

The fecundity and gonad development cycle of the round goby  
(*Neogobius melanostomus* Pallas 1811) from  
the Gulf of Gdańsk

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**Abstract**

The round goby (*Neogobius melanostomus* Pallas 1811) is a non-native species in the Gulf of Gdańsk. The aim of this article is to assess the absolute fecundity and attempt to describe the gonad development cycle of round goby specimens from the Gulf of Gdańsk. The stages of gonad development were classified according to the modified Nikolski (1963) scale for perciformes, which was adapted for the round goby. The sex ratio (female to male) in the population was 3:5. Spawning was most intense during April and July, and there was a pause in June. Fish body size determined fecundity.

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## INTRODUCTION

The round goby (*Neogobius melanostomus* Pallas 1811) is a non-native species in the Gulf of Gdańsk, where it was noted for the first time in 1990 (Skóra and Stolarski 1995). The natural habitats of the round goby include the Black and Caspian seas. This species was probably transported to the Baltic Sea in ballast waters of ships using the inland route from the Black or Caspian seas (Skóra and Stolarski 1996). Since the first sighting, the round goby has increased both its area of distribution and biomass (Chotkowski and Marsden 1999, Skóra et al. 1999, Corcum et al. 2004, Sapota 2004, Wandzel 2003).

This species has also been introduced into the waters of the Great Lakes in North America (Crossman et al. 1992, Jude 1997).

The reproductive biology of *N. melanostomus* has been described by numerous authors including Bilko (1968), Kovtun (1977), Kulikowa & Fandeeva (1976), Michman (1963), Moiseyeva (1983), Tkaczenko (1979), Corcum et al. (1998), MacInnis & Corcum (2000), and Wandzel (2000, 2003).

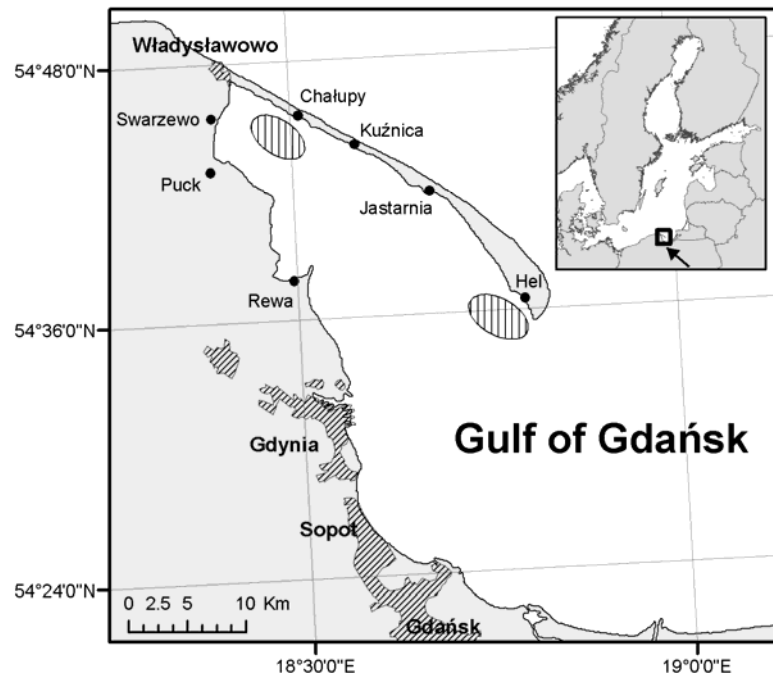
Reproductive strategy plays a significant role during biological invasions, and the majority of successful colonists have a short generation time, high fecundity, and rapid growth rates (Lodge 1993).

The round goby is a batch spawner, which means that females do not spawn all their eggs at one time, but release them in portions throughout the reproductive season. The spawning cycle is repeated five to six times throughout the year at intervals of approximately 17-28 days (Kovtun 1977, Kulikowa 1985, Charlebios et al. 1997, Corcum et al. 1998). The length of the interval between subsequent batches depends on water temperature; higher temperatures mean that intervals are shorter (Charlebios et al. 1997).

The aim of the current study was to determine the fecundity and describe the gonad maturation cycle of *N. melanostomus* specimens from the Gulf of Gdańsk.

## MATERIAL AND METHODS

The fish were collected in July, August, and October 1999 and from March to August 2000 in the vicinity of the village Chałupy and in the fishing harbor of the city of Hel (Fig.1). The fish were caught with fyke-nets and then frozen for further analyses. After thawing, the following measurements were taken: total length (mm); fish weight (to the nearest 0.1 g); gonad maturity stage; gonad weight (to the nearest 0.001 g, females – whole gonad, males – testis and cement gland separately); egg diameter (to the nearest 0.0001 mm). The sex ratio, which is the percentage of males and females in the sample, was



**Fig. 1.** Sampling areas.

determined. The age structure of the sample was determined with otoliths (sagittae).

The eggs were separated from the ovaries by freezing them at a temperature of  $-20^{\circ}\text{C}$  and then thawing them in 80% ethanol (Bleil & Oberst 1993). The eggs were counted and measured, and the maturity stage was determined according to the modified Nikolski scale for perciformes (Nikolski 1963). A five-stage, modified scale was used in the present work.

In order to make the scale more precise for the gobies, the male testes were viewed under magnification and the following descriptive scale was established:

1. juvenile;
2. clearly visible, light beige or slightly grey, not too supple;
3. clearly visible, light beige color but darker than in stage 2, supple;
4. clearly visible, darker than in stage 3 - beige, grey-beige to grey with possible red staining on the edges, supple, engorged;
5. spent (gonad empty).

The maturity stage of the second part of the male gonad, the cement gland, was also determined according to a scale compiled from macroscopic observations:

1. absent (if the testis is in at least stage 2);
2. substantially smaller than the testis, opaque;
3. smaller than the testis, clearly visible, not opaque, grey to beige;
4. approximately the same size as the testis, grey to beige, darker than in stage 3, more supple;
5. absent (if the testis is not present – stage 5 – spent).

Determining female maturity included categorizing the eggs according to the classification presented in Table 1 and measuring the size of the eggs under a stereoscopic microscope (to the nearest 0.0001 mm).

**Table 1**

Ovary maturity stages

STAGE	Characters to determine gonad maturity in females	
1	poorly developed, yellow or pink colored, not clearly visible, sometimes difficult determine sex	
2	small, graves visible, ovaries transparent	egg $\varnothing \leq 0.7589$ mm
3	ovaries yellow or reddish, eggs not transparent, first hydrated oocytes	$0.7589$ mm < egg $\varnothing$ < $2.428$ mm
4	very well-developed graves, eggs transparent, ovaries yellow or reddish, leakage stage	egg $\varnothing \geq 2.428$ mm
5	ovary almost empty with single egg graves, ovaries flabby, reddish	

As the round goby is a batch spawner, the eggs in the ovaries were divided into two fractions referred to as IR (the fraction of eggs in a higher developmental stage) and IIR (found at the same time in the gonad, but in an earlier developmental stage).

The standard gonadosomatic index (GSI) (Ricker 1975) was determined in order to describe changes in gonad development in the males and females.

$$GSI = \frac{B}{W} \times 100 \quad (1.1)$$

where:

- B – gonad weight (g)  
W – fish weight (g)

The average GSI for each gonad developmental stage was compared using nested ANOVA for both sexes.

Relationships between fecundity and length (1.2) and weight (1.3) were estimated in order to determine the round goby reproductive potential.

$$F = \alpha L t^{\beta} \quad (1.2)$$

where:

F – fecundity  
Lt – total length  
 $\alpha$  and  $\beta$  - coefficients

$$F = a + bW \quad (1.3)$$

where:

F – fecundity  
W – fish weight  
a and b – coefficients

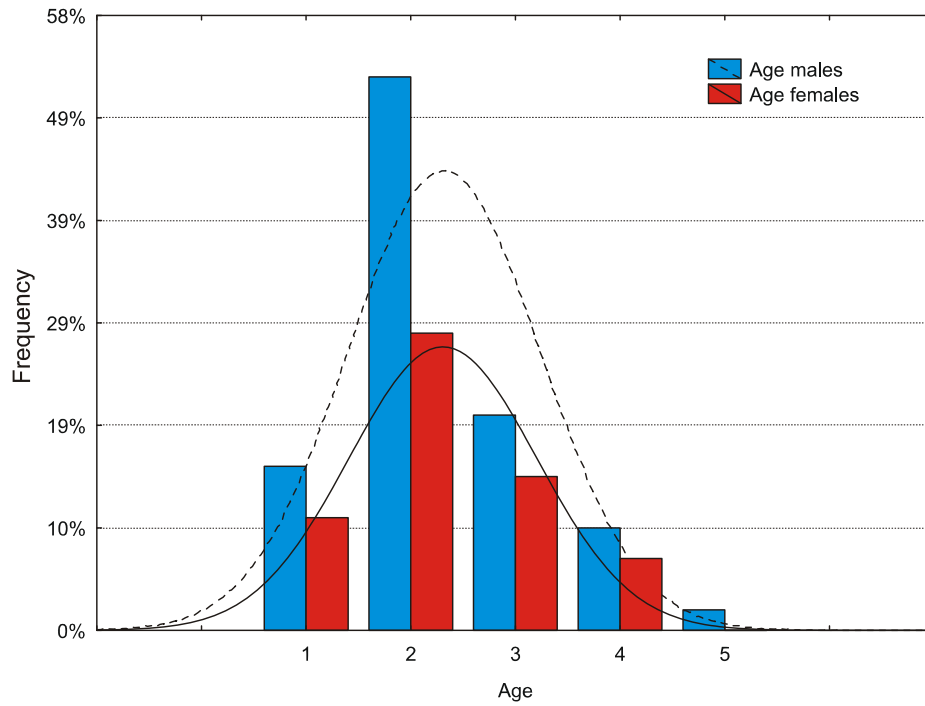
Fecundity-length and fecundity-weight relationships were tested for both egg fractions; only those explaining more than 50% of the variance were selected for further analysis. STATISTICA software was used to perform statistical analyses (StatSoft, Inc. 2001).

## RESULTS

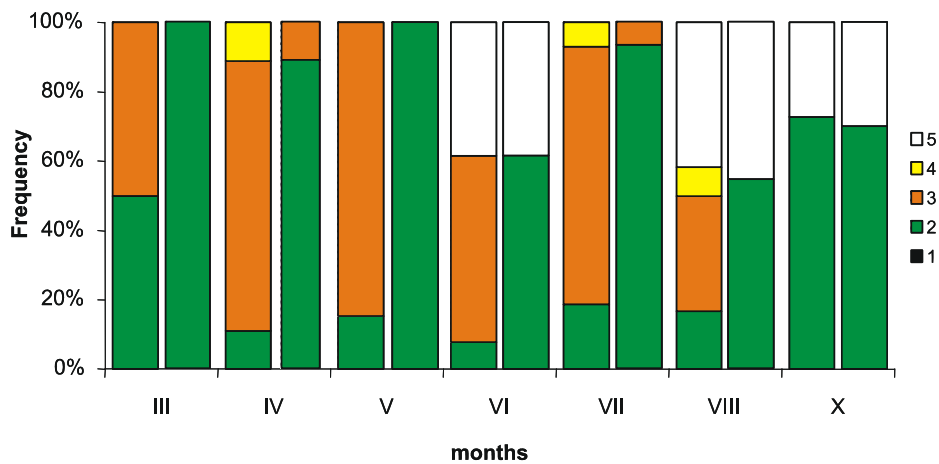
A total of 166 individuals of *N. melanostomus* comprised of 104 males and 62 females were examined. The ratio of females to males was approximately 3:5. The domination of males was less pronounced in spring (April), while in August the numbers of males and females were equal. While the age class 2 dominated among both sexes (Fig. 2), age class 1 also appeared in the samples.

### *Stage of gonad maturity*

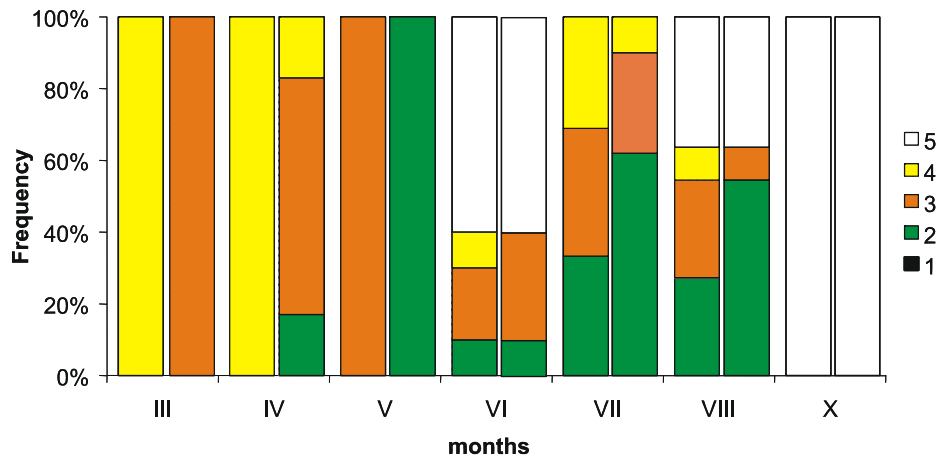
Males are capable of reproducing for an extended period from early March to September. The percentage of specimens with testes in maturity stages 2 and 3 is high in March. In April specimens in stage 4 appeared, while in May most of the specimens were in stage 3. Specimens in stage 5 (spent) comprised a significant share of the fish in June and August. Specimens in stage 4 were noted in July and August. Only fish in stages 2 and 5 occurred in October at a ratio of approximately 7:3 (Fig. 3). No specimens in developmental stage 1 were noted.



**Fig. 2.** Frequency of particular age classes.



**Fig. 3.** Proportion of testis (left bar) and cement gland (right bar) developmental stages by month.



**Fig. 4.** Proportion of ovary developmental stages for the IR (left bar) and IIR (right bar) egg fractions by month.

The cement gland developmental stages observed were lower than that of the testes. During the study season, stage 2 dominated from mid May until mid June, while stage 5 was significant in June, August, and October. A small percentage of cement glands (Fig. 3) were in developmental stage 3 (April, July) (Fig. 3).

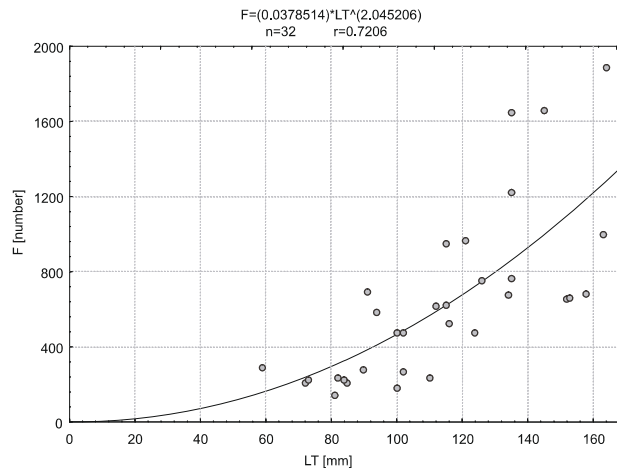
At the beginning of the spawning season (March) the eggs were in stages 4-IR and 3-IIR (Fig. 4). In April, stages 2-IIR and 4-IIR appeared. During May, the gonads contained eggs in stage 3-IR and 2-IIR. In early summer (June), more than half of the round gobies were spent, but fish in stages 2, 3, and 4 were also observed. In July, spent specimens were not noted. The nearly equal frequency of stages 2, 3, and 4 was noted in July for the IR fraction, and stage 5 was noted again in August, when almost 30% of the round gobies were spent. The IR fraction occurred mainly in stages 2 and 3 with a small share of stage 4, while the IIR fraction was dominated by stage 2 with a small share of stage 3. In the fall (October), all the fish were spent (Fig. 4).

### ***Fecundity***

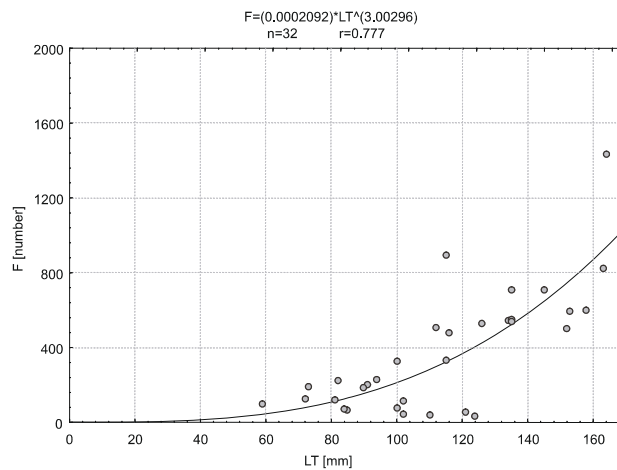
Absolute and relative fecundities were calculated for thirty-two females whose ovaries were in developmental stages 2, 3, and 4. Absolute fecundity ranged from 94 to 2190 eggs, and the mean absolute fecundity was 645 eggs (SD 433). The relative fecundity ranged from 9 to 143 eggs  $g^{-1}$  of female body weight. The mean relative fecundity was 34 eggs  $g^{-1}$  (SD 24) of female body weight.

The fecundity-length relationship is well expressed by a power curve (Fig. 5).

A similar relationship was observed for the IR egg fraction (Fig. 6). However, such a dependence was not noted for the IIR fraction ( $r = 0.09$   $p < 0.005$ ). The number of eggs in the IIR fraction was not correlated with fish length.



**Fig. 5.** Relationship between female absolute fecundity and total fish length (Lt) for both egg fractions.



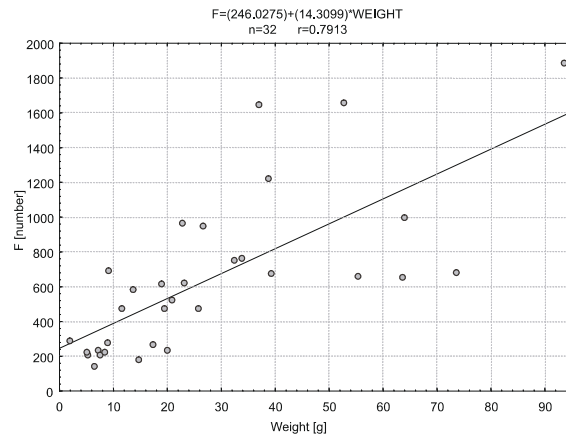
**Fig. 6.** Relationship between female absolute fecundity and total fish length (Lt) for the IR egg fraction.



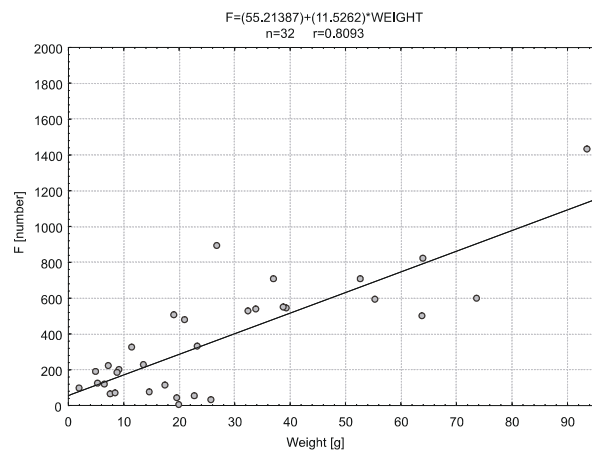
The fecundity-weight relationship is best expressed by a linear equation (1.3) ( $r=0.7913$ ,  $p<0.005$ ). An increase in female body weight of 1 g was accompanied by an increase in fecundity of about 14 eggs (Fig. 7).

There is a significant linear relationship between fecundity and weight for the IR fraction. A weight increase of 1 g caused the number of eggs in the IR fraction to increase by about 11 (Fig. 8).

The dependence between the IIR egg fraction and fecundity and weight was not significant in either the power or linear models.



**Fig. 7.** Relationship between female absolute fecundity and fish weight for both egg fractions.



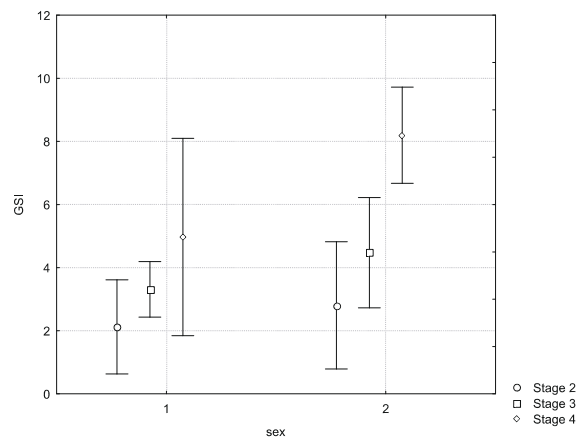
**Fig. 8.** Relationship between female absolute fecundity and fish weight for the IR egg fraction.

### *Dependence of GSI on gonad maturity stage*

In males the highest GSI value of approximately 11 was noted in stage 3 of gonad development. The value of the index in this stage ranged from 0.8 to 11, while in stage 2 it ranged from 0.14 to 9.3. The GSI range for gonad developmental stage 4 was about 1.5 to 8.

The GSI ranges for females were slightly different. In stages 2 and 3, GSI values of between 0.2 and 5 were observed. However, in stage 4 these increased as much as fourfold. In gonad developmental stage 3, the GSI value reached its maximum value of approximately 32.

The ANOVA results demonstrated that the GSI values of females with gonads in developmental stage 4 differed significantly from average GSI values for females with gonads in stages 2 and 3 (Fig. 9, Table 2). No significant differences were observed in males.



**Fig. 9.** Average expected boundary values of GSI for gonad developmental stages (1-males, 2-females). Whiskers - 95% confidence interval.

### DISCUSSION

Although *N. melanostomus* is a new species in the Gulf of Gdańsk, it has already become so widely distributed that it can be considered a permanent element of the ichthyofauna (Borowski 1999, Corcum et al. 2004, Kuczyński 1995, Jackowski 2002, Skóra & Stolarski 1996, Wandzel 2003, Sapota 2004)

In the present work, the 3:5 sex ratio of females to males is markedly higher than the 1:4 ratio of samples collected in 1996 (Skóra & Stolarski 1996) and is similar to results published by Wandzel (2003). In its native waters the sex ratio

of round gobies is approximately 1:1 with females at a slight advantage (Kovtum 1978). However, in the Great Lakes of North America, where it is also an introduced species, the reported ratio was 1:1.9 (Corcum et al. 1998) or even 1.27:1 (McInnis and Corcum 2000). The ratio of males to females in developing populations will probably be advantageous for the males, but this will move towards 1:1 as it is in stable, native populations. As new colonizers, the investigated round gobies should be smaller, mature more rapidly, and have a shorter lifespan than those from the region of the Ponto-Caspian basin; this is due to conditions that differ from their native environment (Corcum et al. 2004). This, however, is not the case in the Gulf of Gdańsk where specimens are larger than those from native regions (Skóra & Stolarski 1996).

While the gonads of both male and female round gobies from the Gulf of Gdańsk are mature from March to August (Fig. 3, Fig. 4), spawning activity intensified from March to May and then from June to August or even September. In comparison with the Ponto-Caspian region, where spawning lasts from April to August (Lindberg et al. 1980), this period is protracted in the Gulf of Gdańsk.

According to Svetovidow (1964) and Tkachenko (1980), in its native regions round goby spawning lasts from early April until the end of August and occurs in waters at temperatures exceeding 12-14°C. In the Baltic Sea, the round goby spawns in waters at temperatures of 17-18°C and with salinity of 8-9 PSU (Skóra 1996). Females from the Gulf of Gdańsk probably also mature more quickly than those from native regions, where females mature at age 2 and males at age 3 (Nikolski 1963). This was confirmed by the observation of age 1 females with gonads in developmental stage 4. The first stage (juvenile) of the Nikolski classification was omitted because even individuals from age class 1 occur with gonads in higher stages.

Egg size categories were determined in relation to the macroscopic gonad descriptions of the Nikolski classification (Nikolski 1963) and were assigned to the corresponding developmental stage. This method allowed for the precise, unequivocal determination of the gonad developmental stage.

Round gobies undertake spawning migrations from deeper zones into shallow, coastal waters at temperatures of 9-26°C (Nikolski 1963). The males

**Table 2**

Standard gonadosomatic index (GSI), nested ANOVA analysis  $p < 0.005$ ,  $MS = 12.515$ ,  $df = 157$ , NIR post-hoc test of ANOVA (ns - not significant)

Males		
Stage	3	4
2	ns	ns
3		ns
Females		
Stage	3	4
2	ns	<b>0.00004</b>
3		<b>0.00184</b>

arrive first followed by the females (Kovtun 1979, Moskal'kova 1996). The round goby is a multiple spawner (Kovtun 1977), and it spawns with multifold intensity (Fig. 3, Fig. 4). One batch of eggs is spawned, followed by another at an interval of approximately 28 days (Kulikowa 1985). This could explain why two fractions of eggs, IR and IIR, were noted in the ovaries. Although two fractions occurred in most ovaries, there were occurrences of ovaries with only one fraction -IIR (see results). The occurrence of only one fraction and the lower number of eggs might be explained by the fact that the prior batch comprised of the IR fraction had already been spawned (Wandzel 2000). This suggests that, after the first spawning event, the IIR fraction matures and becomes the fraction of more mature IR eggs that are spawned in a later spawning phase. By October, the ovaries were spent.

The male gonads were also divided into two morphologically distinct parts - the testis and the cement gland, as proposed by Miller (1984). The role of the cement gland is not yet fully understood (Miller 1984). The fluid it contains might improve sperm contact with eggs or it might incite females to spawn subsequent batches of eggs (Miller 1984, Charlebios et al. 1997).

The males also exhibit two clear phases in the spawning process with a pause in June (Fig. 3). The developmental cycle of the cement gland, which is similar to the ovaries, also exhibits a decline in spawning activity in June. However, throughout the study period it occurs in developmental stage 2, but also in small numbers in stage 3 (April, July) (Fig. 3). This suggests that cement gland maturity is somewhat dependent on the maturity of the testes. If the testis is spent, the cement gland is absent.

In the current study, absolute fecundity is defined as the number of eggs in the ovaries in developmental stages 2, 3, and 4. In the Gulf of Gdańsk, absolute fecundity ranged from 94 – 2190 eggs (mean 645, SD 423) at a female length of 59 - 187 mm. Wandzel (2000) reported a higher absolute fecundity range of 89 to 3824 eggs (mean 1739) at a mean female length of 74-166 mm for round gobies from the Gulf of Gdańsk during the same period. The estimated absolute fecundity is lower than that from the native round goby regions, where it ranges from 328 to 5221 eggs (Kovtun 1878). Although in the Great Lakes of North America, which have also been colonized by the round goby, fecundity ranges from 259 to 1818 eggs (MacInnis & Corcum 2000). The relative fecundity of the round gobies from Gulf of Gdańsk ranged from 9 to 143 eggs g<sup>-1</sup> of female body weight. This range was wider than that reported by Wandzel (2000).

It was confirmed that there was a power relationship between absolute fecundity and total length ( $r = 0.7206$   $p < 0.005$ ) (Fig. 5) and a significant correlation between absolute fecundity and female body weight ( $r = 0.7913$   $p < 0.005$ ) (Fig. 7). Kovtun (1978) and Wandzel (2000) both reported similar

relationships. The anticipated absolute fecundity of females 150 mm in length was 1068 eggs, while Wandzel (2000) reported this as 3036 eggs.

The analysis of the correlations of absolute fecundity with fish weight and total length for the IR and IIR oocyte fractions indicates that they are only significant for the IR egg fraction ( $r=0.777$   $p<0.005$  and  $r=0.809$   $p<0.005$ ) and that the number of eggs depends on the weight and length.

Apparently, after the first period of spawning intensity the number of eggs which mature from the less mature IIR fraction to the IR does not depend on the length or weight of the fish. After the second period of spawning intensity, the eggs have already been spawned, and the gonads are empty.

As the gonad developmental stage changes over time, so does the GSI. Although the values do not differ significantly, the male GSI increases with each subsequent gonad developmental stage (Fig. 9). The insignificant differences between average GSI values for females with gonads in stages 2 and 3 (Fig. 9) indicate that the gonads develop slowly with little weight increase. When the ovaries mature to stage 4, gonad weight increases rapidly and the GSI values are about fourfold higher, and then the differences between stage 4 and lower stages are significant (Fig. 9, Table 2).

To summarize, the population of round gobies in the Gulf of Gdańsk is dominated by males, which is in contrast to the population inhabiting the Great Lakes. The sex ratio of males to females is 5:3 and has decreased since 1996. The spawning period extends from March to August. The most advanced gonad stages were noted in April and July, which indicates that the spawning intensifies at these times. A kind of pause in spawning intensity was observed in June. Absolute fecundity ranges from 94 to 2190 eggs and is positively correlated to body length and weight. Two fractions of eggs, less and more mature, were observed in the ovaries. There is a power correlation between the number eggs in the IR fraction and total length, while that between the number of eggs in the IR fraction and body weight is linear. The number of eggs from the IIR fraction does not depend on fish size (length or weight).

Although the round goby can spawn more than twice during a reproductive season (Kovtun 1977), in the current study only two egg fractions were observed in the ovaries. Further research into the reproduction of the round goby is necessary.

The existing hypothesis regarding round goby multiple spawning suggests that after the first spawning the less mature IIR fraction developed into the IR, while the less mature IIR egg fraction developed from the connective tissue at the same time. This, however, needs to be verified as the present study confirmed only two spawning batches under the conditions in the Gulf of Gdańsk.

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