Metazoan parasite fauna of the round goby *Neogobius melanostomus* Pallas, 1811 (Perciformes: Gobiidae) collected from the Black Sea coast at Sinop, Turkey

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Abstract
Round gobies (*Neogobius melanostomus*) were collected from a small stream connected to the Black Sea coast of Sinop, Turkey and examined for metazoan parasites. A total of 122 gobies was investigated throughout a 1-year period. Long-term investigations can be useful for finding rare parasites and for determining the diversity and seasonality of parasites. Parasite species recovered were a Digenea gen. sp., *Pygidiopsis genata*, *Ascocotyle* sp., *Gyrodactylus proterorhini*, *Dichelyne minutus*, *Hysterothylacium aduncum*, *Spiroxys* sp., *Neoechinohynchus rutilii*, and *Ergasilus sieboldi*. Overall infection prevalence (%) and mean intensity values were 97.5% and 78.2 ± 23.1 parasites per infected fish, respectively. Infection prevalence and mean intensity values for each parasite species in relation to season, fish size, and sex were also determined and discussed.

Keywords: Black Sea, Gobiidae, metazoan parasites, *Neogobius melanostomus*, Pisces, Turkey

Introduction
The round goby, *Neogobius melanostomus* Pallas, 1911, is a common gobiid in near-shore areas of the Black and Caspian Seas, Seas of Azov and Marmara (Charlebois et al. 1997), and is present in coastal waters off Sinop in Turkey (Berg 1949), where this study was conducted. The parasite fauna of the round goby and the existence of parasites in the native environments of this fish species have been reported by several authors (Bykovskaya-Pavlaskaya et al. 1964; Najdenova 1974; Yusupov and Urazbaev 1980; Kvach 2002a, 2005), and in the North American Great Lakes following their translocation via freighter ballast water (Charlebois et al. 1997). The gobiid parasite fauna is more heterogeneous than the gobiid fish fauna itself (Najdenova 1974) and its species composition depends mostly on the ecology of host fish (Kvach 2005).

In the present study, the existence of metazoan parasites in relation to season, the sex, and size classe of the round goby were investigated throughout a 1-year period at the Black Sea coast of Sinop, Turkey. This investigation is also the first parasitological survey of this fish species in Turkey.
Materials and methods

Specimens of *N. melanostomus* were collected by hand net and cast net in Sırakaraağaçlar stream (42°21'24"N, 35°1'5"E) which connects with the Black Sea on the coast of Sinop, Turkey. The sampling area is described in Özer (2003), but, briefly, the stream system is characteristically slightly brackish during the late autumn and early spring months (October to March) when the water level rises and connects with the Black Sea. In summer and early autumn, however, the water level drops, the connection is broken and the stream turns to fresh water. Sırakaraağaçlar stream is brackish in the autumn and winter (1–4 psu) but becomes increasingly fresh water in spring and summer (0 psu). The mean depth of the sampling site is approximately 1.5 m and its bottom is muddy and sandy in some parts.

Sampling was carried out on a monthly basis from October 1999 to September 2000. For parasitological examinations, fish were transported alive in local water directly to Sinop Fisheries Faculty Laboratory. A total of 122 round goby were investigated. At necropsy, they were weighed, the total length measured, and their sex determined. Gills, eyes, peritoneal cavity, mesenteries, and peritoneal viscera were examined for parasites using conventional methods. Fish were examined within 48 h of capture. Routine procedures were used to collect and process parasites.

The prevalence and mean intensity levels of the parasite species were determined according to Bush et al. (1997). Mean intensity values are given ± SE. Variance-protected Tukey–Kramer analysis was performed to identify significant differences in the mean intensity values of each parasite species across season, fish size, and fish sex classes. Statistical significance was tested by using ANOVA, and if the ANOVA *P* for a group was <0.05 a Tukey–Kramer post-test, which is a multiple comparison procedure that is used when more than two-group comparison indicated significant group differences, was performed. Differences in parasite loading on male and female goby were tested using the Student’s *t* test. The significance level (*a*) was set at 0.05 for all the statistical tests. *a* is used to indicate the probability of rejecting the statistical hypothesis tested when in fact that hypothesis is true. Before conducting any statistical test, it is important to establish a value for *a* and it is customary to set *a* at 0.05. Statistical significance was accepted for probability values of *P*<0.05, which is *a* level here. Statistical tests and terms used here are in accordance with Zar (1984).

Results

The current study is the first to report parasites from round gobies captured from its natural environment on the Black Sea coast of Turkey. A total of nine parasite species was identified (Table I). If the number of parasite species in the individual host (infra community) is regarded, it is apparent that the majority of round goby hosts were infected with only two parasite species (Figure 1). The overall infection prevalence (%) and mean intensity values recorded from a total of 122 fish specimens were 97.5% and 78.2 ± 23.1 parasites per infected fish, respectively. Digenea gen. sp. was the most abundant parasite and had 84.4% infection prevalence and mean intensity values of 83.8 ± 26.3 parasites per infected fish. On the other hand, *Ergasilus sieboldi* was the least abundant parasite species (Table I). Seasonal prevalence and mean intensities of all parasite species infecting *N. melanostomus* (component community) are reported in Table II. Only Digenea gen. sp., *Pygidiopsis genata*, *Ascocotyle* sp., and *Dichelyne minutus* were in sufficient number to make any statistical comparison of their seasonal mean intensity values and statistically significant
differences were determined between some seasons \( (P<0.05) \) (Table II). When the numbers of parasite species are regarded, increases in spring and summer are evident (Figure 2). Statistically significant differences were also determined between the fish size classes (Table III): large fish had higher infection prevalence and intensities than did small fish. A similar pattern, more parasite numbers in larger fish, was also seen in the prevalence values of the number of parasite species detected in three different fish size classes (Figure 3). Both infection prevalence and mean intensity values of all individual parasite species, in general, were slightly greater in females but no statistically significant difference was determined \( (P>0.05) \) (Table IV). The numbers of parasite species in both sexes were very similar and had the same pattern (Figure 4).

**Discussion**

The round goby belongs to the second largest fish family, Gobiidae, which is distributed worldwide in both salt and freshwater habitats (Charlebois et al. 1997), and its parasite fauna differs in native environments and in places of invasion. The total of nine metazoan parasite species identified here is fewer than the numbers reported by Gayevskaya et al. (1975; 47 species), by Machkevsky et al. (1990; 16 species), by Kvach (2002a; 15 species), and by Kvach (2005; 16 species), all from the native Black Sea environment, and by

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**Table I. Infection prevalence and mean intensity levels of metazoan parasites found in Neogobius melanostomus.**

| Parasite species                             | Prevalence (%) | Mean intensity \( \pm \text{SE} \) |
|----------------------------------------------|----------------|---------------------------------|
| Digenea gen. sp. (gills)                    | 84.4           | 83.8 ± 26.3                    |
| *Pygidiopsis genata* Looss, 1907 (liver, intestine) | 40.2           | 2.4 ± 0.2                      |
| *Ascoctyle* sp. (gills, intestine)          | 14.8           | 1.9 ± 0.2                      |
| *Gyrodactylus proterorhini* Ergens, 1967 (gills, fins, skin) | 16.4           | 4.0 ± 0.7                      |
| *Dichelyne minutus* Rudolphi, 1819 (intestine) | 50.8           | 3.9 ± 0.5                      |
| *Hysterobothrium aduncum* Rudolphi, 1802 (intestine) | 11.5           | 3.1 ± 1.1                      |
| *Spirocyx* sp. (intestine)                  | 16.4           | 5.4 ± 1.3                      |
| *Neoechinorhynchus rutili* Müller, 1780 (intestine) | 7.4            | 1.4 ± 0.3                      |
| *Ergasilus sieboldi* von Nordmann, 1832 (gills) | 2.5            | 9.0 ± 7.5                      |
| Overall                                      | 97.5           | 78.2 ± 23.1                    |

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Figure 1. Infection prevalence (\%) with respect to the number of parasite species detected in the round goby throughout the investigation period.
Table II. Seasonal infection prevalence and mean intensity levels of metazoan parasites found in *Neogobius melanostomus*.

| Parasite species        | Autumn (11–23°C) (n=30) | Winter (4–6°C) (n=21) | Spring (8.6–24.8°C) (n=30) | Summer (26.4–31°C) (n=41) |
|-------------------------|--------------------------|------------------------|-----------------------------|-----------------------------|
|                         | Prevalence (%) | Mean intensity (± SE)  | Prevalence (%) | Mean intensity (± SE)  | Prevalence (%) | Mean intensity (± SE)  | Prevalence (%) | Mean intensity (± SE)  |
| Digenea gen. sp.        | 66.7          | 156.4±111.5 A          | 100             | 72.9±30.9 A          | 80             | 38.8±17.5 AB          | 90.2          | 27.3±5.6 B            |
| *Pygidiopsis genata*    | 36.7          | 3.9±1.0 A              | 57.1            | 2.3±0.3 AB           | 26.7           | 1.5±0.2 B             | 46.3          | 1.9±0.2 B             |
| *Ascocotyle* sp.        | 10            | 3.0±1.0 A              | 19              | 2.0±0.4 A           | 6.7            | 1.0±0.0*              | 22            | 1.9±0.4 A             |
| *Gyrodactylus proterorhini* | 3.3          | 1±0.3 A                | 0               | 0±0.4 A             | 57             | 3.4±0.5               | 0             | 0                    |
| *Dichelyne minutus*     | 13.3          | 1.5±0.3 A              | 9.5             | 1±0.0 A             | 60             | 2.1±0.3 A             | 87.8          | 5.3±0.7 B             |
| *Hysterothyacium aduncum* | 26.7         | 3.6±1.8 A              | 23.8            | 2.6±1.1 A           | 0              | 0                      | 0             | 0                    |
| *Spiroxys* sp.          | 0             | 0                      | 7               | 2.0±1.0 A           | 7              | 42.5                   | 4.3±0.9       |
| *Neoechinorhynchus rutili* | 0             | 0                      | 0               | 0±0.4 A             | 19.5           | 1.5±0.4               | 19.5          | 1.5±0.4               |
| *Ergasilus sieboldi*    | 6.7           | 1.5±0.5 A              | 0               | 0±0.5 A             | 0              | 2.4                    | 24*           |
| Overall                 | 90            | 119.2±83.5 A           | 100             | 75.1±31.2 A         | 100            | 105.4±47.5 A          | 100           | 33.2±5.5 A            |

Values followed by the same uppercase letter are not significantly different, analysis of variance-protected Tukey–Kramer, z=0.05. *Too few values for statistical analysis.*
Ondrackova et al. (2005; 13 species) from the Danube River, Slovakia. The present total is higher than the numbers given by Kvach (2002b; seven species) in a salty closed lagoon of the North-Western Black Sea region, Ukraine; by Kvach (2002a; five species) in the Baltic Sea, and the numbers given by Muzzall et al. (1995; seven species) and Pronin et al. (1997; seven species), both following the establishment of this fish species in US fresh waters. In addition to the differences in the numbers of parasite species of the round goby in its native and expanded ranges, species composition of these parasites was also quite different as a result of different environmental, ecological, and host factors. The nematode species *Dichelyne minutus* and a digenean *Pygidiopsis genata* metacercariae were the only parasites present here and in native habitats in the north-western part of the Black Sea (Kvach 2002a, 2005). On the other hand, *H. aduncum* was first reported by Machkevsky et al. (1990) in the Gulf of Yagorlyk and later by Kvach (2002a) in the Gulf of Gdansk, Baltic Sea. *Ergasilus sieboldii* was found in Goloviţa Lake (Radulescu and Vasiliu 1951) and in the Gulf of Gdańsk (Kvach 2002a).

The parasites of the subclass Digenea are usually found encysted, encapsulated, or occasionally as free metacercarial or juvenile forms from tissues (occasionally lumen of gut: forms from eye may be free and active) (Gibson et al. 1996). *Digenea* gen. sp. was the most abundant parasite, both in prevalence and mean intensity values, and such an unidentified metacercariae has not been reported by any of the researchers who have conducted studies on the round goby so far. Some heterophyid parasites, such as *Cryptocotyle concavum* Creplin, 1825, has been reported as the dominating parasite species from the round goby and some other gobiids such as *Gobius niger* L. 1758, *Pomatoschistus minutus* Pallas, 1769, *P. pictus* Malm, 1865, *P. microps* Krøyer, 1838, and *Gobiusculus flavescens* Fabricius, 1779 (Zander and Kesting 1998; Kvach 2002a, 2005; Zander 2005a, 2005b), but they were not found in the present study. *Dichelyne minutus* and *P. genata* had the second and third

![Figure 2. Seasonal infection prevalence (%) with respect to the number of parasite species detected in the round goby.](image-url)
highest infection prevalence rates, respectively. Kvach (2005) reported lower infection prevalence rates but higher (nearly three times for *D. minutus* and 50 times for *P. genata*) mean intensity values than in the present study for the same fish species from the NW Black Sea. *Hysterothylacium aduncum* from the NW Black Sea and *E. sieboldi* from the Gulf of Gdańsk (southern Baltic) were reported at low levels (Kvach 2002a) and similar results were also recorded in the present study. *Spiroxys* sp. has only been found in the round goby from a river and lake in Michigan, USA by Muzzall et al. (1995). As far as I am aware, *Ascocotyle* sp. and *Neoechinorhynchus rutili* have not been reported from the round goby so far and these parasites are also recorded for the first time from Turkey, along with *P. genata* and *G. proterorhini*.

Table III. Infection prevalence and mean intensity levels of metazoan parasites of *Neogobius melanostomus* according to the size classes of fish.

| Parasite species         | Prevalence (%) | Mean intensity (± SE) | Prevalence (%) | Mean intensity (± SE) |
|--------------------------|----------------|----------------------|----------------|----------------------|
|                          | ≤60  | 61–80  | ≥81   | ≤60 (n=43) | 61–80 (n=44) | ≥81 (n=35) |
| Digenea gen. sp.         | 79.1 | 88.6   | 85.7  | 22.1±4.6 A | 39.6±11.8 AB | 211.3±85.3 B |
| *Pygidiopsis genata*     | 34.9 | 29.5   | 57.1  | 2.4±0.3 A  | 2.5±0.7 A  | 2.2±0.3 A  |
| *Ascocotyle* sp.         | 9.3  | 9.1    | 28.6  | 2.8±0.9 a  | 2.3±0.6 a  | 1.6±0.2   |
| *Gyrodactylus proterorhini* | 6.9  | 29.5   | 11.4  | 3.7±0.9 A  | 3.4±0.8 A  | 6.3±1.4 A  |
| *Dichelyne minutus*      | 4.6  | 72.7   | 80    | 1.0±0.0 a  | 2.2±0.2 A  | 6.1±0.8 B  |
| *Hysterothylacium aduncum* | 18.6 | 11.4   | 2.9   | 1.5±0.2 A  | 4.8±2.8 A  | 7 a       |
| *Spiroxys* sp.           | 0    | 11.4   | 42.9  | 0            | 3.4±0.5 A  | 6.1±1.7 B  |
| *Neoechinorhynchus rutili* | 0    | 9.1    | 8.6   | 0            | 1.0±0.0 a  | 1.8±0.6 a  |
| *Ergasilus sieboldi*     | 2.3  | 2.2    | 2.9   | 1 a          | 2 a        | 24 a      |
| Overall                  | 93   | 100    | 100   | 20.6±4.0 A  | 39.7±10.8 A | 192.3±74.3 B |

Values followed by the same uppercase letter are not significantly different, analysis of variance-protected Tukey–Kramer, α=0.05. aToo few values for statistical analysis.

Figure 3. Infection prevalence (%) with respect to the number of parasite species detected in the round goby of different size classes.
Many gobies are short-lived fishes that become adult in the first year and die mostly after the spawning season (Zander 1993). *Neogobius melanostomus*, however, can live for 3 years or longer (Charlebois et al. 1997). Young fish appear in summer and grow up to juveniles, which emigrate in autumn to deeper waters; when temperatures rise in the following spring they immigrate into shallow waters (Mayskiy 1960). The gobiid larvae and young individuals live in the suprabenthos (Zander and Kesting 1998). During this period they can ingest some nematode larvae together with their copepod prey (Zander 1993). Three metacercariae species: Digenea gen. sp., *Pygidiopsis genata*, and *Ascocotyle* sp., together with a species of nematode, *Dichelyne minutus*, have been found firstly infecting the round goby in autumn and in the other seasons. Study of the prey composition of the round goby was beyond the scope of the present study, but the first occurrence of three digeneans and a nematode species within the host in autumn could be the result of above-mentioned

![Figure 4. Infection prevalence (%) with respect to the number of parasite species detected in the round goby of different sexes.](image)

Table IV. Infection prevalence and mean intensity levels of metazoan parasites found in *Neogobius melanostomus* according to the sex of host.

| Parasite species          | Prevalence (%) | Mean intensity (± SE) |
|---------------------------|----------------|----------------------|
|                           | Female (n=82)  | Male (n=40)         | Female (n=82)  | Male (n=40)         |
| Digenea gen. sp.          | 83.9           | 87.5                 | 110.0±39.4 A   | 32.9±6.3 A          |
| *Pygidiopsis genata*      | 40.2           | 40                   | 2.4±0.3 A      | 2.4±0.2 A           |
| *Ascocotyle* sp.          | 15.9           | 12.5                 | 2.2±0.4 A      | 1.6±0.2 A           |
| *Gyrodactylus proterorhini* | 17.1        | 15                   | 4.1±0.9 A      | 3.7±1.1 A           |
| *Dichelyne minutus*       | 53.7           | 45                   | 4.2±0.6 A      | 3.3±0.6 A           |
| *Hysterohylacium aduncum* | 7.3            | 20                   | 2.3±0.9 A      | 3.6±1.8 A           |
| *Spiroxys* sp.            | 19.5           | 10                   | 6.1±1.5 A      | 2.5±1.2 A           |
| *Neoechinorhynchus radii* | 8.5            | 5                    | 1.1±0.1 A      | 2.1±1.5 A           |
| *Ergasilus sieboldi*      | 2.4            | 2.5                  | 12.5±11.5      | 2                   |
| Overall                   | 96.3           | 100                  | 100.9±34.4 A   | 33.2±5.9 A          |

Values followed by the same uppercase letter are not significantly different, analysis of variance-protected Tukey–Kramer, α=0.05.
host–parasite interactions. A clear seasonality was observed in the occurrence of some parasite species such as *G. proterorhini*, *H. aduncum*, *Spiroxys* sp., *N. rutili*, and *E. sieboldi*, which appeared only in some seasons. *Gyrodactylus proterorhyni* and *E. sieboldi* can be differentiated from the rest by mode of life and infestation process. Both species had clear seasonal appearances. While *G. proterorhyni* occurred within the temperature range 11–24.8°C, especially May in spring, the predominant season for appearance, *E. sieboldi* occurred at a higher temperature range of 23–31°C, September in autumn and July in summer. Changes in water temperature are probably the most important cause of this specificity in their occurrences as was noted by Chubb (1977) for *Gyrodactylus* and other monogeneans. The occurrence of both parasites also coincides with 0–1 psu salinity in the stream system. Infection of the round goby with the nematode *H. aduncum* showed a seasonal appearance only in autumn and winter, and the same pattern of this parasite in another gobiid fish *Gobius niger* was observed by Zander and Kesting (1998). They reported this seasonal occurrence as a result of the dying off of parasites and the continuous re-infection during the spring and summer via ingestion of crustaceans that harbour parasite larvae. On the other hand, *Spiroxys* sp. found in the present study had a reverse occurrence pattern, firstly low appearance in spring and then higher infection prevalence and mean intensity values in summer. This could be a result of a longer accumulation period up to summer. This assumption is supported by the record of largest-sized round goby, coinciding with the summer season, with the greatest infection prevalence and mean intensity values. The number of parasite species and their corresponding seasonal infection prevalence increased in general in spring and summer. From late spring to early autumn, the connection of the stream with the Black Sea was broken and promoted eutrophication and, hence, the production of detritus-rich sediments, which is an advantageous environment for intermediate hosts that caused more successive parasitization, as was observed by Zander and Kesting (1998).

In general, there is a positive relationship between parasitism, age, and size. Larger, older individuals are more likely to be parasitized because they have been around longer and there is a greater probability that they will encounter parasites (Krist and Lively 1998). Here, in the present study, the number of parasite species and the specimen numbers of each parasite species, infection prevalence, and mean intensity values increased as the size of fish increased. In most of the parasite species, however, mean intensity values increased more consistently than those corresponding infection prevalence values. Similar results, in part, were also observed in some gobiid fish species by Zander and Kesting (1998).

Parasites may infect both sexes differently, because male and female fish often have different feeding habits (Rohde 1993). In our study, host sex did not have a significant influence on the parasite fauna of *N. melanostomus*, suggesting that habitat use and diet are similar for both sexes of this species. The parasite species numbers detected in both sexes of fish presented only slight differences, infection prevalence and mean intensity values for each parasite species were also very close in both sexes and this may also indicate that no difference was present in the susceptibility to parasites.

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