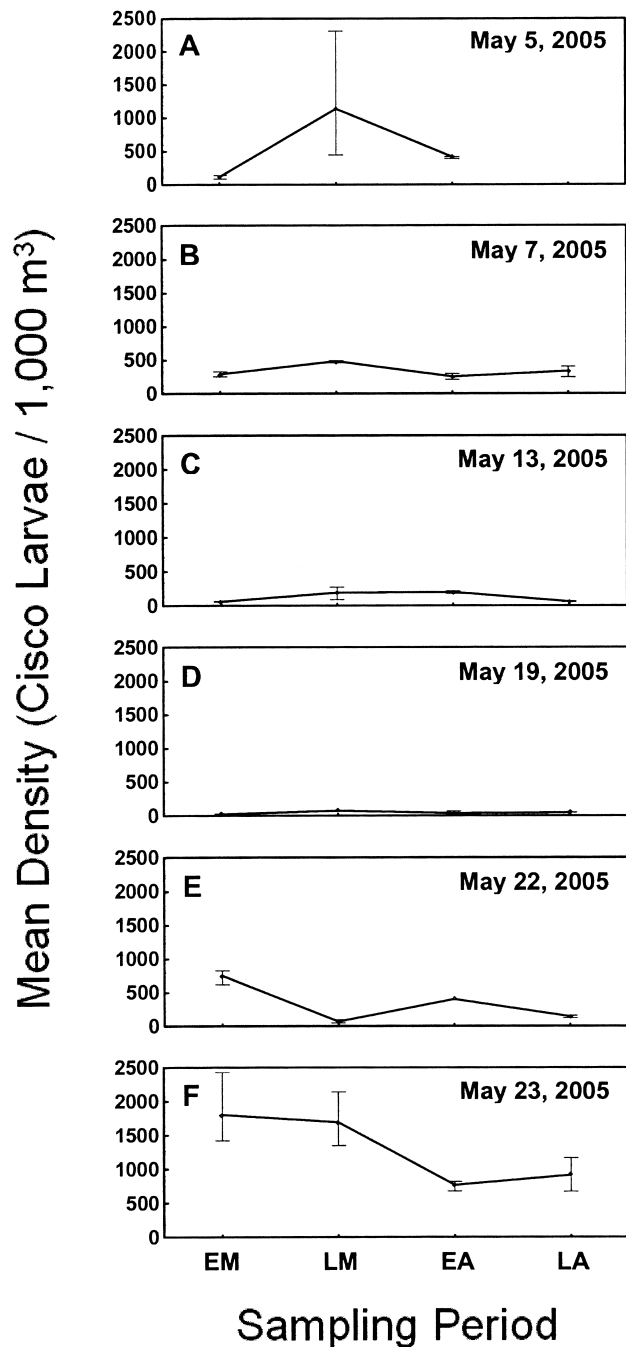
**FIG. 2.** (A)  $\text{Log}_{10}$ -transformed larval cisco density ( $\#/1,000 \text{ m}^3$ ) estimates and (B) cumulative length frequency distributions of larval cisco collected with side-by-side tows using a  $2 \times 1\text{-m}$  paired neuston net and a  $0.5\text{-m}$  (diameter) conical net. (C)  $\text{Log}_{10}$ -transformed larval cisco density ( $\#/1,000 \text{ m}^3$ ) and (D) the cumulative length frequency distributions of larval cisco collected using sinusoidal and straight tow patterns with either a  $2 \times 1\text{-m}$  neuston net ( $\blacklozenge$ ) or a  $0.5\text{-m}$  (diameter) conical net ( $\square$ ). The dashed lines (A and C) are 1:1 lines.

was no pattern in density estimates during daylight. When analyzing days with three replicate tows for all four time periods, there were no significant trends throughout the period of an individual day ( $F = 2.16$ ,  $P = 0.194$ ,  $n = 3$ ). Likewise, when repeating the analysis using only the first tow for the four time periods, there was no evidence of a pattern in larval cisco density within a day ( $F = 0.32$ ,  $P > 0.8$ ,  $n = 5$ ).

## DISCUSSION

Several studies have examined the dynamics of cisco early life history by attempting to estimate abundance of swim-up larvae. Evidence from many of these studies has shown emergent cisco to be distributed throughout the water column but concen-

trated primarily in the surface stratum (Anderson and Smith 1971, Selgeby *et al.* 1978, Hatch and Underhill 1988, Oyadomari and Auer 2004), leading us to use only surface horizontal tows to index density. Selgeby *et al.* (1978) evaluated the performance of several gear types (Clarke-Bumpus sampler,  $0.5\text{-m}$  tow net,  $1 \times 1\text{-m}$  net) and concluded, similar to the results of our study, that the effective sampling area of a gear does not appear to affect estimated density of larval cisco. Hatch and Underhill (1988) also conducted a gear comparison ( $0.5\text{-m}$  Bongo sampler and  $1\text{-m}$  Tucker trawl) and found no difference between density estimates from the two gears. However, neither study addressed sampling design issues in light of their results. Because Selgeby *et al.* (1978) conducted a limited number of



**FIG. 3.** Average larval cisco density (#/1,000 m<sup>3</sup>) as a function of time of day. Sampling events occurred in the early morning (EM), late morning (LM), early afternoon (EA), and late afternoon (LA) for six individual days at different sites. Error bars represent the range of estimated densities for each sampling event.

paired comparisons ( $n = 5$ ) and Hatch and Underhill (1988) did not report detailed diagnostics for their analysis, we felt that our further investigation of appropriate sampling devices was warranted.

Our results suggest that samples from the conical net are comparable to those from the much larger neuston net. Because conical nets are affordable, can be deployed from any small vessel without special requirements (e.g., hydraulic winch), and result in smaller catches (equating to less laboratory processing time), we recommend the conical net for future surveys of larval cisco. We note that smaller sample volumes and the patchiness of local distributions may cause surveys using conical nets to be vulnerable to greater variation in density estimates. To some extent this can be offset by increasing the number of tows conducted. Depending on the objectives of future studies this may be an important consideration when choosing a gear to deploy. The length frequency distributions of larvae captured by the two gears were significantly different, with the neuston net capturing slightly larger individuals (Fig. 2B). We consider this difference, while significant according to the K-S test, is small enough to be unimportant from a practical viewpoint. Both nets captured a limited range of larval cisco sizes, possibly due to the emigration of larger age-0 cisco from the surface waters or gear avoidance by larger larvae. We feel alternate sampling strategies and gears would be needed to assess age-0 cisco once they reach larger sizes, a goal beyond the scope of the present study.

A recent study showed bow-mounted push nets had an increased efficiency for sampling of larval sunfish (*Lepomis* spp.) but not for gizzard shad (*Dorosoma cepedianum*), suggesting that vessel interference may influence larval fish catchability for certain species (Claramunt *et al.* 2005). Because of the sea conditions encountered in Lake Superior and the rigid mounting apparatus used to deploy this gear type, its use was not considered as it would often rise above the water's surface with the rise and fall of the vessel's bow. Instead, we tested for effects of vessel interference within our sinusoidal tow versus straight tow analysis. Despite the concerns, we did not observe an effect on estimated density caused by tow pattern. The differences in length frequency distributions for the two tow patterns, while statistically significant, were very small (Fig. 2D) and are very unlikely to represent differences that would influence the biological interpretation of survey results. Given the availability of global positioning systems, we recommend sam-

pling larval cisco using straight tows to simplify protocols and facilitate straightforward measurements of volume filtered (distance x net mouth area). We do not however suggest abandoning the use of flowmeters, as their use may serve as an added measure of distance.

Finally, our investigation showed no trends in density over the light period of a day, which indicates density estimates across space within a given day are comparable and can be used to estimate mean density for the sampled region. Recent efforts to measure spawner and recruit abundance are based on statistical grids (10' latitude by 10' longitude) because this scale is of interest to managers within Lake Superior. Assessments of larval cisco will be of more utility when they are executed to the same scale. Collectively, our results suggest that multiple vessels/agencies can be used to conduct larval cisco surveys of major geographic regions within Lake Superior, facilitating large-scale research initiatives.

When designing sampling programs, it is imperative to understand the biases associated with different gears and strategies. Studies targeting the larval phase of cisco should employ gears known to efficiently capture the demographics of this life stage. However, it is also important to make effective use of time and resources. Developing protocols that balance these conditions will ensure that surveys achieve a rigorous estimate of abundance across the widest possible spatial range. Using a coordinated effort to advance the level of understanding of cisco early life history in Lake Superior will facilitate research initiatives and ultimately provide a greater understanding of cisco population dynamics.

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