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SHORT PAPER

Relationships between Length-Weight, Age, and Body Condition of the Round Goby *Neogobius Melanostomus* (Pallas) in a Tributary and Harbor Embayment in Southern Lake Michigan, USA

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Abstract

Gender relationships between total length (*LT*) and standard length (*LS*) (mm) were compared to weight (*MB*)(mg) in the round goby, *Neogobius melanostomus*, for tributary and harbor embayment habitat in southern Lake Michigan. Length and weight relationship (n = 413), *LT* length-frequency distribution, and sex ratios were determined. A strong positive correlation (P>0.001) was found between length (*LT* and *LS*) and weight for both males and females. Male populations showed positive allometric growth rates with b-values and Fulton Condition indices above 3.0, females populations showed positive allometric growth in the tributary populations, but a negative allometric growth rate in the harbor population.

Keywords: Growth, Fulton's condition factor, relative mass index, Laurentian Great Lakes.

Introduction

The round goby, *Neogobius melanostomus* Pallas, is a benthic fish native to the Black and Caspian Seas of southeastern Europe that was introduced to the Laurentian Great Lakes region in 1990 to North America through ballast water transfer in the Detroit River, near Detroit, Wayne County, Michigan, USA (Jude *et al*., 1992). It first appeared in Lake Michigan in 1992 at Calumet Harbor near Chicago, Cook County, Illinois, USA. Since its introduction to the Great Lakes, the round goby has spread rapidly (Charlebois *et al*., 2001; Clapp *et al*., 2001) and has significantly altered the local ecosystem (Schaeffer *et al*., 2005; Kipp *et al*., 2012; Kornis *et al*., 2012), of southern Lake Michigan.

The successful colonization of the round goby in North America has been attributed to its ability to spawn multiple times per season, male nest protection, its high survival rate in areas with low water quality, and its ability to often outcompete native benthic fish species such as the mottled sculpin (*Cottus bairdii*), johnny darter (*Etheostoma nigrum*), and logperch (*Percina caprodes*) (French and Jude, 2001; Janssen and Jude, 2001; Balshine *et al*., 2005). It is a food source for gamefish such as the smallmouth bass (*Micropterus dolomieu)*, yellow perch (*Perca flavescens)*, walleye (*Sander vitreus)*, and lake trout (*Salvelinus namaycush)*. In the Laurentian Great Lakes the round goby is considered a non-native, introduced, invasive species due to its rapid spread and negative effects on ecosystem equilibrium and economy. The success of the species has been attributed to its consumption of zebra and quagga mussels, which are both introduced species. Zebra and quagga mussels comprise a significant portion of its diet, but may be overestimated in its importance in the diet composition (Brush *et al*., 2012; Thompson and Simon, 2013).

The Lake Michigan basin is the third largest Laurentian Great Lake and sixth largest lake worldwide based on surface area with an average depth of 85 m (USEPA and Environment Canada, 1995). The southern Lake Michigan basin has a mixture of diverse land uses including industrial and natural landscapes (Simon and Stewart, 1999). The study area includes the original dispersed location of the round goby from its North American introduction in Calumet Harbor, which is along the Lake Michigan breakwall at the entrance to the harbor (Jude *et al*., 1992). Comparisons were made between the size of individuals from the original harbor site and individuals in nearby recently colonized tributaries. Individuals from two tributary locations from the Grand Calumet River Area of Concern were included from the East Branch Grand Calumet River upstream of Clark Road in Gary, Lake County, Indiana, USA, and West Branch Grand Calumet River west of

Indianapolis Boulevard, East Chicago, Lake County, Indiana, USA (Figure 1).

In order to effectively manage introduced species, basic life history information is needed to evaluate patterns in life history strategies. Relationships between length and weight in the round goby in both native and introduced portions of its range has shown differences in maturity, fecundity, longevity, and mean generation time (Thompson and Simon, 2015). Limited information is available on population differences between tributary and harbor habitats and less is available from Great Lakes Areas of Concern (Poos *et al*., 2009). The second wave of invasion into tributary streams of the Laurentian Great Lakes has cause for concern based on life history strategies used by the round goby and the potential dispersal into the Mississippi River basin. We evaluate patterns in growth in both length and weight, sex ratio, and body condition using the Fulton Condition Index between the original source populations in Calumet Harbor and newly invaded habitats in a tributary of Lake Michigan.

Materials and Methods

Fish were collected during daylight hours in areas associated with an anthropogenic concrete breakwall including varying strata in the harbor and coastal shoreline of Lake Michigan and from two riverine habitats. Lake habitats are associated with steel sheet piling and sand beaches (depth ≤ 3 m). Riverine habitat types included rip-rap, large boulder habitat associated with an anthropogenically modified shallows, sand and muck substrates in the river itself with concrete erosion barriers (depth = 2–5m), and open lake deep waters in the nearshore (depth<5m) around the breakwall. River habitats included natural and artificial substrates including sand, riprap, and large cobble particle sizes similar to those defined by Moran and Simon (2013).

Active sampling methods used for capture at deep breakwall habitats included angling with spin cast fishing rods with 3-5 hooks that had steel shanks that were 10mm long with #10 snelled bait hooks evenly spaced 1 m apart and 4m common sense minnow seine with 3.2 mm mesh size. Minnow seines were used to ensure capture of individuals of the smallest age 1 year class (Dopazo *et al*., 2008), which were seined along the beach outside the Calumet Park breakwall. Individuals collected from tributary sites used boat mounted DC electrofishing gear with a 3500 watt generator and 2-3 watt amperage. The collected *Neogobius melanostomus* were immediately anaesthetized in MS-222, then preserved in a 10% formalin solution and later transferred to 70% ethanol. Electrofishing was conducted between Indianapolis Boulevard and Roxanna Marsh and in the East Branch upstream of the Clark Road Bridge.

Fish were identified by sex and measured for length and total mass using a digital caliper and ocular fit micrometer for small individuals and a Sartorius analytical balance for weight. Size distribution, age composition, and sex ratios were then calculated. Individuals selected for analysis were randomly chosen individuals distributed throughout the entire size range. Due to the precision of the equipment used in the analysis it would have been impractical to attempt to measure live individuals in the field. While the data represented may not completely reproduce the length-weight relationships of live individuals, it is assumed that because each specimen was preserved using the same procedure, the overall relationship between length and weight would be analogous.

In the laboratory, two measures of length, total length [L_T] and standard length [L_S], were used in the

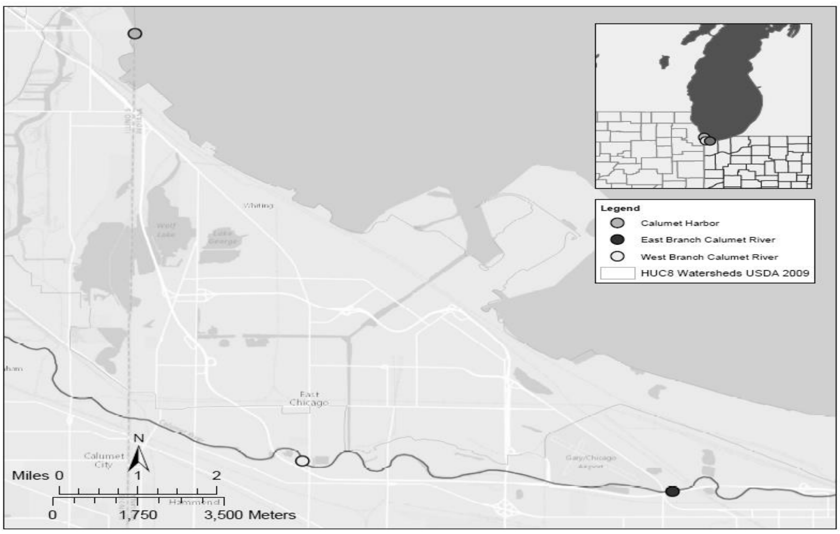

Figure 1. Study area showing the Calumet River in northeastern Illinois. The original introduction reach and two river locations in the Grand Calumet River Area of Concern were surveyed during August to October 2014 and 2015.

analysis. These correlations can be used to determine the health and body condition of each round goby population associated with Lake Michigan and tributaries. The current study hypothesized that a difference in growth would occur between males and females. We will utilize this information to assess whether round goby growth patterns are influenced by sex in southern Lake Michigan. Individuals were weighed using a Sartorius balance with a precision of 0.1 mg and measured with digital calipers with a precision of 0.01 mm. Sex was determined by observing either the tubular gonopod in males or a cloaca with a short ovipositor in females. Both measurements of length were recorded to the nearest 0.01 mm. The methods for measuring the length and weight of the specimens followed standard methods (Hubbs *et al*., 2004). The SL was measured along the horizontal body axis from the tip of the snout to the base of the hypural plate, while TL was measured along the horizontal axis from the tip of the snout to the tip of the depressed caudal fin. Simple linear regression analysis was used to determine if a significant relationship was observed between length and weight in round goby (Zar, 2010). The length and weight measurements were log normalized and graphed using a best fit trend line. We use linear regression on the normalized data to evaluate body condition and the Fulton Condition Index (Ricker, 1975; Nash *et al*., 2006). Gender based populations with slope > 3 have positive allometry, < 3 have negative allometry, while a b = 3 is isometric. Positive allometry indicates that weight is gaining faster than length, while negative allometry indicates length is gaining faster than weight. Gender based trend lines determined best-fit regression models and residuals (r^2) graphed for L_T (mm) and M_B (g) (Nash *et al*., 2006). To determine the effects of sex on the growth rate, a Student t-test was used to compare relationships in body weight and length (L_S and L_T) between females and males and between harbor and tributary populations (Zar, 2010). To evaluate patterns in habitat degradation on body condition and length-weight ratios, a Student t-test was used to compare male and female round goby individuals from the East and West Branches of the Grand Calumet River and Calumet Harbor.

Age relationships were determined using cohort analysis and size modeling (Froese and Binohlan, 2000). Individuals were aged by using a frequency distribution analysis of total length (Nielsen and Johnson, 1996). Individual L_T was displayed based on 3 mm size intervals from the smallest to the largest individual collected during this study. Length frequency distribution is considered a standard approach for evaluating size at age categories in fish (Nielsen and Johnson, 1983). Age groups of round goby for harbor and tributary habitats were determined using length-frequency distribution analysis, where ages were decided according to elevated peaks in the distribution. Normal curve size distributions represent hypotheses of estimated age class in the population, and are consistent with hard structure techniques using scales or otoliths. We did not have age 0 individuals in this analysis based on the known reproductive season, so it is anticipated that age 0 individuals are likely under-represented in this dataset.

Body condition was determined using Fulton's condition factor (K), which was calculated from the equation (Ricker, 1975):

$$K = M_B\ L_T^{-3} \times 10^{5}$$

where, M_B is the weight of the individual, L_T is the TL of the individual. Fulton's condition factor (*K*; Nash *et al*., 2006) is a measure of fish health using mass (Ricker, 1975). It assumes that *M*B is proportional to the cube of the *L*T. The interpretation of population health using the *K* value is based on the slope factor (*b*) for the population. If $b < 3$, the population is considered healthy as the population has uniform condition throughout the L_T range (Froese, 2006). The value of *K* was calculated for anthropogenically modified harbor and river habitats. *F* tests were used to compare growth between sex and habitat. Simple linear regression evaluated relationships between the L_T and M_B by sex. All statistical analyses were performed using Microsoft Excel for Windows 2011 (MicrosoftCorp., Excel for Windows OS X. 14.3.0; www.microsoft.com/windows) and StatPlus (AnalystSoft Inc., StatPlus:windows-statistical analysis program for Windows OS. 2009; www.analystsoft.com/en). Figures and tables were created using Microsoft Excel for Windows 2011 (Microsoft Corp., Excel for Windows OS X. 14.3.0).

Results

Round goby from the tributary (East Branch of the Grand Calumet River (n=144) and West Branch of the Grand Calumet River (n = 119)), and the original invasion site in Calumet Harbor (n = 150) were evaluated to determine the effect of sex and habitat on length-weight relationships (n = 413) in southern Lake Michigan populations. There was no significant difference in L_S (T-test, F = 1.56, P = 0.064) or L_T (F = 1.56, P = 0.0636) between males and females.

Male round goby individuals showed a significant positive correlation between L_T and M_B (r^2 = 0.981, F = 5068.73, d.f. = 276, P<0.001; Figure 2a) and between L_S and M_B (r^2=0.985, F=6288.45, d.f.= 76, P<0.001; Figure 2b). Females showed a positive correlation between L_T and M_B (r^2=0.991, F=5405.34, d.f. =135, P<0.001; Figure 2c) and L_S and M_B (r^2 = 0.990, F= 4661.57, d.f.=135; P<0.001; Figure 2d). Combined population data for both sexes found a strong relationship between L_T and M_B (r^2 = 0.985, F = 9623.23, d.f. = 412, P<0.001; Figure 3b) and between L_S and M_B (r^2 = 0.986, F=10931.82, d.f. = 412, P<0.001; Figure 3a), which was consistent with

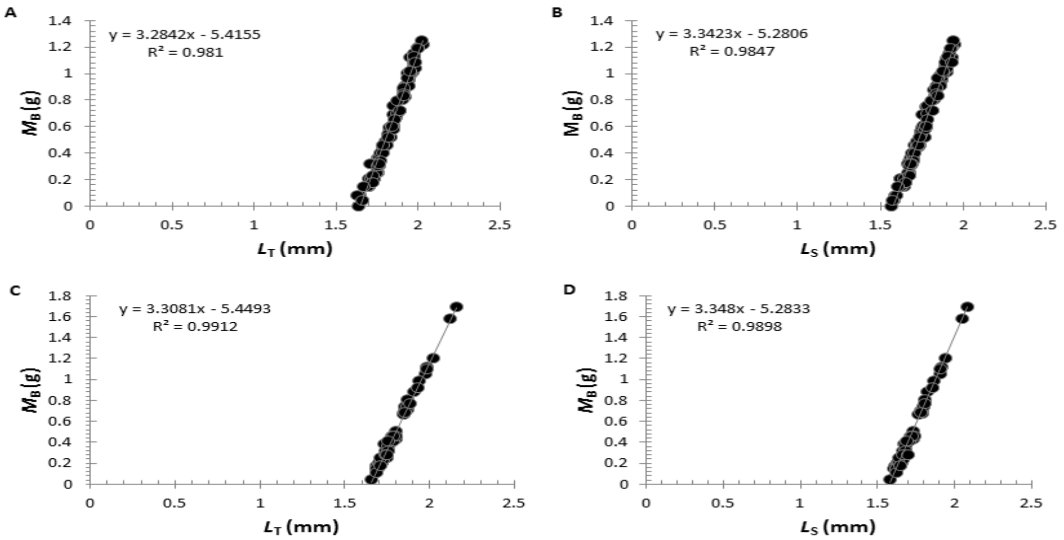

Figure 2. Log normalized linear trend line for total length (L_T) and total mass (M_B) relationships for (a) male and (c) female *Neogobius melanostomus* from the southern basin of Lake Michigan from August 2014 to October 2015. The curves were fitted by (a) y = 3.2842x - 5.4155 (r^2 = 0.981) and (c) y = 3.3081x - 5.4493 (r^2 = 0.991). Log normalized linear trend line for standard length (L_S) and total mass (M_B) relationships for (b) male and (d) female. The curves were fitted by (b) y = 3.3423x - 5.2806 (r^2 = 0.985) and (d) y = 3.348x - 5.2833 (r^2 = 0.990).

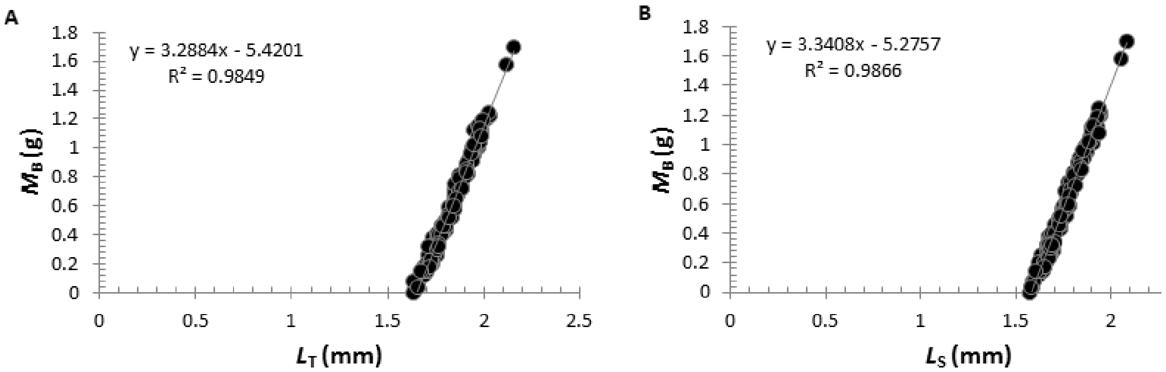

Figure 3. Combined population data for both sexes found a strong relationship between L_T and M_B (r^2 = 0.985, F = 9623.23, d.f. = 149, P<0.001) and between L_S and M_B (r^2 = 0.987, F = 10931.82, d.f. = 149, P<0.001), which was consistent with the individual sexes.

the individual sexes. There was no significant difference between sexes.

River and harbor populations including both sexes exhibited a strong relationship between L_S (F = 10931.82, d.f. = 412, P<0.001) and between L_T and body weight (F = 9623.23, d.f. = 412, P<0.001). Females ranged from 44.92–143.04 mm L_T and males ranged from 42.67–107.14 mm L_T. The body weight of males ranged from 1.0-17.70 g, while females ranged from 1.10-50.04 g. The sex ratio of males to females was 1.0: 0.5. Growth patterns of male and female round goby did not differ significantly in L_S (F = 1.56, P = 0.064) or L_T (F = 1.56, P = 0.0636); however, there was a significant difference observed in the body weight M_B between males and females (F = 4.35, P<0.001).

Comparison of individuals in the Grand Calumet River and Calumet Harbor showed **s**tatistically significant differences in L_T (F = 3.63, P<0.001), L_S (F = 3.67, P<0.001) and M_B between river and harbor populations (F = 4.04, P<0.001). Female round goby showed a significant difference in both L_T (F = 7.69, P<0.001), L_S (F = 8.03, P<0.001), and M_B between the river and harbor populations (F = 90.13, P<0.001). Male round goby individuals showed no significant difference in L_S and L_T, while a significant difference in M_B (F = 4.87, P<0.001) was observed between the river and harbor populations.

Age classes ranged from 0+ to 3+ years with the majority (96.8%) being age 1 and age 2 years. Males did not attain age 3 in harbor populations (Figure 4). The L_T and M_B relationships showed that both sexes grew exponentially with male growth rate increasing faster than female. L_T-at-year class composition with a June hatch date showed that during the study period, age classes were age I (17 months) (mean ± s.d. = 57.52 ± 7.92 mm, range: 42.7–71.8 mm), 29 months (mean ± s.d. = 86.59 ± 9.03 mm, range: 65.6–104.5 mm) and 41 months (mean ± s.d. = 137.3 ± 8.12 mm, range: 131.6–143.0 mm).

There was a significant difference observed in the M_B between males and females (t-test, F = 4.35, P<0.001). Population growth was at a positive allometric rate for males in Calumet Harbor, Grand Calumet River, and Southern Lake Michigan and females in the Grand Calumet River and Southern Lake Michigan (Table 1). Females from the harbor population grew at a negative allometric rate. There was no significant difference in the Fulton Condition Index (*K*), a body condition index, between males and females (Figure 5). The relationship between Fulton Condition Factor (K) and L_T was explained by the logarithmic equation, $K = 5.22225e^{-0.121\,(L_T)}$ (r^2 = 0.969, $P > 0.001$).

Discussion

The second introduced population of round goby found in North America was in the Calumet Harbor at the mouth of the Calumet River, Cook County, Illinois (Jude *et al*. 1992; Charlebois *et al*. 1997). Limited information on population dynamics for southern Lake Michigan exists, including length-weight relationships, sex ratio, and age and growth structure (Charlebois *et al*. 1997; Cooper *et al*. 2009).

Positive relationships between length (L_S and L_T) and weight (M_B) infer that round goby exhibit positive allometric growth patterns. The lack of significant differences in length between males and females indicate that round goby do not have a growth pattern that is influenced by sex. The significant difference between weight in males and females may be a result of the sample size of female round goby in the study or potential life history strategies favoring larger sizes in females to maximize reproductive fitness.

Gutowsky and Fox (2011) found male and female ratio to highly favor males in southeastern Ontario, Canada. Male outnumbered female 1:0.455, while the original introduced population was slightly higher with a ratio of 1: 0.714. The sex ratio was expected to be close to 1:1, representing an even distribution of males and females. This study instead found the total population ratio to be 1 : 0.5, with a higher ratio of males to females in Calumet Harbor (1

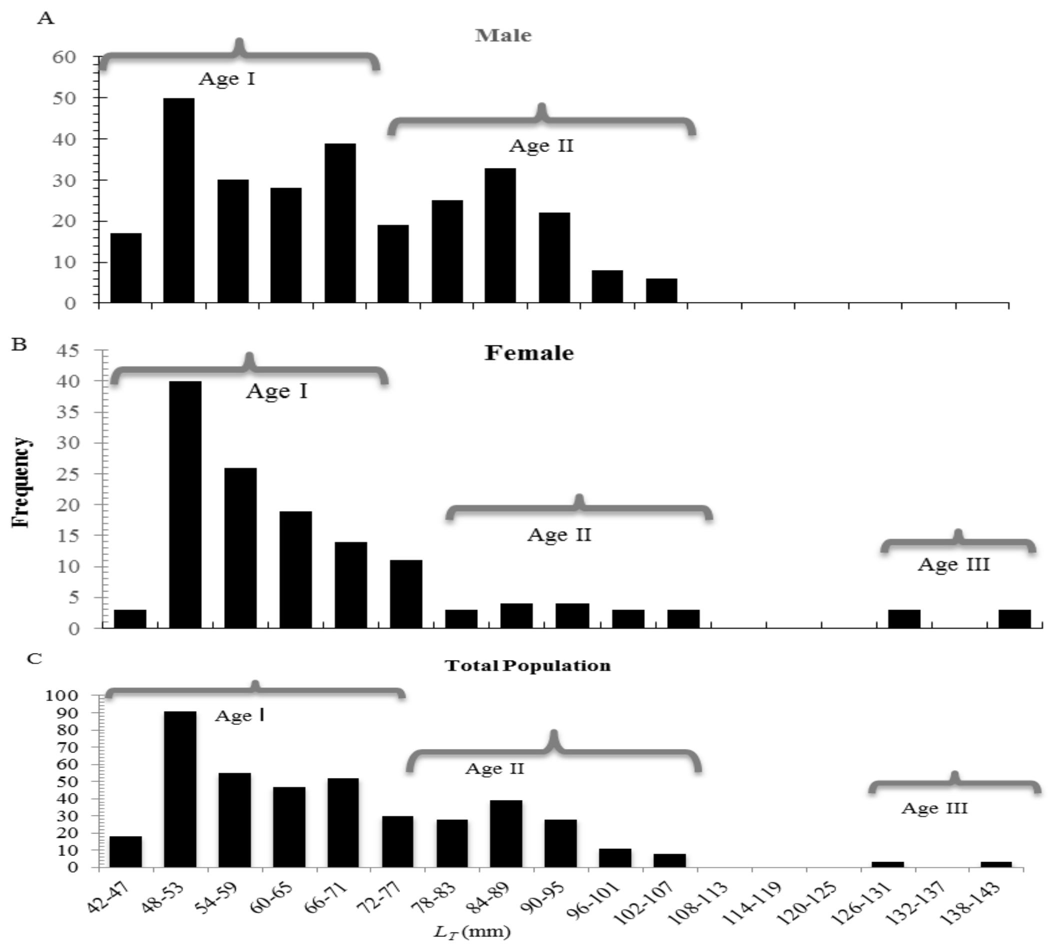

Figure 4. Total length (L_T) frequency distribution of *Neogobius melanostomus* in southern Lake Michigan August-October 2014. (a) males, (b) females), and (c) total population.

Table 1. Comparison of length (L_T) at age for introduced round goby (*Neogobius melanostomus*) populations in the Laurentian Great Lakes

Basin	Location		0	1	2	3	4	5	6	*b*	Study
Lake Huron	Hammond Bay*	Male		45	72	87	98				French & Black (2009)
	Harbor Beach*	Male		40	61	77	89	98			French & Black (2009)
Lake Erie	Upper Detroit River	Male		76	91						MacInnis & Corkum (2000)
		Female		70	78	100					
	Pennsylvania tributary streams	All	44	68	80	94					Phillips *et al.*(2003)
	Central Basin	Male	54 - 55	74-76	104	176-6					Johnson *et al.*(2005*a*)
		Female	49	70-72							
	Bass Islands (July)	All	16	34	56	77					Thompson &Simon (2015)
	Trent River	Male		47	67						Gutowsky & Fox (2011)
		Female		48	65						
Lake Michigan	Lake Michigan	Male			71	84	94	108			Huo *et al.*(2014)
		Female			70	80	91	99	111		
	Grand Calumet River	Male		42-61	65-107					3.412	This study
		Female		45-77	74-106	128-143				3.002	
		All		42-77	65-107	128-143				3.180	
	Calumet Harbor	Male		42-71	69-96	96-101				3.787	This study
		Female		45-74	72-106					2.513	
		All		42-74	69-106	96-101				3.377	
	Southern basin Lake Michigan	Male		42-67	65-107					3.284	This study
		Female		45-77	72-106	128-143				3.308	
		All		42-85	69-107	128-143				3.288	

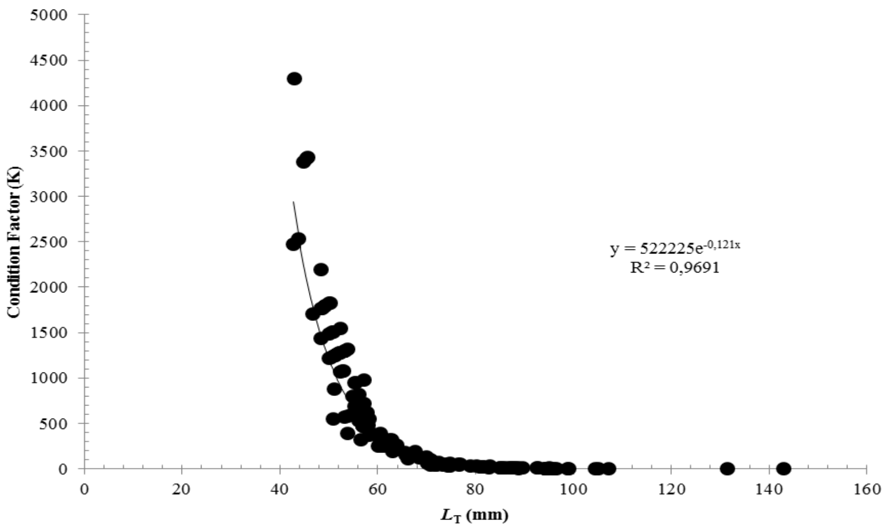

Figure 5. Fulton Body Condition index (K) relationship with L_T (n = 413) for individuals from the Grand Calumet River and Calumet Harbor.

: 0.642) than in the Grand Calumet River (1 : 0.34). Gutowsky and Fox (2011) considered this to be a density dependent intraspecific competition relationship.

Reproduction strategies used by round goby suggest that fewer males would be expected based on the polygyny reproductive mode (Simonovic *et al*. 2001; Kornis *et al*. 2012); thus, each male can reproduce with multiple females. The increased male dominated sex ratio may be required for nest guarding. This would suggest that males should be larger than females and greater in number than females; however, there is no significant size difference between males and females. Males are much more vulnerable to predation during reproductive periods due to nest guarding duties (Stammler and Corkum 2005). The number of males is statistically higher than females, which may be due to the need to replace males that expire due to competition, exhaustion, and increased mortality (Charlebois *et al*. 1997; Kornis *et al*. 2012). An alternate explanation may involve hermaphroditism, which has been reported in goby genera *Paragobiodon* (Lassig 1977), *Gobiosoma* (Robertson and Justines 1981), *Coryphopterus* (Robertson and Justines 1981; Cole 1983; Cole and Shapiro, 1990), and *Evidota* (Cole 1990), with large females switching sex when males are reduced in number.

The value of parameter *b* ranged from 2.513 to 3.787, which is within the expected 2.5–3.5 range according to Froese (2006). All populations studied grew at a positive allometric rate, which increases weight at a higher rate than length. Only the female Calumet Harbor population grew at a negative allometric rate and increased length at a rate higher than weight. Although there are certain exceptions, the parameter *a* value can be used as an indicator of the body shape of a fish (Froese, 2006). In this study, the body shape of most species was classified as elongate trending away from fusiform based on Froese (2006). Because the fishes in this study inhabit standing waters, body shapes did not need to be streamlined.

No significant differences were expected in length and weight between goby populations in the Grand Calumet River and Calumet Harbor since the sites are in close proximity and should allow fish to migrate freely. However, significant differences in all growth variables between the populations were observed. There were significant differences in both measures of length and weight for females between the river and harbor populations, while males only differed in weight. The significant difference in growth between populations for females may be skewed by the female sample size of round goby in the study. This length and weight difference may suggest density dependent regulation in the Calumet Harbor population based on intraspecific competition, which puts stress on the fish reducing its size due to limited resources. Once the population reaches maximum capacity, the population expands to new areas, leading to more resources, less competition and larger body size and mass. An alternate hypothesis would be that the original source population has reached carrying capacity and are stunted in growth, while expanded populations in tributary reaches have access to greater resources and uncontested habitat. A third hypothesis may be the reduced predation pressure in newly colonized habitats may enable greater growth and individual longevity to increase.

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