GREAT LAKES FISHERY COMMISSION

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Otolith microchemistry for percid production dynamics in Lake Erie

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ABSTRACT

Walleye stock structure in Lake Erie has been explored using a variety of genetic and meristic techniques, however, the proportional contribution of individual stocks has yet to be determined. Knowledge of relative stock contribution may assist management decisions surrounding preservation or rehabilitation efforts concerning habitats crucial to one or more stocks.

A mixed stock analysis was performed on samples of juvenile age-0 walleye from the 2003 cohort using otolith microchemistry and linear discriminant analysis. Otolith-based stock discrimination is possible for walleye due to the differences in spawning and nursery habitats used by the various stocks throughout the basin. Subtle differences in trace-metal concentrations at natal environments are recorded by the otolith and can be used to distinguish between stocks. A library of larval walleye otolith signatures from the 2001 year-class was used as the basis of the discriminant analysis. All major stocks within the western basin could be discriminated at a classification accuracy of 67% to 100% using Mn, Sr, and Ba concentrations. Juveniles from reef and river-spawned stocks exhibited clear differences in Sr and Mn concentrations, emphasizing the exposure to environments within western Lake Erie. In August 2003, most juvenile walleye exhibited characteristics of the Maumee River and Hen Island stocks. The knowledge that multiple stocks contribute differentially to the Lake Erie walleye population allows for the creation and testing of hypotheses concerning individual stock dynamics to begin.

INTRODUCTION

Lake Erie supports the most valuable freshwater fishery in North America. Although more than 20 species contribute to the catch, two percid species, walleye *Stizostedion vitreum* and yellow perch *Perca flavescens* comprise the majority of the harvest. While walleye have been exploited commercially from the beginning of recorded fishery history until the present day, percid species have also been an important component of Lake Erie recreational fisheries. The walleye standing stock size has fluctuated greatly between the 1970s and today. During the period of 1978 until 2003, adult (age-2+) walleye population size in Lake Erie increased to an estimated 69 million fish during the late 1980s, before gradually declining to an estimated 29 million fish in 2003 (Walleye Task Group (WTG) 2004). With the downturn in walleye abundance over the past decade, new research regarding walleye recruitment and stock structure are welcomed by managers of the fishery; stock discrimination of Lake Erie walleye stocks has been identified by the Great Lakes Fishery Commission as a guiding principle in the creation of fish-community management objectives for Lake Erie (Ryan et al. 2003).

Lake Erie's fisheries are jointly managed by the Michigan Department of Natural Resources (MDNR), the New York State Department of Environmental Conservation (NYSDEC), the Ohio Department of Natural Resources (ODNR), the Ontario Ministry of Natural Resources (OMNR), and the Pennsylvania Fish and Boat Commission (PFBC). In 2000, these agencies developed a Coordinated Percid Management Strategy, designed to protect and rebuild percid populations (WTG 2001). A critical goal of this long-term strategy is to identify factors that regulate production, so that management models can be refined (WTG 2001, Yellow Perch Task Group 2001). The need to identify and discriminate stocks is central to this goal, and a recent external science review of the stock assessment program confirmed that identification of stock structure is critical to ensuring future sustainability. Identifying stock discreteness is also a top priority with other fisheries in the Great Lakes (Kutkuhn 1981; Marsden et al. 1989; Bronte et al. 1996) and elsewhere (Begg et al. 1998, 1999; Campana et al. 1999; Stephenson 1999).

There is evidence that the walleye population of Lake Erie consists of multiple stocks, yet the stock structure of Lake Erie walleye population, or the extent that individual stocks contribute to the western basin population, is still not fully known. Although adult walleye move throughout the lake during the majority of the year, they separate into discrete spawning stocks during the late winter and early spring in order to breed (Todd and Haas 1993). Generally, walleye stocks are classified as either river- or reef-spawning, with walleye returning to the same spawning grounds year after year (Olson and Scidmore 1962, Mion et al. 1998, Robert Haas, Michigan DNR, pers. comm.). That walleye choice of spawning habitat has been demonstrated to have a heritable component (Jennings et al. 1996) adds to the

evidence that distinct walleye stocks likely exist in Lake Erie.

Stock discrimination of walleye in Lake Erie has been attempted previously using genetic techniques. Differences between spawning stocks could not be detected in two studies (Billington and Herbert 1988, Todd and Haas 1993, McParland et al. 1999). There is conflicting evidence that the walleye breeding groups from the Maumee River and Sandusky River can be separated (Merker and Woodruff 1996, Stepien and Faber 1998). Major histocompatibility genes have been explored as potential genetic stock discrimination tools for walleye (Fujiki et al. 2001), however broad scale application has yet to be evaluated for Lake Erie walleye.

Stock discrimination of fishes using otolith microchemistry has been employed in numerous studies (reviewed by Campana 1999). Otoliths are natural chronologic structures that can serve as biological tracers of fish given that they are metabolically inert and assimilate trace elements in relation to the concentrations present in the environment as they increase in size (Campana and Thorrold 2001). The otolith is thus a permanent record of the chemistry of the environment, or environments, a fish has experienced (Campana et al. 2000). In turn, the time-keeping properties of otoliths have been used successfully to detect movements of fish between habitats such as marine and freshwater (Tzeng and Tsai 1994, Radke et al. 1998, Zlokovitz et al. 2003).

Although not as markedly different as moving between marine to freshwater environments, potential differences between the river and reef natal areas within Lake Erie provide an opportunity to use otolith microchemistry to determine the proportional stock contribution to the walleye population of western Lake Erie. To better understand the walleye stock dynamics occurring in western Lake Erie, we used a pre-existing library of larval walleye otolith elemental concentrations to conduct a mixed stock analysis (MSA) based on otolith microchemistry. The completion of an MSA would allow for the assessment of the proportional contributions of individual stocks occurring within the western basin. In addition, an MSA would establish if there is complete mixing of juvenile walleye from different natal areas occurring in the western basin. The sorting of individual age-0 fish into stock groupings allows stock-specific condition and growth to be assessed.

This project specifically addresses fishery research priorities identified by the Council of Lake Committees and the Lake Erie Committee regarding walleye stock discrimination. The application of our otolith microchemistry techniques to a mixed stock analysis using samples collected from the Lake Erie agency recruitment indexing program will ensure that our results are directly relevant and easily incorporated into their current management strategies.

Our work also supports the Great Lakes Fishery Resources Restoration Study (GLFRRS) recommendation 12 *(restoration and enhancement of exploited and / or declining species*) by focussing

on one of the most important species in the Lake Erie, and Great Lakes, sport and commercial fishery. First, by identifying and documenting the potential differential contribution of stocks, and then by evaluating the factors contributing to this differential survival, growth and production we will further enhance the ability resource management agencies to recognise and account for this uncertainty in their management models.

OBJECTIVES

Project Objective 1:	To verify that production of walleye in Lake Erie is a consequence of differential recruitment of stocks produced in the western basin.
Results:	Completion of a mixed stock analysis based on sagittal otolith microchemistry identified three stocks (Maumee River, Ontario reefs, and Sandusky River) which contributed unequal numbers of individuals to the population.
Status:	Objective accomplished.
Project Objective 2:	To determine if different walleye stocks yield young-of-year (YOY) of different size and condition.
Results:	Comparison of average TL for both of the stocks with adequate sample sizes (Maumee River and Ontario reefs) indicated a slight statistically significant difference in average TL between stocks.
Status:	Objective accomplished.
Project Objective 3:	To determine the relative contribution of genetic and environmental factors in controlling differential stock survival and growth.
Results:	Otolith chemical signatures in juvenile fishes may be influenced by ontogeny (Fitzgerald et al. 2004). The changes in elemental uptake for larval and juvenile walleye need to be assessed before otolith elemental profiles can be used successfully to trace movements of fish. Jason van Tassel (Ph.D. candidate, Ohio State University) is pursuing this research question as part of his dissertation.
Status:	Objective not accomplished.
Projective Objective 4:	To investigate the use of otolith microchemistry during larval growth stages to determine their changing physicochemical environment.
Results:	We were unable to recruit a student to conduct uptake experiments during the

time scheduled for the project. A study of the uptake mechanisms in juvenile fish is now underway.

Status: Study ongoing.

METHODS

Mixed stock analysis of age-0 walleye in Lake Erie

Walleye collections. Age-0 walleye (70-142 mm TL) were obtained from an interagency percid recruitment monitoring program conducted by the Ontario Ministry of Natural Resources and the Ohio Department of Natural Resources. The program, has run continuously since 1987 and consists of approximately 80 ten-minute standardized bottom trawls conducted by the two agencies annually every August in the western basin of Lake Erie (Figure 1). Trawls were stratified into four depth strata (0-3 m, 3-6 m, 6-9 m, and >9 m) in proportion to the area in each depth strata. Trawling was completed with a two-seam, modified Biloxi bottom trawl with a 10.3 m headrope, 11.8 ground line, and a 13 mm mesh cod end. The program sampled 78 sites across the western basin during August 2003. Additional age-0 walleye samples were obtained from the United States Geological Survey (USGS) office in Sandusky, Ohio. All age-0 walleye samples were frozen and stored until processing.

Subsampling design for 2003 age-0 walleye collections. Two different subsampling strategies were employed in order to address both density and distribution of juveniles in western Lake Erie. The first subsample (n = 100) was weighted by trawl abundance (Figure 1). In this sample, the total basinwide catch was allocated to the subsample in proportion to its catch [i.e. high catch areas contributed more to the sample than low catch areas (Table 1)]. The abundance-weighted subsample was considered to be representative of the 2003 year-class as a whole, and was used to infer stock structure for the 2003 walleye cohort. The second subsample (n = 100) was weighted by the area of the western basin. The western basin was divided into 10 quadrats (Figure 2). Each quadrat contributed individuals to the subsample in proportion to its relative surface area (Table 2). The area-weighted sample was analysed to investigate if juvenile walleye exhibited stock specific aggregations within the western basin of Lake Erie and to assess how far juveniles had dispersed from nursery areas by August of their first year.

Sample preparation. Fish were thawed, weighed (to nearest 1 g), and measured for total length (to nearest 1 mm) prior to otolith extraction. Sagittal otoliths were removed using the "up through the gills" method (Secor et al. 1991) using non-metallic forceps. Visible tissue was removed from otoliths prior to placement in vials, where they dried in open air for 24 hours.

Dry otoliths were mounted on acetate squares for polishing using cyanoacrylate glue (Krazy® glue) as the mounting medium. Otoliths were mounted with the sulcus side upward and then polished

down the sagittal axis with 30-, 12-, 9-, 3-, and 1- m lapping film (Precision Surfaces International, Houston, TX) in order to expose the core. Otoliths were viewed under a light microscope at 100x-200x magnification in order to verify that the core region was at the otolith surface. Polished otoliths were mounted to glass petrographic slides in groups of 16 for laser ablation inductively-coupled mass spectrometry (LA-ICP-MS) analysis. Prior to LA-ICP-MS analysis, each otolith was photographed under 100x-200x magnification using either a Nikon monochrome CCD video camera mounted on a Nikon Labophot-2A microscope or a CoolSnap Pro color CCD video camera mounted on an Olympus BX51 microscope. In order to improve the clarity of the otoliths, ultra-pure Milli-Q water was applied to each slide during photography. Slides were allowed to air dry prior to further handling. Sonication of the slides in ultra-pure Milli-Q water for 10 minutes occurred before LA-ICP-MS analysis in order to remove polishing residues and other potential contaminants. Sonicated slides were rinsed three times with ultra-pure Milli-Q water, dried under a HEPA-filtered, laminar-flow hood for 24 hours, and stored in acid-washed Petri dishes prior to LA-ICP-MS analysis. All final stages of cleaning, drying, and storage occurred in a class 100 clean room.

Laser ablation. Otoliths were chemically analysed using LA-ICP-MS at the Great Lakes Institute of Environmental Research, University of Windsor. A Thermo Elemental X7 ICP-MS was operated at low resolution using argon as the carrier gas. The purpose-built laser sampling system was based on a non-homogenized, high power, frequency-quadrupled (wavelength: 266 nm, pulse rate: 20 Hz) Nd: YAG laser (Fryer et al. 1995). The laser beam was focussed onto the sample using an Olympus BX-51 petrographic microscope and an Optics For Research 266 nm 10X objective lens. A 1.5 mm pinhole beam constrictor was used. Otoliths were ablated in lots of sixteen, along with four ablations (two before and two after) of a glass standard reference material (NIST SRM 612) for calibration and to correct for ablation yield and instrument drift. Ablation proceeded in transects targeting the core across each otolith. Laser traverse speed was kept constant at 5 µm/s. Each otolith was analysed via LA-ICP-MS once. Otoliths were viewed under 100x-200x magnification following ablation in order to confirm that the laser ablation trajectory crossed the core region. Otoliths with ablation transects that did not cross over the core region were removed from further analysis.

Otolith elemental signature selection. The analytical data corresponding to the region immediately after the core (5 seconds) was isolated from each otolith laser ablation transect using one of two methods. The first method of core detection relied on the existence of the an elevated Mn concentration in the core region. Previous studies have observed that sagittal otolith core regions of both freshwater and marine fishes have elevated levels of manganese (Hedges 2002, Brophy et al. 2004, Ludsin et al. in press). The elevated manganese (Mn) core concentration (or Mn "spike") can be used successfully to pinpoint the

region within an ablation transect data series that corresponds to the core. When the Mn spike was absent, the otolith ablation transect was viewed and measured under 200x magnification using SigmaScan® (SPSS Science, Chicago, IL.). The distance from the start of the ablation to the core region was measured (μm) and the time to the core region (measured in seconds) was calculated. The value for the time to the core region was increased by 3 seconds in order to compensate for the delay in analysis material in the ICP-MS. The adjusted time point was used to mark the beginning of the five-second (~25μm), post-core span. To verify that this method successfully located core regions, a subsample of otoliths that exhibited an Mn spike were processed using the microscope-measurement method. Calculated core region measurements were compared to the core region timing of the Mn spike and values obtained from the microscope-measurement method were found to agree with timing of the actual Mn spike signal.

Core regions were integrated using LAMTRACE, a spreadsheet based program for LA-ICP-MS data reduction, in order to calculate an elemental signature for each otolith processed (Van Achterbergh et al. 2001). The internal standard used for the calculations was ^{43}Ca. The lower limits of detection (LLDs) were calculated for each element in each sample using an algorithm developed by Longerich et al. (1996). The LLDs take into account the background noise, the length of integrated background interval, the drift-corrected, normalised sensitivity for each element, the length of ablation signal that was integrated, and ablation yield (Van Achterbergh et al. 2001). If an elemental concentration fell below the LLD for a sample, the value was considered to be missing.

Due to a limited supply of age-0 fish, a group of otolith elemental signatures was shared between the abundance- and the area-weighted subsample. Additional otolith elemental signatures were dedicated to only one of the two subsamples in order to minimise the overlap of individual fish otoliths in each subsample. All age-0 walleye elemental concentrations for the 2003 cohort were $\log_{10}(x+1)$ transformed to achieve normality prior to the performance of the mixed stock analyses.

Generation of the linear discriminant function (LDFA) and performance of the mixed stock analysis (MSA). Using initial samples with cases of known origin, LDFA identifies gradients of variation within a given dataset and generates a linear mathematical classification function that maximises the variation between predetermined groups (McGarigal et al. 2000, Rosner 2000). LDFA is also capable of classifying additional cases through the application of the discriminant function to new comparable data. The discriminant function from LDFA of 2001 larval walleye from western Lake Erie was used to perform an MSA of age-0 walleye from the 2003 year-class.

Larval walleye otolith elemental signatures from across western Lake Erie previously analysed by Hedges (2002) were the source for the training set used to generate the discriminant function (Table 3).

The LDFA was performed using the classic complete estimation (or direct) procedure. Structure coefficients (also referred to as "factor correlations") were examined to determine which elements were influential in the analysis. Classification functions produced by the LDFA were tested with a re-sampling procedure to estimate classification accuracy.

Raw elemental concentrations from the library were $\log_{10}(x + 1)$ transformed to achieve normality. Inclusion of an element as a potential discriminating variable in the LDFA occurred when two selection criteria were satisfied: (1) the mean coefficient of variation (CV = 100 • SD/mean) for the glass standard reference samples was less than 10.5 %, an indication of an acceptable level of precision (Gillanders and Kingsford 1996); and (2) the element was detected above the LLD for at least 90 % of all cases. If an elemental concentration fell below the LLD for a sample, the value was considered to be missing. Missing values were replaced with the group (spawning location) median concentration for each element (Legendre and Legendre 1998). Since outliers can greatly influence a discriminant analysis (McGarigal et al. 2000, McCune and Grace 2002), groups were tested for outliers by calculating the extreme studentised deviate statistic for each group and cases with scores above the 95th percentile (three cases) were removed (Rosner 2000)

The classification functions from the LDFA were then applied to the two subsamples from the 2003 age-0 walleye cohort of western Lake Erie in order to complete the MSA of the 2003 year-class. Application of the classification function to the individuals from the 2003 year-class allowed for the creation of percent composition of both the abundance- and area- weighted samples.

Elemental uptake mechanisms of young fish

The objective of this study is to examine the uptake and incorporation of Ba, Sr, and Mg (at three concentrations) into guppy (*Poecilia reticulata*) otoliths. Experiments will be run using one element at a time (i.e. Ba experiment is conducted first). Guppy broods will be born into the experimental conditions (element at one of three concentrations or control) and reared until 6 weeks of age. Otoliths will be removed and analysed from a sample of each brood at 2, 4, and 6 weeks of age. Otolith signatures from both within (same conditions) and between (different Ba concentrations) broods will be compared and assessed.

Gravid female guppies were placed into one of 8 experimental (1.5 L) tanks. All tanks were originally filled (1.4 L) with tap water and allowed to stand overnight to allow chlorine to evaporate. Different amounts of a 1% Ba solution were added to six of the tanks in order to create Ba concentrations in the experimental tanks that were 2X, 4X, and 16X of the background water concentration (~20 ppb). Each concentration level was assigned to two of the tanks (random selection). The Ba concentration of the two control tanks were left at background levels (20 ppb). Females were kept in experimental tanks until

each gave birth to a brood of young. Females were removed as soon as juvenile guppies were observed in a tank. Juvenile guppies were raised in experimental tanks. After 14 days, a sample of the juvenile guppy brood were removed from the experimental tank and sacrificed. Guppy samples were frozen for further analysis (otolith extraction). Broods were sampled in the same manner as described above at 24 and 36 days.

Otolith extraction and analysis. Sagittal otoliths were removed from sacrificed fish. Otoliths will be secured to acetate and polished with lapping film to create a flat plane for imaging. After polishing, otoliths will be secured to glass petrographic slides, sonicated and analysed via LA-ICP-MS.

RESULTS

Mixed stock analysis of age-0 walleye in Lake Erie

Generation of the LDFA from 2001 larval otolith elemental signatures. Four potential stocks were selected for inclusion in the LDFA based on evidence of breeding stocks within the western basin: Maumee River, the reefs of the Ohio waters, the reefs in Ontario waters, and Sandusky River (Goodyear et al. 1982, Roseman et al. 2002). The elements that satisfied the LDFA inclusion criteria were determined by measuring the following isotopes: ^{55}Mn, ^{86}Sr, ^{88}Sr, ^{137}Ba, and ^{138}Ba (Table 3). Only ^{86}Sr and ^{137}Ba were used in the analysis due to high intra-isotopic correlations (Sr: r = 0.98, df = 68; Ba: r = 0.96, df = 68).

There were differences in Mn and Sr concentrations between groups (Figure 3). River spawning stocks (Maumee, Sandusky) had lower Mn and higher Sr than the reef stocks (Ohio and Ontario reefs) whereas there was little variation in Ba among stocks. The differences between Mn and Sr concentrations detected suggested that an LDFA would be able to successfully divide the 2001 elemental concentrations into groupings. Two discriminant functions were needed to separate the larval walleye elemental signatures from the 2001 year-class into discrete groups (Wilk's lambda = 0.06, $F(9, 155) = 38.87$, $p < 0.001$) (Figure 4a). Together, these functions explained >98% of the variation between individual fish (Chi square = 188.23, df = 9, $P < 0.001$). Standardised canonical coefficients for function 1 (axis 1) were 0.23, -0.96, and 0.41 for Mn, Sr, and Ba, respectively. Standardised coefficients for Mn, Sr, and Ba for function 2 (axis 2) were -1.00, -0.47, and 0.16, respectively.

To properly assess which variables were associated with each function, the product-moment correlations between the canonical functions and the individual elements (also referred to as "structure coefficients") were examined (McGarigal et al. 2000). Coefficients that had an absolute value of >0.32 were interpreted for each function, as coefficients above this threshold account for >10% of the variance of a function (Tabachnick and Fidell 1996). Manganese had structure coefficients of 0.43 and -0.90 for functions 1 and 2, respectively. Strontium had a structure coefficient of -0.90 for function 1 and its

coefficient was below threshold limits for function 2. Barium was below threshold limits for both functions and thus contributed marginally to the discrimination.

Classification accuracy (McGarigal et al. 2000, Rosner 2000) for the discriminant function was significantly better than chance (Cohen's K statistic = 0.90, SE ± 0.07). The discriminant function correctly classified an average of 93% of the cases in the dataset (Table 4). Classification accuracy for individual stocks in the LDFA ranged from 67% to 100%. Sandusky River and the Ontario reef fish were always correctly classified; Maumee River fish were misclassified 33% of the time (3 of 9 cases). A jackknifed classification matrix of the training set, produced for cross-validation purposes (Engelman et al. 2004) correctly classified 91% of cases (Table 4). The decline in classification accuracy in the jackknifed dataset was due to one misclassification in the Ontario reef sample. The first function of the LDFA separated river from reef spawning stocks on the basis of Sr and Mn (Figure 4). Sandusky River and Ontario reef fish were most dissimilar, while Maumee River and Ohio reef fish were centred about the origin indicating less distinct Sr and Mn concentrations for larvae from these sites.

MSA of 2003 age-0 walleye. Data were prepared for mixed stock analysis by ensuring that no zero values existed in the data matrix. A simple regression was developed to predict Mn from Mg ($r^2 = 0.50$, $F = 90.23$, df = 96, $p < 0.001$). Missing Mn values (due to failure of the sample to comply with all the criteria identified in the methods) were generated for nine age-0 walleye in 2003 (Table 1).

The discriminant functions developed during the LDFA were then applied to the otolith elemental signatures for the 2003 age-0 walleye cohort. The abundance-weighted sample ($n = 81$) was separated into three groups: Maumee River (64%), Ontario reefs (33%), and Sandusky River (2%) (Figure 4b). The area-weighted sample ($n = 83$) yielded a similar stock contribution: Maumee River (61%), Ontario reefs (37%), and Sandusky River (1%) (Figure 4c). There was no relationship between stock contribution and proximity to spawning site for the area-weighted sample, suggesting that age-0 walleye in August are a well-mixed assemblage of stocks throughout the basin (Table 5).

Stock-specific assessment. As only two individuals were identified as originating from Sandusky River, this stock was not included in the analysis of stock-specific size. The average total length of individuals (TL) was different for the Maumee River and Ontario reefs stocks (ANOVA, $F = 6.682$, $p = 0.011$). Juvenile walleye identified as part of the Maumee River stock being slightly larger than walleye identified as originating from the Ontario reefs (Figure 5).

DISCUSSION

The mixed stock analysis of samples from the western basin of Lake Erie suggests that the majority of juvenile fish produced during the 2003 spawning season originated in the Maumee River.

Offshore reefs in the northern half of the basin (Ontario) also successfully produced juvenile walleye in 2003. No fish were classified as originating from the Ohio reefs, and production of the Sandusky River was much less than that of the Maumee River. Walleye production in the Sandusky River (when compared to walleye production of the Maumee River) has been reported before (Mion et al. 1998), based on larval catch rates. It must be remembered that these stock contributions are for 2003 only, and similar proportions may not be produced in other years. Additional study years are needed to better understand the broader-scale contributions of different stocks.

As only the pattern of contribution for the 2003 cohort is known, no inference can be made into the dynamics behind strong and poor year classes. The unequal production of individual stocks could suggest mechanisms as to why there is large annual variation in walleye production in Lake Erie. Individual stocks spawn in separate areas within the western basin. Meteorological events (such as violent winds) may occur in the vicinity of some spawning grounds and not others, causing the failure of one stock's offspring while other stocks are able to successfully produce young. While the search for reliable environment-recruitment relationships for western Lake Erie has been occurring for the last 30 years with little success (Bartnik, 2005) some intuitive relationships have been proposed that illustrate how environmental factors may affect different spawning and nursery areas differentially.

The lack of any spatial gradient in stock composition of the individual quadrats from the area-weighted sample show that age-0 walleye from various spawning areas in the western basin have mixed thoroughly by August of their first year. Since otoliths record the ambient environmental conditions a fish experiences throughout its life, it may be possible to track individual juvenile walleye for the first few months of its life. As the increments of an otolith can be used to age a fish (Wright et al. 2002), the chemistry of the otolith corresponding to an increment (or a series of increments) can be extracted and compared to reference elemental signatures from across the western basin. A reference library of elemental water signatures from numerous sites within the western basin would need to be constructed for the most valuable results, and a more thorough understanding of the nature of the relationship between water chemistry and walleye otolith chemistry would need to be established for such an analysis to be worthwhile.

The results of the MSA provide evidence for the occurrence of differential stock contribution in the western basin of Lake Erie. With evidence of differential contribution of stocks to the net abundance of a year-class, management could conduct additional MSAs in western Lake Erie to discover the relative stock contributions of western Lake Erie walleye over subsequent years, as the contribution proportions of the 2003 cohort cannot be assumed to reflect any long-term or persistent patterns in relative stock contributions. The practice of completing periodic mixed stock analyses (at a frequency of 3-4 years, a

simple generation for walleye) could track the viability of the various stocks in the western basin of Lake Erie.

Unsustainable exploitation of a single stock in a multiple stock fishery is possible if stock structure is unknown or ignored. Unsustainable fishing and the consequent extirpation of a vulnerable stock could occur (Begg et al. 1999). The extirpation of the stock may not dramatically reduce the yield of the fishery yet the resultant loss of genetic variation could be detrimental, especially if the extirpated stock was adapted to a specific environment. In the case of Lake Erie walleye, there is no method available currently to determine if reef- or river-spawning walleye stocks are being harvested with equal effort, however the MSA results of the spatially-weighted sample indicate that the age-0 walleye are well mixed by August. Adult walleye from different stocks also mix throughout the basin when not spawning (Todd and Haas, 1993), so overexploitation of one stock during the harvest season would be unlikely. Knowledge of the stock dynamics of western Lake Erie walleye could lead to a better understanding of early life history and recruitment dynamics of walleye, as well as provide the impetus to start or support remediation of spawning habitats throughout the basin.

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Table 1.—Composition of the abundance-weighted sample of age-0 walleye from western Lake Erie, 2003. Collectively the sample size generates an approximate equal proportion of the total basin wide trawl catch.

Trawl Location (D°M'S")						Total Catch	Sample Size
Latitude (N)			Longitude (W)				
41	43	32.9	82	44	15.0	47	3
41	53	34.8	82	43	18.1	25	1
41	56	22.9	83	2	52.8	27	2
41	49	18.1	82	53	46.7	48	2
41	50	3.5	82	47	40.2	12	1
41	56	24.7	82	47	57.5	37	1
41	56	47.4	82	41	49.9	57	3
41	51	54.7	82	31	34.7	9	0
41	48	56.9	82	32	34.8	9	1
41	43	43.7	82	32	33.0	14	1
41	41	3.1	82	35	0.6	11	1
41	41	54.6	82	43	3.7	135	8
41	48	1.1	82	44	1.3	101	5
41	59	52.1	82	44	55.3	65	3
41	57	2.5	82	59	8.5	23	1
41	53	8.5	82	52	38.3	37	2
41	54	0.7	82	46	45.1	166	10
41	54	43	82	37	45.1	100	6

Trawl Location (D°M'S")						Total Catch	Sample Size
Latitude (N)			Longitude (W)				
41	51	28.8	82	43	28.2	46	2
41	45	49.7	82	46	10.9	87	4
41	45	49.7	82	46	10.9	33	2
41	39	42.1	82	45	36.0	65	2
41	40	12.0	82	51	18.0	10	0
41	36	6.1	82	54	11.9	9	1
41	33	42.1	82	51	29.9	30	0
41	31	54.1	82	52	59.9	42	3
41	39	47.9	82	55	36.1	123	0
41	46	0.1	82	58	30.0	50	2
41	50	12.1	83	3	24.1	84	5
41	51	42.1	83	6	24.1	9	1
41	53	30.1	83	5	53.9	35	2
41	45	54.0	83	18	11.9	55	3
41	42	6.1	83	14	30.1	12	1
41	44	35.9	83	18	42.1	17	1
41	28	41.9	82	44	24.0	18	1
						TOTAL	81

Table 2.—Composition of the area-weighted sample of age-0 walleye from western Lake Erie, 2003. The sample size was chosen to provide a number of fish equal to the proportional area of that quadrat to the whole basin.

	Quadrat Boundaries (D°M'S")								
Quadrat	Direction	Latitude (N)			Longitude (W)			Area (m^2)	Sample Size
A	East				83	15	0		
	South	41	48	0					
								166.53	0
B	East				83	0	0		
	South	41	48	0					
	West				83	15	0		
								589.33	12
C	East				82	45	0		
	South	41	48	0					
	West				83	5	0		
								457.13	12
D	East				82	30	0		
	South	41	48	0					
	West				82	45	0		
								482.38	12
E	North	41	48	0					
	East				83	15	0		
								224.48	6
F	North	41	48	0					
	East				83	3	0		
	West								
								448.98	12
G	North	41	48	0					
	East				82	45	0		
	South	41	36	0					
	West				83	3	0		
								446.06	11
H	North	41	48	0					
	East				82	30	0		
	South	41	36	0					
	West				82	45	0		
								421.04	11
I	North	41	36	0					
	East				82	45	0		
	West				83	0	0		
								276.22	7
J	North	41	36	0					
	East				82	30	0		
	West				82	45	0		
								386.62	0
								TOTAL	83

Table 3. —Isotopes measured, the limits of detection (LLD) of elemental concentrations, percent of samples above LLD, and the coefficient of variation (CV) for the data used to create the linear discriminant function (2001 larval walleye) and the data tested by the discriminant function (2003 age-0 walleye). Elemental concentrations were originally measured in ppm. Elements in bold were included in the development of the discriminant function.

Isotope	LLD (± SD)	% > LLD	CV (± SD)
	2001 larval walleye		
^{26}Mg	35.27 ± 100.70	87	2.07 ± 1.63
^{55}Mn	**2.64 ± 8.64**	**94**	**1.40 ± 1.10**
^{65}Cu	16.33 ± 59.05	2	2.91 ± 2.36
^{66}Zn	4.67 ± 14.85	60	4.41 ± 3.54
^{86}Sr	**32.73 ± 69.79**	**99**	**3.92 ± 3.97**
^{88}Sr	**0.49 ± 1.85**	**100**	**1.58 ± 1.20**
^{89}Y	0.26 ± 0.88	19	1.87 ± 1.23
^{137}Ba	**1.31 ± 4.11**	**99**	**3.24 ± 2.39**
^{138}Ba	**0.24 ± 0.82**	**100**	**2.25 ± 1.91**
^{208}Pb	0.39 ± 1.34	35	6.59 ± 8.38
^{238}U	0.16 ± 0.50	6	3.19 ± 2.46
	2003 age-0 walleye		
^{7}Li	0.17 ± 0.18	55	7.44 ± 6.17
^{25}Mg	**1.42 ± 1.20**	**100**	**2.23 ± 1.18**
^{55}Mn	**0.26 ± 0.23**	**92**	**1.25 ± 0.90**
^{65}Cu	1.00 ± 0.87	18	3.13 ± 1.28
^{66}Zn	**0.47 ± 0.64**	**96**	**2.48 ± 1.61**
^{85}Rb	**0.06 ± 0.05**	**96**	**2.41 ± 1.06**
^{86}Sr	**0.46 ± 0.42**	**100**	**1.59 ± 0.80**
^{88}Sr	**0.05 ± 0.02**	**100**	**2.81 ± 0.83**
^{89}Y	0.03 ± 0.03	4	1.53 ± 0.69
^{111}Cd	0.10 ± 0.10	15	3.91 ± 1.64
^{120}Sn	0.05 ± 0.04	68	2.28 ± 1.06
^{137}Ba	**0.09 ± 0.08**	**100**	**2.31 ± 0.82**
^{138}Ba	**0.02 ± 0.01**	**100**	**1.73 ± 0.92**
^{140}Ce	0.01 ± 0.01	38	1.85 ± 1.04
^{208}Pb	0.01 ± 0.01	88	2.9 ± 1.2
^{238}U	0 ± 0	54	3.3 ± 2.9

Table 4.—Classification matrices from linear discriminant functions (LDF) used to classify YOY walleye from western Lake Erie to natal stock. LDF was based on Hedges (2002) larval walleye otolith library.

Actual Stock	% Correct	Classification Stock				*n*
		Maumee River	Ohio reefs	Ontario reefs	Sandusky River	
		Training set				
Maumee River	67	**6**	3	0	0	9
Ohio Reefs	90	1	**18**	1	0	20
Ontario Reefs	100	0	0	**18**	0	18
Sandusky River	100	0	0	0	**23**	23
Total	93	7	21	19	23	71
		Jack-knifed classification				
Maumee River	67	**6**	3	0	0	
Ohio reefs	90	1	**18**	1	0	
Ontario reefs	94	0	1	**17**	0	
Sandusky River	100	0	0	0	**23**	
Total	91					

Table 5.—Classifications of age-0 walleye in western Lake Erie in 2003 using the area-weighted sample set.

Stock Group	Quadrat							
	B	C	D	E	F	G	H	I
Maumee River	50%	67%	67%	50%	67%	64%	64%	57%
Ohio reefs								
Ontario reefs	50%	25%	33%	50%	33%	36%	36%	43%
Sandusky River		8%						

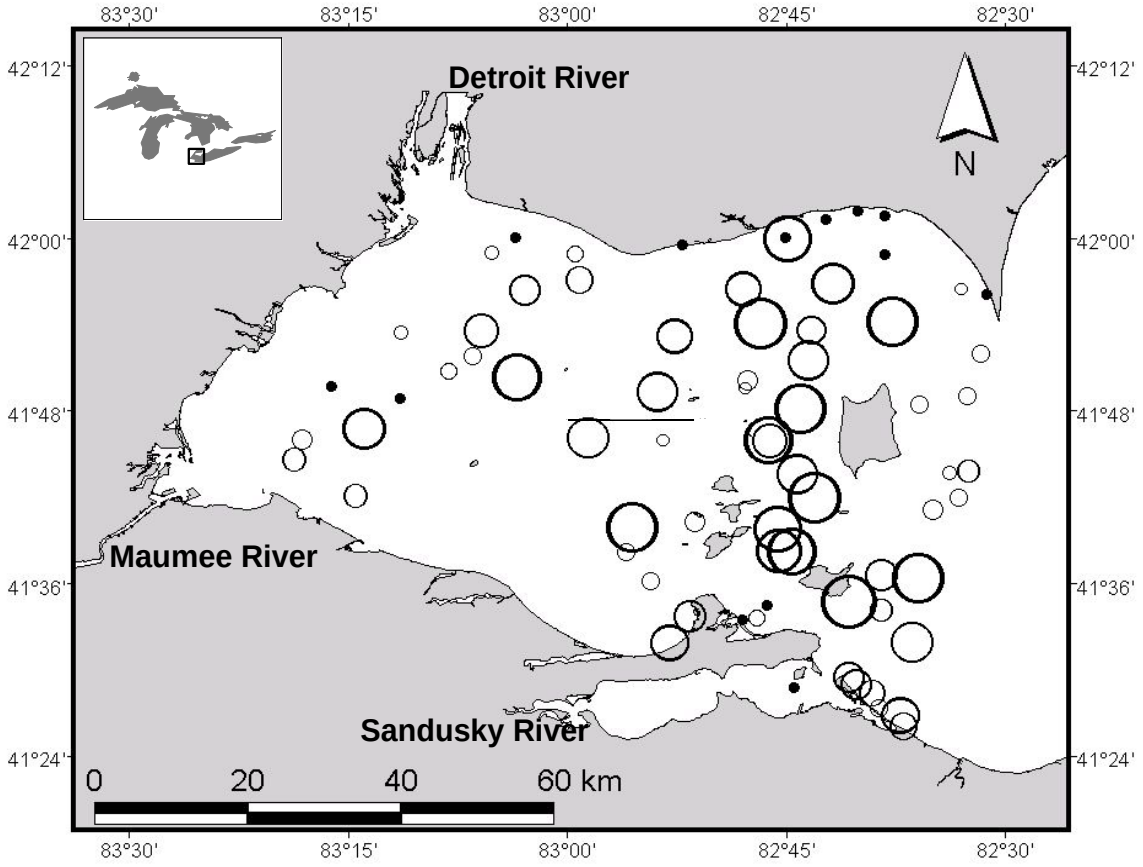

Figure 1.—Abundance of age-0 walleye by collection site in western Lake Erie, 2003. Dots indicate sites where no walleye were caught.

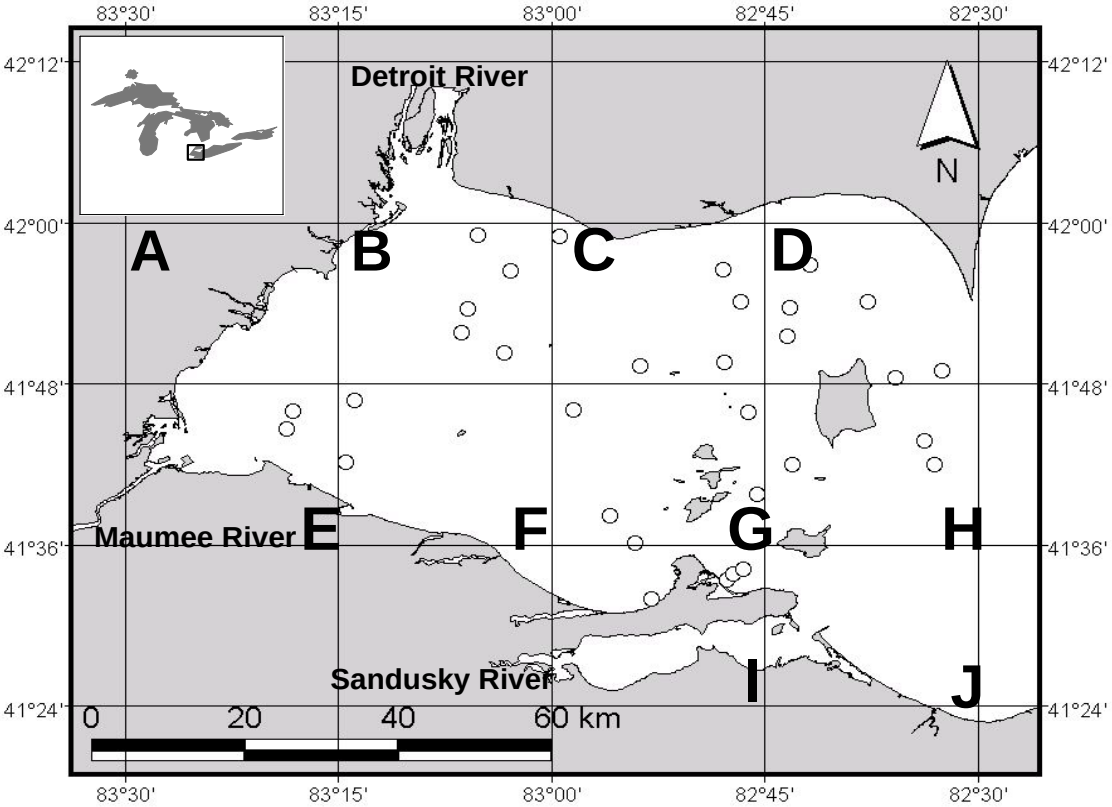

Figure 2.—Grid design used to select age-0 walleye for the area-weighted mixed stock analysis in western Lake Erie, 2003. The area of open water (non-filled regions, Table 2) was used to determine how many fish to include in the subsample for each quadrat.

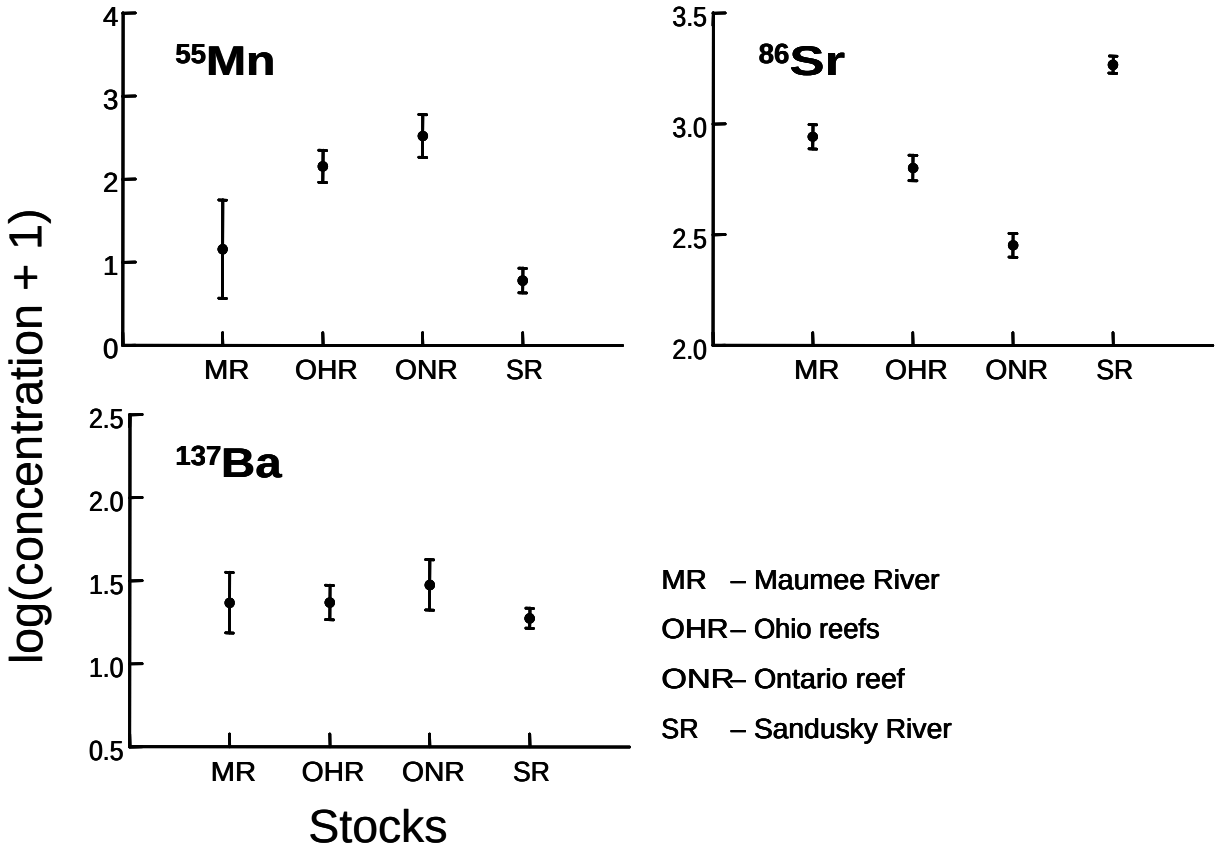

Figure 3.—Mean (± 2SE) concentration of elements for the four dominant walleye spawning stocks in western Lake Erie.

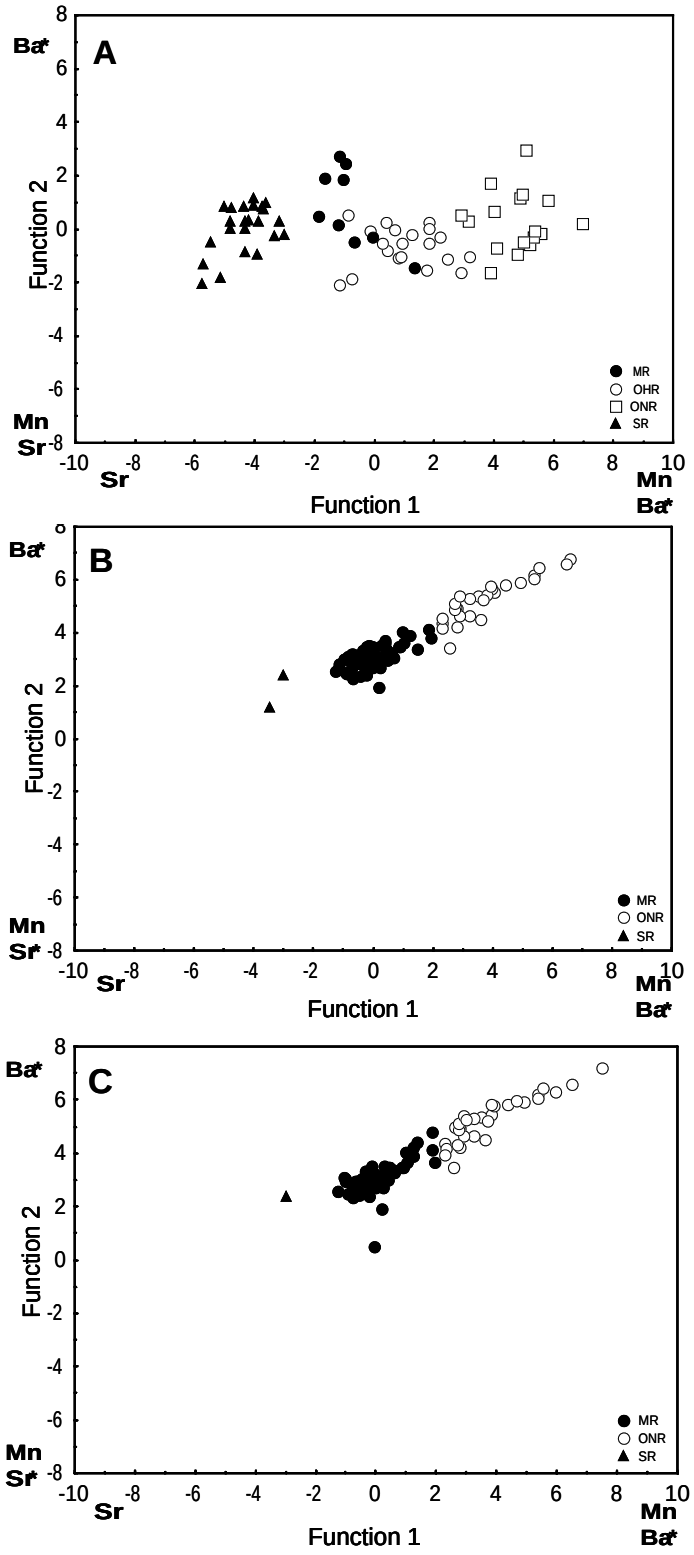

Figure 4.—Scatterplot of the canonical scores for: larval walleye and age-0 walleye from the abundance-weighted and area-weighted samples in western Lake Erie in 2001. The element labels indicate the direction of the association with the functions. Elements marked with an asterisk indicate non-significant contributions to the function.

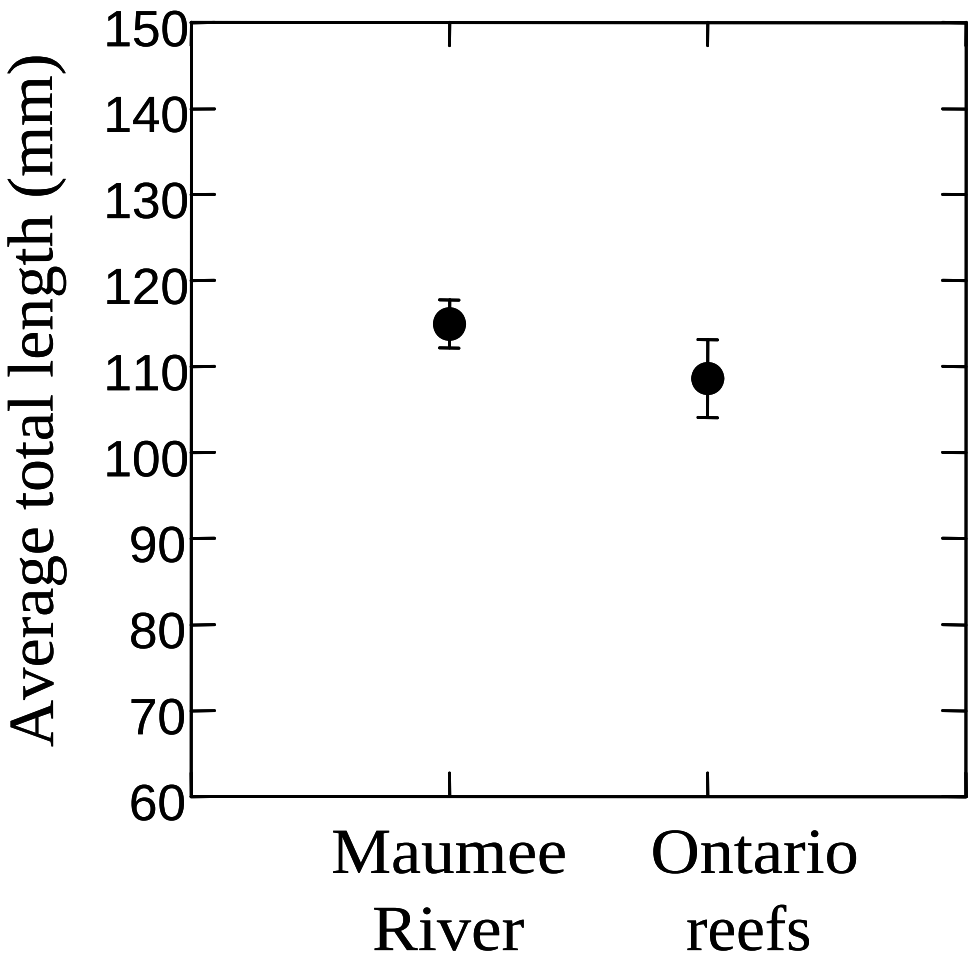

Figure 5.—The difference in mean size (TL) of August age-0 walleye identified as originating from the Maumee River and the Ontario reefs. Whiskers indicate two standard errors about the mean.

DELIVERABLES

This project provided partial support to one M.Sc. student (Sarah Bartnik) whose research described the factors regulating walleye production in Lake Erie. In addition to the completion of the M.Sc. thesis (March 2005), a manuscript relaying the results of the mixed-stock analysis is in preparation for publication. Prior to the defense of the M.Sc. thesis, preliminary results from the mixed stock analysis were presented in an oral presentation entitled "Mixed stock analysis of a walleye cohort in the western basin of Lake Erie" at the 47th Annual Conference of the International Association for Great Lakes Research. Upon completion of the study regarding the uptake mechanisms of young fish, a separate manuscript will be prepared for submission and publication.

APPENDIX

Oral presentation abstract:	Mixed stock analysis of a walleye cohort in the western basin of Lake Erie
Presented at:	47th Annual Conference of the International Association for Great Lakes Research, May24-28, 2004, Waterloo, Ontario.

Lake Erie walleye are thought to consist of stocks associated with geographically distinct spawning areas within the lake. Local variation in the production from individual spawning areas may explain the patterns of juvenile walleye production observed for the entire western basin. To explore whether differential contribution is occurring among stocks, we conducted a mixed stock analysis using juvenile (age-0) fish collected in August 2003. These fish were obtained from a depth-stratified random bottom-trawling program covering the entire western basin of Lake Erie (n = 76 sites). Discriminant function analysis was used to assign the juvenile fish back to their natal stock. We discuss how the relative contribution of the different stocks to the lakewide population can influence management decisions concerning harvest restrictions or habitat restoration.