Ecological niche of

invasive round goby *Neogobius melanostomus* (Pallas, 1814) in the Kiel Canal and adjacent section of the Elbe River

Dissertation

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vorgelegt von

Mattias Hempel

aus Braunschweig

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- 1. Gutachter: Prof. Dr. Ralf Thiel
- 2. Gutachterin: Jun. Prof. Dr. Mathilde Cordellier

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1 Summary

The round goby *Neogobius melanostomus* (Pallas, 1814) was introduced into numerous waters in Europe and North America in the late 20th and early 21st centuries. The highly invasive species is able to build up dense populations in invaded areas within only a few years. Therefore, it can strongly influence native species by direct predation or competition and alter the invaded ecosystems.

This thesis focuses on the role of *N. melanostomus* in brackish and freshwater habitats in Northern Germany, especially in the federal states Schleswig-Holstein and Hamburg. The considered habitats have recently been invaded by the species and hold established populations nowadays.

Research into trophic interactions between round goby and its predators and prey organisms can contribute to the knowledge of invasive species and lead to a better understanding of processes in the invaded ecosystems. First, a recent range expansion of *N. melanostomus* has been shown by reporting the first records of the species in the Elbe River, Germany. Second, with regard to a possible further range expansion into regions with higher salinities, an aquaria experiment was conducted measuring the salinity tolerance of juvenile specimens. Third, the role of round goby in the Kiel Canal was studied in detail with emphasis on its feeding ecology, growth and reproductive biology. This information gives important insights into the adaptability of the species to artificial brackish water habitats. Fourth, the growth and diet composition of zander *Sander lucioperca* (Linnaeus, 1758) as a hypothesized predator of *N. melanostomus* was studied to exemplify potential effects of the newly available prey on higher trophic levels.

In Chapter 4, the first records of round goby in the Elbe River, Germany were stated. The described 36 specimens were found from May 2008 to June 2013 mainly in the tidal river section close to the city of Hamburg.

In Chapter 5, the results of an aquaria experiment dealing with the effects of salinity on juvenile round goby from the brackish Kiel Canal were shown. In detail, survival, daily food intake and growth were compared between salinities of 0.1, 7.5, 15 and 30 over 12 weeks. All specimens were fed *ad libitum* with mysid shrimps and measured and weighted weekly. Mean daily food intake increased from salinity level 0.1 to 15. Specimens at level 30 showed a more diversified and lower mean food intake. Mean cumulative body weight gain appeared greatest at a level of 7.5, closely followed by 0.1 and 15. In contrast, fish growth was significantly lower at a salinity level of 30. The results of the experiment indicate that the assumed salinity tolerance of round goby made by other authors is too low at least regarding specimens from brackish habitats. A further spread of this species into regions with higher salinities seems to be possible.

The results of research into diet, growth and reproductive biology of *Neogobius melanostomus* from the Kiel Canal, northern Germany, were shown in Chapter 6. Crustaceans, annelids and chironomids had the highest importance in the diet of all three inspected size classes of round goby. Only large specimens consumed mussels and fish, which contributed to the diet to a minor extent. Diet showed a high interannual variability. In comparison to other ecosystems, round goby in the Kiel Canal has a relatively low asymptotic total length and a high relative fecundity. The inspected population has an almost balanced sex ratio and the highest spawning activity takes place in May and June. Direct predation on eggs and fry of native fish species appear to be too low to have negative effects. Due to the high abundance of round goby in the Kiel Canal it seems likely that changes will occur in the ecosystem due to competition with native fish species and predation on invertebrates.

The diet and growth of zander from the Kiel Canal as a possible predator of round goby was studied in Chapter 7 to analyse effects of the newly available potential prey. Round goby represented the most important fish prey taxon in the zander diet in 2011–2013. Inspected zander of total lengths \geq 200 mm and \geq 400 mm fed heavily on round goby, at least in the warmer quarters (2nd and 3rd) of the years under study. In the colder quarters of the inspected years (1st and 4th), zander \geq 400 mm showed a high amount of cannibalism. In comparison with data from 1995/1996 - before the round goby invasion in 2006 - zander showed considerable differences in diet composition. They also showed faster growth and a better condition in this study compared to the data from 1995/1996. As catch numbers of round goby dropped while zander catches increased, abundance of round goby seems to be controlled by high predator abundance.

This thesis contributes significantly to an improved state of knowledge about the invasive round goby, particularly in artificial brackish water habitats. It provides new insights into the spread of the round goby in Northern Germany and hints for a possible further development. A reassessment of the role of *Neogobius melanostomus* in the food web of the Kiel Canal should be conducted after a couple of years in order to estimate the changes of the ecosystem due to the invader.

2 Zusammenfassung

Die Schwarzmundgrundel *Neogobius melanostomus* (Pallas, 1814) gelangte im späten 20. beziehungsweise im frühen 21. Jahrhundert in zahlreiche Gewässer Europas und Nordamerikas, in welchen die Art zuvor nicht heimisch war. Die stark invasive Art bildet in neu kolonisierten Bereichen häufig bereits nach wenigen Jahren abundante Populationen aus. Daher kann sie, sowohl durch direkten Fraßdruck als auch durch Konkurrenz, einen starken Einfluss auf Ökosysteme und deren Artenzusammensetzung haben.

Die vorliegende Arbeit befasst sich mit der Rolle von *N. melanostomus* in Brack- und Süßwasserhabitaten in Norddeutschland, insbesondere in den Bundesländern Schleswig-Holstein und Hamburg. In den betrachteten Gebieten trat die Art erstmals vor etwa einem Jahrzehnt auf. Mittlerweile haben sich dort etablierte Populationen ausgebildet.

Die Erforschung von trophischen Interaktionen der Schwarzmundgrundel mit Prädatoren und Beutetieren kann wichtiges Grundwissen liefern und zu einem besseren Verständnis der in den kolonisierten Ökosystemen ablaufenden Prozesse führen. Zunächst wurde mit dem Erstnachweis der Schwarzmundgrundel für die Elbe in Deutschland ein Beitrag zur Ausbreitungsgeschichte geleistet. Darauf folgend wurde ein Aquarienexperiment durchgeführt, welches die Salinitätstoleranz juveniler Individuen untersuchte, um Rückschlüsse auf eine mögliche weitere Ausbreitung ziehen zu können. Um außerdem Anpassungen Schwarzmundgrundel künstliche regionale der an Brackwasserlebensräume zu untersuchen, wurden im Nord-Ostsee-Kanal detailliert die Nahrungsökologie, das Wachstum und die Reproduktion betrachtet. Ebenfalls im Nord-Ostsee-Kanal wurden Wachstum und Nahrungszusammensetzung des Zanders Sander lucioperca (Linnaeus, 1758) untersucht. Dadurch konnten mögliche Auswirkungen der potentiellen neuen Nahrungsquelle Schwarzmundgrundel auf höhere Trophieebenen studiert werden.

Mit Kapitel 4 wurden die ersten Nachweise der Schwarzmundgrundel für die Elbe in Deutschland veröffentlicht. Die beschriebenen 36 Individuen wurden zwischen Mai 2008 und Juni 2013, vor allem aus dem tidebeeinflussten Bereich des Flusses nahe der Stadt Hamburg, gemeldet.

Kapitel 5 beschreibt die Ergebnisse eines Aquarienexperiments zum Einfluss der Salinität auf juvenile Schwarzmundgrundeln aus dem brackwassergeprägten Nord-Ostsee-Kanal. Dazu wurden die Überlebensrate, die tägliche Futteraufnahme und das Wachstum bei verschiedenen Salzgehalten (0,1; 7,5; 15; 30) über die Dauer von 12 Wochen verglichen. Alle Individuen wurden *ad libitum* mit Schwebgarnelen gefüttert und wöchentlich gemessen und gewogen. Die tägliche Futteraufnahme stieg vom Salzgehalt 0,1 bis zum Salzgehalt von 15 an. Bei einem Salzgehalt von 30 wurde eine durchschnittlich geringere Futteraufnahme mit großen Schwankungen zwischen den einzelnen Individuen festgestellt. Die kumulative Gewichtszunahme war beim Salzgehalt 7,5 am höchsten, dicht gefolgt von der Zunahme bei den Salzgehalten 0,1 und 15. Das Wachstum beim Salzgehalt 30 dagegen war signifikant geringer. Das Experiment lässt den Schluss zu, dass die bisher von einigen Autoren für die Schwarzmundgrundel angenommene Salzgehaltstoleranz zu niedrig angesetzt ist, wenn Exemplare aus Brackwassergebieten betrachtet werden. Eine weitere Ausbreitung in Gebiete mit höheren Salzgehalten erscheint durchaus möglich.

Die Ergebnisse der Forschungsarbeit zu Nahrung, Wachstum und Reproduktionsbiologie von *Neogobius melanostomus* aus dem Nord-Ostsee-Kanal in Norddeutschland sind in Kapitel 6 dargestellt. Crustacea, Annelida und Chironomidae hatten die höchste Bedeutung in der Nahrung der drei untersuchten Größenklassen von Schwarzmundgrundeln. Nur von großen Individuen wurden auch in geringen Anteilen Muscheln und Fische konsumiert. In der konsumierten Nahrung zeigten sich große interannuelle Schwankungen. Im Vergleich zu anderen Ökosystemen zeigten die untersuchten Schwarzmundgrundeln aus dem Nord-Ostsee-Kanal eine relativ geringe asymptotische Totallänge bei einer hohen relativen Fruchtbarkeit. Die Population hatte ein annähernd ausgeglichenes Geschlechterverhältnis, und die stärkste Laichaktivität wurde im Mai und Juni verzeichnet. Der direkte Prädationsdruck auf Eier und Larven heimischer Fischarten wurde aufgrund der gewonnenen Daten als relativ gering eingeschätzt. Durch die hohe Abundanz der Schwarzmundgrundel im Nord-Ostsee-Kanal erscheint es jedoch wahrscheinlich, dass es durch Konkurrenz mit heimischen Fischarten und/oder Fraßdruck auf Invertebraten zu Veränderungen im Ökosystem kommt.

Um Effekte der neuen Nahrungsquelle Schwarzmundgrundel auf einen möglichen Prädator zu untersuchen, wurden in Kapitel 7 Nahrung und Wachstum des Zanders im Nord-Ostsee-Kanal betrachtet. Die Schwarzmundgrundel hatte die höchste Bedeutung in der Nahrung der Zander im Untersuchungszeitraum 2011 bis 2013. Zander der Größenklassen ≥200 mm und ≥400 mm Totallänge konsumierten, zumindest in den warmen 2. und 3. Jahresquartalen, zu großen Teilen Schwarzmundgrundeln. In den kalten 1. und 4. Quartalen war bei Zandern ≥400 mm vermehrt Kannibalismus festzustellen. Im Vergleich mit Daten aus den Jahren 1995 und 1996 - aus der Zeit vor dem Auftreten der Schwarzmundgrundel in 2006 - wurden starke Unterschiede in der Nahrungszusammensetzung des Zanders festgestellt. Außerdem zeigten die Zander in der aktuellen Studie ein schnelleres Wachstum und eine bessere Kondition als früher. Da fallende Fangzahlen bei der Schwarzmundgrundel und steigende Fangzahlen beim Zander festgestellt werden konnten, wird eine Abundanzkontrolle durch den Prädator vermutet.

Die vorliegende Arbeit leistet einen wichtigen Beitrag zu der Erweiterung des Kenntnisstands über die invasive Schwarzmundgrundel, insbesondere in künstlichen Brackwasserlebensräumen. Sie liefert

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neue Erkenntnisse zur Ausbreitung der Grundel in Norddeutschland und gibt Hinweise auf die mögliche weitere Entwicklung der Situation. Um einschätzen zu können, inwieweit Änderungen im Ökosystem des Nord-Ostsee-Kanals zu erwarten sind, sollte dort die Rolle von *Neogobius melanostomus* im Nahrungsnetz nach einigen Jahren nochmals untersucht werden.

3 General introduction

3.1 Neobiota and invasive species in aquatic ecosystems

By definition, neobiota are taxa establishing in the wild beyond their natural distribution ranges due to humans or human-related activities after the year 1492. Newly established animals are named neozoans and plants neophytes. If the species have a relevant impact on the ecosystem they are introduced to, they are called invasive species (e.g. VITOUSEK ET AL. 1997, GEITER ET AL. 2002, HELENO ET AL. 2009). This is applicable to numerous examples regarding species of plants as well as animals. Caused by the increase in international trade intensities, the number of introductions of invasive species rose quickly in recent decades (e.g. MARGOLIS & SHOGREN 2004, WESTPHAL ET AL. 2008).

Currently, a massive biotic homogenization of the earth's surface can be seen as the effect of the breakdown of major biogeographic barriers that have historically kept the floras and faunas of the various continents quite distinctive (MILLS ET AL. 2000). Biotic change, as much as climate change, has the capacity to alter the whole nature of ecosystems. Thus, it can result in large ecological and economic consequences and is subject to international concern, embedded e.g. in the Convention on Biological Diversity (MILLS ET AL. 2000). Invasive species are widely accepted as one of the major causes of species extinction and biodiversity loss in the world (e.g. LOWE ET AL. 2000, ALLENDORF & LUNDQUIST 2003, DIDHAM ET AL. 2005, GOLLASCH ET AL. 2006).

For aquatic animals, possibilities of reaching new habitats were strongly promoted by human activities, most notably the development of new connections between water bodies by canalization and shipping traffic. Canalization can eliminate natural barriers for dispersal of aquatic organisms like watersheds between rivers or land bridges between seas (GALILETAL. 2007). For example, the Kiel Canal in northern Germany, with the highest number of ship transits worldwide, was built to shorten the voyage between the Baltic and the North Seas by up to 450 NM. This canal was mentioned to represent an important migration pathway and 34 non-native species (plants and animals) are known to occur in it or in adjacent waters (GOLLASCH ET AL. 2006). If a new connection is build, both natural dispersal of mobile species and ship-induced dispersal can take place. As the first navigable canal was built around 2200 BC linking the Tigris and Euphrates rivers in Mesopotamia (GALIL ET AL. 2007), support of species invasions in the mentioned way has taken place for a long time. Due to proceeding globalization and the related boost in shipping traffic world-wide, there are numerous examples for ship-induced dispersal since the 20th century. Large transoceanic vessels, carrying high amounts of ballast water, are able to release a lot of eggs, larvae or small sized adult animals while exchanging ballast water in harbours, representing a major vector of introduction (ELLIS & MACISAAC 2009).

Many invasive aquatic species have broad abilities in tolerating different salinity levels (e.g. PAAVOLA ET AL. 2005, ELLIS & MACISAAC 2009). They can often handle short-term salinity changes, which occur during ballast water exchange, easily and survive in conditions different from those in their native habitats. On the one hand, a lot of invasive species are well adapted to intermediate salinities. On the other hand, these intermediate salinities in brackish water seas (e.g. Baltic Sea) hold the lowest native species richness and the highest amount of "empty" niches (PAAVOLA ET AL. 2005). Thus, non-indigenous species are often able to develop high species richness and high abundance when colonizing brackish waters.

The Ponto-Caspian region serves as a major source for successful invaders of the aquatic world (see RICCIARDI & RASMUSSEN 1998), especially for waters in Europe. Canalisation and the subsequent shipping traffic have connected rivers in the west with eastern ones, serving as "invasion-highways" (GALIL ET AL. 2007). Additionally, the Ponto-Caspian region has a history of fluctuating water levels and salinities and therefore holds a large number of species that are able to tolerate various salinities (RICCIARDI & MACISAAC 2000).

Examples for successful invasive species with Ponto-Caspian origin come from several classes of animals. The zebra mussel *Dreissena polymorpha* (Pallas, 1771), native to basins of rivers draining into the Black Sea and the Sea of Azov and in estuarine reservoirs and other freshwater influenced parts of these seas (Son 2007), represents an important example of invasive bivalves. In Europe, multiple introductions of zebra mussel have taken place since the beginning of the 19th century (KARATAYEV ET AL. 1997). In North America the invasion of dreissenids was first detected in the western basin of Lake Erie, Canada, in 1986 (CARLTON 2008). One species of the genus *Dreissena*, the quagga mussel *Dreissena bugensis* (Andrusov, 1897), was even back-introduced from North America to Europe (BOLTOVSKOY 2015).

Malacostraca, in particular gammarid amphipods, is one of the macroinvertebrate groups with the highest number of invasive species in the world (BACELA-SPYCHALSKA & VAN DER VELDE 2013). One of them, the killer shrimp *Dikerogammarus villosus* (Sowinsky, 1894), which was native to the Ponto-Caspian basin, has spread into many waters in Europe.

Within the class Actinopterygii (ray-finned fishes), the family Gobiidae comprises some successful invaders of the late 20th and the early 21st century that stem from the Ponto-Caspian basin. The species round goby *Neogobius melanostomus* (Pallas, 1814), monkey goby *Neogobius fluviatilis* (Pallas, 1814), racer goby *Babka gymnotrachelus* (Kessler, 1857), bighead goby *Ponticola kessleri* (Günther, 1861) and western tubenose goby *Proterorhinus semilunaris* (Heckel, 1837) have extended their distribution areas in Europe and beyond (e.g. GRABOWSKA & GRABOWSKI 2005, RIZEVSKY ET AL. 2007, GRABOWSKA ET AL.

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2009, NEILSON & STEPIEN 2009, CAMMAERTS ET AL. 2012, BORCHERDING ET AL. 2013). Due to high tolerance of different environmental variables, e.g. in salinity, temperature and oxygen content of the inhabited waters, the gobiids are very successful invaders (see LFV BAYERN 2013). While some neozoan fish species only appear for a short period of time and are not able to reproduce in their new environment, a minor number of species have established populations as a permanent component of the invaded ecosystems.

In Germany, 63 fish species were reported as neozoans in 2000, eight of these as established (GEITER ET AL. 2002). In 2010, 15 non-native fish species already had developed established populations in Germany (WIESNER ET AL. 2010). Of these established species, only gobiids are actually spreading throughout the waterways, while stocks of other non-indigenous fishes tend to decline or are at most stable (WOLTER & RÖHR 2010).

Northern Germany represents a region that is inhabited by many established invasive species, especially by aquatic taxa. In the Elbe River various non-indigenous invertebrate species occur. Considering the whole extent of the river, NEHRING (2006) described 31 alien macrozoobenthic species, 21 of them occurring in the estuary in northern Germany (near to the study site of Chapter 4). The Kiel Canal (study site of Chapters 6 and 7) has not only been colonised by numerous invasive species (e.g. BULNHEIM 1980, NEHRING 2000, NEUKAMM 2009), it also acts as a potential way of introduction by connecting the North Sea with the Baltic Sea (NEHRING 2003, GOLLASCH ET AL. 2006). In the studied areas, only one species of invasive gobiids has been found in high numbers until now, the round goby. Its distribution nowadays and the supposed invasion history are described in the following in detail.

3.2 Characteristics, biological aspects and distribution of round goby

The species investigated in detail in this dissertation, the round goby, is a relatively large benthic gobiid species reaching a maximum total length of about 250 mm (SAPOTA 2006). It is mostly described in current literature with terms like invasive, non-indigenous or introduced. Regardless of ongoing discussion on terminology in invasive species (e.g. COLAUTTI & MACISAAC 2004), these terms are used synonymously in this thesis.

The round goby has an elongated body, circular in cross section, a terminal mouth with thick lips and a slightly notched tongue (KORNIS ET AL. 2012). The posterior angles of its jaws extend to the anterior quarter of the eye. Coloration of the species is variable, with brown, yellow and/or grey pigmentation, mostly associated with darker brown lateral blotches. Breeding males have a dark, nearly black, coloration with white-edged fins (MILLER 1986), while females possess a lighter coloration throughout the entire season. *N. melanostomus* has two dorsal fins separated by a small interspace. First dorsalfin usually has a large black spot in posterior part (MILLER 1986, Figure 3.1). Sometimes specimens without this spot occur (ISSG 2013). Pelvic fins are fused, forming a suction disc almost reaching the anus. Two pairs of pharyngeal tooth plates are present near the first two gill arches (KORNIS ET AL. 2012). Round goby lacks a gas bladder and the body is scaled. Meristic counts of the major characters (Table 3.1) were given for the native range of round goby by MILLER (1986), by N. Delventhal & J. Lyons (pers. comm. cited in KORNIS ET AL. (2012)) for the introduced range, Lake Michigan and by KNEBELSBERGER & THIEL (2014) for the introduced range, western Baltic Sea area.



Figure 3.1. Habitus of round goby Neogobius melanostomus.

Table 3.1. Meristic characters of round goby given for the native range by MILLER (1986), for Lake Michigan by N. Delventhal & J. Lyons (pers. comm. cited in KORNIS ET AL. (2012)) and for the western Baltic Sea area by KNEBELSBERGER & THIEL (2014), n.i. = character not indicated.

Meristic character	native range	Lake Michigan	western N Baltic Sea	
Number of specimens analysed	unknown	48	7	
First dorsal-fin spines (D1S)	5–7	5–6	6	
Second dorsal-fin rays (D2R)	13–16	15–17	13–16	
Anal-fin rays (AR)	11–14	11–15	11–13	
Pectoral-fin rays (PR)	17–20	n.i.	18	
Total vertebrae (Ve)	31–34	31–34	31–34	
Lateral line scales (LL)	45–57	42–59	n.i.	

Differentiation of male and female specimens of the species based on external morphology is possible with some practice at a total length of about 60 mm or more (M. Hempel pers. obs.). The most important external character for differentiation is the shape of the erectile urogenital papilla between the anus and the base of the anal fin. It is broad and blunt in females and longer and pointed in males (see KORNIS ET AL. 2012). Males mature at an age of 3–4 and females at an age of 2–3 years in the native distribution area (MILLER 1986). Younger ages at maturity were found in invaded areas in North America and northern Europe (e.g. MACINNIS & CORKUM 2000, TÜRK & SCHNELL 2015), indicating that the species can even mature after one year of growth. Round goby is a multiple spawner which can repeat spawning every 18–20 days at water temperatures of 9–26 °C (CHARLEBOIS ET AL. 1997) for up to six times a year (MILLER 1986). Males guard nests, usually in cavities, with up to 10,000 eggs from four to

six females (CHARLEBOIS ET AL. 1997). Males do not feed while nest guarding and die after the reproductive season (MILLER 1986). The sex ratios of round goby are reported to differ between native and introduced populations. In introduced populations there are typically at least twice as much males as females (KORNIS ET AL. 2012). In contrast, native populations in the Sea of Azov are reported to have 1.6–1.9 females per male, sampled 1961 to 1975 (KOVTUN 1979). The used sampling gear seems to have a strong effect on the sex ratio found with higher proportions of males when using passive sampling gear or while angling (e.g. BRANDNER ET AL. 2013c).

N. melanostomus is native to the Ponto-Caspian region. There it naturally inhabits the Black Sea, Sea of Azov, Caspian Sea and the tributaries of those waters (MILLER 1986). The species prefers areas with hard substratum due to better possibilities of spawning, hiding and feeding (e.g. BELANGER & CORKUM 2003, SAPOTA 2006, KORNIS ET AL. 2012). When stony nesting areas are limited, any solid elements on the bottom may be utilized as nest substrate (e.g. SAPOTA ET AL. 2014). Round goby typically occurs in shallower inshore areas with depths of up to 20 m (MILLER 1986) but, especially in winter, it can be found in deeper areas, in up to 60 or 130 m (WALSH ET AL. 2007), depending on the inhabited water body.

The diet of *N. melanostomus* chiefly consists of bivalves, crustaceans and polychaetes, but small fish and chironomid larvae are also found (MILLER 1986). The species has an opportunistic feeding strategy (e.g. KIPP & RICCIARDI 2012, BORCHERDING ET AL. 2013, BRANDNER ET AL. 2013a) and a strong adaptability to manmade changes in prey availability (KUDRENKO & KVACH 2005). An ontogenetic diet shift from an amphipod-dominated to a mollusc-based diet was described by numerous authors (e.g. BORCHERDING ET AL. 2013, BRANDNER ET AL. 2013, BRANDNER ET AL. 2013, BRANDNER ET AL. 2013a). Although bivalves (mostly *Dreissena* spp.) are, caused by long retention time of shells in the digestive tract, often overestimated in the diet of round goby (BRUSH ET AL. 2012), it is a major prey source, especially for larger specimens, in many regions (e.g. JUDE ET AL. 1992, RAY & CORKUM 1997, THOMAS 1997). BRANDNER (2014) confirmed high plasticity of round goby diet in comparison with the bighead goby.

The ability to tolerate multiple water conditions makes *N. melanostomus* very successful in establishing new populations. The species is able to adapt to a wide range of salinities, inhabiting fresh, brackish and marine waters (SkóRA ET AL. 1999). In pure freshwater ecosystems (e.g. Rhine River and Danube River, Europe, and Great Lakes, North America), as well as in brackish waters (Baltic Sea), it has developed large populations shortly after introduction. In contrast, it was supposed that full oceanic salinity was not accessible for round goby as a permanent habitat (e.g. ELLIS & MACISAAC 2009, KARSIOTIS ET AL. 2012). *N. melanostomus* is also able to tolerate a wide range of temperatures. Water temperature of many inhabited ecosystems can drop to near 0°C in winter. The upper limit for positive growth of the round goby was estimated to be 28.9°C, the energetic optimum temperature 26°C (LEE & JOHNSON

2005). Tolerance of very low water oxygen concentrations is another factor, which can support the spreading of *N. melanostomus*. The oxygen threshold level is variable, depending on body weight, from 0.3 to 0.9 mg l⁻¹ (BALÁŽOVÁ-L'AVRINČÍKOVÁ & KOVÁČ 2007). Lifespan of round goby is described to be up to four years in its native area (MILLER 1986). In the invaded southern Baltic Sea, an age of up to six years was reported by SOKOŁOWSKA & FEY (2011) and some inspected specimens from Lake Michigan, North America, reached an age of seven years (HUO ET AL. 2014).

N. melanostomus was introduced into numerous waters in North America and Europe (Figure 3.2) in the late 20th and the early 21st century. In North America, specimens of round goby were first detected in 1990 by anglers in the St. Clair River, connecting Lake Erie and Lake Huron, two of the Laurentian Great Lakes (JUDE ET AL. 1992). Introduction to the Great Lakes was supposed to have taken place via ballast water from transoceanic vessels (e.g. KORNIS ET AL. 2012) originating from the southern Dnieper River, a Black Sea tributary (BROWN & STEPIEN 2009). Since its first detection in 1990 round goby has spread throughout the Great Lakes at a faster rate than any previous fish invader (KORNIS ET AL. 2012). In Europe, it has spread and is currently still spreading in many rivers (KALCHHAUSER ET AL. 2013) and in the Baltic Sea area (e.g. SAPOTA & SKÓRA 2005, KORNIS ET AL. 2012).



Figure 3.2. Native round goby distribution area after MILLER (1986) (green) and invasive range extensions (red). Redrawn after KORNIS ET AL. (2012) and BRANDNER (2014) with additional records after BRUNKEN ET AL. (2012), MICHALEK ET AL. (2012), MANNÉ ET AL. (2013), LSFV NIEDERSACHSEN (2014), the U.S. GEOLOGICAL SURVEY USGS NONINDIGENOUS AQUATIC SPECIES DATABASE (2014), ROCHE ET AL. (2015) and BERLIN.DE (2017). The rectangle shows the position of detailed map (Figure 3.3).

Since the supposed first introduction into the Baltic Sea, detected in 1990 in the Bay of Gdańsk, Poland (SAPOTA & SKÓRA 2005), round goby has spread both west- and eastwards along the Baltic coasts (Figure

3.3). Westwards it was detected in the estuary of Odra River, border of Poland and Germany, in 1996 (Anonymous, cited in CZUGAŁA & WOŹNICZKA 2010), near Rugia Island in 1998 (WINKLER 2006) and in the Trave River, Germany, in 2008 (pers. comm. Dr. M. Brunke, State Agency for Agriculture, Environment and Rural Areas, Schleswig-Holstein, Germany). In 2006, round goby probably appeared in the Kiel Canal, northern Germany, for the first time (NEUKAMM 2009). Westwards of the study area, round goby was found in the tidal part of the Weser River, Germany, in 2012 (BRUNKEN ET AL. 2012).



Figure 3.3. Map of round goby distribution in northern Germany and nearby areas of the Baltic Sea. Red numbers refer to the years of first occurrence described in literature. Weser River: BRUNKEN ET AL. (2012), Kiel Canal: NEUKAMM (2009), Trave River: pers. comm. Dr. M. Brunke, State Agency for Agriculture, Environment and Rural Areas, Schleswig-Holstein, Germany, Denmark: AZOUR (2011), German Baltic Sea coast: K. Skóra, University of Gdańsk, unpublished data cited in CORKUM ET AL. (2004), Rugia Island: WINKLER (2006), Odra River estuary: Anonymous, cited in CZUGAŁA & WOŹNICZKA (2010), Bornholm: MICHALEK ET AL. (2012), southern coast of Sweden: BJÖRKLUND & ALMQVIST (2010), Bay of Gdańsk: SAPOTA & Skóra (2005), coastal waters of Lithuania and Latvia: RAKAUSKAS ET AL. (2008). Red arrows show the supposed directions of range extensions from the Bay of Gdańsk.

3.3 Effects of round goby and other invasive animals on aquatic food webs

Invasive gobiids, especially when occurring in high abundance, can influence food web structures by changing composition and abundance of benthic invertebrates (e.g. BARTON ET AL. 2005, LEDERER ET AL. 2008, KIPP & RICCIARDI 2012) and by competition with native species for food and/or habitat (e.g. KARLSON ET AL. 2007, VAN KESSEL ET AL. 2011). Introduction of fish species, wanted as well as undesired, has often resulted in important changes of ecosystem communities. The Nile perch *Lates niloticus* (Linnaeus, 1758) is probably the most cited example for invasive fish species. Native in several rivers and lakes of the African continent, it was introduced into Lake Victoria, Africa, in 1954 to counteract

overfishing effects. Occurrence of Nile perch in the lake led to the extinction of more than 200 endemic fish species through predation and competition for food (LOWE ET AL. 2000).

Strong impacts of introduced species on food webs in aquatic ecosystems have also been described for different invertebrate taxa, e.g. Bivalvia and Crustacea. After the occurrence of the invasive Bivalvia *Dreissena* spp. (zebra and quagga mussel) in Lake Ontario, North America, a strong impact on the plankton composition, according to an increase in water clarity, and a decline in native clams was found (MILLS ET AL. 2003). The killer shrimp *D. villosus*, one of the most studied species of invasive Crustacea, is a predatory feeder that can eliminate both native and exotic species (DICK & PLATVOET 2000). In the Rhine River, *D. villosus* is very common and has a huge impact on the food web (VAN RIEL ET AL. 2006). Another well-known example of invasive Crustacea is the Chinese mitten crab *Eriocheir sinensis* H. Milne Edwards, 1853, native in central Asia (RUDNICK ET AL. 2003). It has spread to Europe and to North America (e.g. HERBORG ET AL. 2003). Chinese mitten crab is supposed to compete with native crayfish species in freshwater (CLARK ET AL. 1998). In the ecosystems of the Baltic Sea a recent increase in Chinese mitten crab abundance poses an additional risk of structural and functional changes, especially for benthic communities (OJAVEER ET AL. 2007).

As the understanding of ecosystems and the stability, structures and functions therein, strongly depends on the understanding of the food web structure (see BECKERMAN ET AL. 2006), research on this structure is very helpful to quantify the impact of invasive gobiids on invaded ecosystems. Diet of these gobiids, often comprises large amounts of other non-indigenous species. BORCHERDING ET AL. (2013) found high percentages (56%) of Crustacea (mainly invasive *D. villosus*) in the diet of three invasive gobiid species in the Lower Rhine, Germany. A similar situation was described for the prey consumption of round goby and bighead goby in the upper Danube River (BRANDNER ET AL. 2013a), with up to two thirds of relative importance index contributed by amphipods (again mainly *D. villosus*) and about 92% of gut contents consisting of non-native prey species (BRANDNER 2014). Accordingly, the gobiid fishes seem to benefit from previous invasions of exotic prey species.

For the Great Lakes region, numerous authors (e.g. RAY & CORKUM 1997, FRENCH & JUDE 2001) reported high amounts of zebra and quagga mussels (up to 58%) in the guts of round goby. KORNIS ET AL. (2012) summarize in their review paper that, considering all known studies, by mass 64.5% of round goby diet in Eurasia consists of molluscs. Although this high amount might be an overestimation, caused by long retention time of the shells in the guts (see BRUSH ET AL. 2012), *Dreissena* spp. and other molluscs definitely represent important prey taxa. *N. melanostomus* is the only common forage fish in the Great Lakes and in the Baltic Sea area that feeds heavily on mussels (KORNIS ET AL. 2012). Thus, it facilitates the transfer of energy from mussels to higher tropic levels in both systems. It was supposed that this could support the accumulation of toxins in species of higher trophic level (AZIM ET AL. 2011). Apart from that, predatory species can benefit from the establishment of round goby in their inhabited ecosystems. High reliance on the newly available prey was observed in birds, snakes and fishes (reviewed in KORNIS ET AL. 2012). Concerning birds, examples include the grey heron *Ardea cinerea* Linnaeus, 1758 at the Baltic coast (JAKUBAS 2004) and different cormorant species in North America (double-crested cormorant *Phalacrocorax auritus* (Lesson, 1831) (SOMERS ET AL. 2003, JOHNSON ET AL. 2010, VAN GUILDER & SEEFELT 2013)) and at the Baltic coast (great cormorant *Phalacrocorax carbo* (Linnaeus, 1758) (BZOMA 1998)). Round goby can constitute up to 95% of these birds diet. Range expansions of double-crested cormorants are supposed to be associated with massive occurrence of round goby (TUCKER & SEEFELT 2014). Similarly, for inspected colonies of grey heron, the increasing number of nests is supposed to be associated with huge numbers of *N. melanostomus* nearby (JAKUBAS 2004). Island populations of northern water snake *Nerodia sipedon* (Linnaeus, 1758) from Lake Erie exhibited a massive diet shift to high amounts of round goby after invasion of the lake by this fish species (KING ET AL. 2008). The snake profits from the newly available prey, as recognizable by increased size of females and higher offspring number.

Furthermore, round goby has become important food to many predatory fishes in the newly established habitats (KORNIS ET AL. 2012). In the region of the Great Lakes and in Europe, e.g. in the Dyje River, Czech Republic, burbot Lota lota (Linnaeus, 1758) consumes large quantities of round goby (HENSLER ET AL. 2008, MIKL ET AL. 2017). The increased mean length at age for burbot seems to be related to the high reliance on round goby as food source (e.g. JOHNSON ET AL. 2005). Additionally, REYJOL ET AL. (2010) recorded round goby as prey item in 65% of stomachs of inspected sauger Sander canadensis (Griffith & Smith, 1834) in Lake St. Pierre, St. Lawrence River, Canada. For young-of-the-year smallmouth bass Micropterus dolomieu Lacepède, 1802 in Lake Erie, STEINHART ET AL. (2004b) found that 75% of consumed fish was N. melanostomus. JOHNSON ET AL. (2005) found similar percentages for dry weight of round goby in the prey of adult smallmouth bass. In the Baltic Sea area, ALMQVIST ET AL. (2010) identified Atlantic cod Gadus morhua Linnaeus, 1758 (22.4% of prey mass overall) and European perch Perca fluviatilis Linnaeus, 1758 (69% of prey mass overall) as major predators of round goby. Positive effects on the predatory fishes, with high reliance on round goby as food source, were specified by numerous authors (e.g. JOHNSON ET AL. 2005, CRANE ET AL. 2015). These effects comprise higher total length and total weight at age and a better condition of the predators. However, a N. melanostomus dominated diet does not in any case lead to better condition of the predatory species. Effects can differ strongly between observed species as well as between specimens of the same species in different ecosystems (CRANE ET AL. 2015).

3.4 Objectives

The invasive round goby is currently spreading in many regions of Europe and North America. One of these regions is northern Germany, including the study sites of Chapters 4, 6 and 7. Additionally, the fish used in the aquaria experiments (Chapter 5) are from the same area.

As the result of the short period since their first occurrence, populations of round goby in that area are nowadays subject of fast changes. Research activities in northern Germany can lead to new findings on spreading of round goby and related effects. This newly established populations of *N. melanostomus* were investigated in the artificial Kiel Canal and the river systems of Trave and Elbe in order to estimate possible impacts on the ecosystem communities.

The tidal Elbe River is connected to the systems of Kiel Canal and Trave River (Figure 3.3), both heavily colonised by round goby recently. Therefore, occurrence of round goby there seems to be only a matter of time. The Kiel Canal and the Trave River, situated north-easterly of the Elbe River, were obviously invaded from the Baltic Sea coast (e.g. NEUKAMM 2009, reports of occurrence from the Trave River system - pers. comm. Dr. M. Brunke, State Agency for Agriculture, Environment and Rural Areas, Schleswig-Holstein, Germany). About 100 km south-western of the Elbe River, in the North Sea area, first records of round goby were obtained from the Weser River, Germany, in 2012 (BRUNKEN ET AL. 2012). Performing the first comprehensive study to gather records of *N. melanostomus* specimens in the tidal Elbe River (Chapter 4), the gap in knowledge on a possible distribution of the species between the Baltic Sea and Northeast Atlantic region (North Sea) patches should be closed.

When predicting a possible enlargement of the round goby distribution in the North Sea area, it is important to know the salinity tolerance of that species. Coastal areas of the North Sea exhibit a salinity of almost 30–32 (JANSSEN ET AL. 1999) or lower in direct estuary regions. As literature only gives hints on a very wide salinity range for *N. melanostomus* (e.g. SAPOTA 2006, KARSIOTIS ET AL. 2012, KORNIS ET AL. 2012), an aquaria experiment has been conducted to observe growth of juvenile specimens, from the brackish Kiel Canal, under different salinity conditions (Chapter 5). Salinities of up to 30 were used to simulate different ecosystem conditions. This is the first published experiment analysing the salinity tolerance of round goby specimens that stem from brackish water habitats.

For the assessment of possible impacts of established round goby populations on invaded ecosystems it is important to know about the species diet, spawning behaviour and life cycle. All this can diverge strongly between specimens from different ecosystems. For example, the role of *N. melanostomus* as a predator on invertebrates and fishes can strongly influence native species in invaded areas. One of the investigated systems, the Kiel Canal, represents a special case as it is an artificial canal with brackish water conditions. From such an ecosystem no detailed study concerning the population biology of

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round goby is available. With Chapter 6, a study was performed that gives an insight in the feeding ecology, growth and reproductive biology simultaneously.

Hypothetically, the appearance of round goby has the potential to enhance the situation of predatory species in invaded ecosystems. In the study area of the Kiel Canal, a main predatory fish species is the zander *Sander lucioperca* (Linnaeus, 1758). A study on the diet and growth of zander in the Kiel Canal before the round goby invasion (KAFEMANN & THIEL 1998, KAFEMANN 2000) offered the chance to investigate changes after some years of invasion. In Chapter 7, a detailed study was performed on diet composition, growth and condition of zander in a section of the Kiel Canal that was heavily populated by *N. melanostomus*.

First records of the round goby *Neogobius melanostomus* (Pallas, 1814) in the Elbe River, Germany

A similar version of this Chapter was published as:

HEMPEL M, THIEL R (2013) First records of the round goby *Neogobius melanostomus* (Pallas, 1814) in the Elbe River, Germany. BioInvasions Records 2, 291–295.

Abstract

The round goby *Neogobius melanostomus* (Pallas, 1814), a Ponto-Caspian gobiid species, was recorded for the first time in the Elbe River, Germany in May 2008. Between then and June 2013, 22 records comprising 36 specimens of *N. melanostomus* have been collected from the Elbe River, mainly in the tidal river section of the city of Hamburg.

Key words: *Neogobius melanostomus*, round goby, Gobiidae, Elbe River, Germany, non-indigenous species

Introduction

The round goby *Neogobius melanostomus* (Pallas, 1814) is a Ponto-Caspian gobiid species. Its natural distribution comprises the Black, Azov and Caspian Seas and their tributaries (MILLER 1986). The species prefers waters with rocky substrates like stone packing and cobble (RAY & CORKUM 2001) but also inhabits flat sandy or muddy soft areas (JOHNSON ET AL. 2005) if more diverse substrate for spawning is available in nearby areas (SAPOTA & SKÓRA 2005). The round goby reaches a maximum total length of 250 mm (SAPOTA 2006) and has a broad diet chiefly feeding on bivalves, small crustaceans and polychaetes (MILLER 1986, CORKUM ET AL. 2004). Its tolerance of a wide range of environmental factors (CORKUM ET AL. 1998, SAPOTA 2006) and ability to spawn several times a year and high fecundity (MACINNIS & CORKUM 2000) promotes the successful spreading of the species.

Materials and methods

On May 9th 2008, the first individual of *N. melanostomus* was caught in the tidal Elbe River, Germany at 53°31'28"N and 9°59'11"E (Figure 4.1, Appendix 4.1) by a commercial fisherman and was subsequently frozen. The fish was transferred (by Reinhard Diercking) to the fish collection of the Zoological Museum Hamburg (ZMH) for species identification and preservation in 70% ethanol after defrosting. The specimen was deposited in the ZMH fish collection under catalogue number ZMH 25592. From 2011 to 2013, 20 further records were provided, mostly by anglers, comprising 34 specimens of round goby from the tidal Elbe River section of the city of Hamburg (Appendix 4.1). One additional record (Appendix 4.1, record No. 9) was obtained further upstream close to the city of Geesthacht. Five of these specimens were transferred to the ZMH, preserved in 70% ethanol and stored under separate catalogue numbers in the fish collection (Table 4.1). Morphometric measurements were done on the left side of each specimen following HUBBS & LAGLER'S (1958) method. Meristic characters were counted and fish were identified according to MILLER (1986) and KOTTELAT & FREYHOF (2007). Vertebrae, and the spines and rays of dorsal and anal fins of all specimens were counted from radiographs (Figure 4.2) using an X-ray imaging system. Sexes were identified following KORNIS ET AL. (2012), based on the shape and length of the urogenital papilla.



Figure 4.1. Map with the new records of *Neogobius melanostomus* in the Elbe River. Red spots and ZMH numbers indicate specimens stored in the ZMH fish collection. Black triangles and numbers show additional recorded specimens.



Figure 4.2. Round goby, *Neogobius melanostomus*, ZMH 25592, 189 mm *TL*, captured from the tidal Elbe River, Germany, on May 9th 2008. Picture (above) and X-ray (below) taken after preservation in ethanol, meristic characters taken from X-rays are indicated.

Results and discussion

Brief description of the specimens deposited in the ZMH fish collection:

These six specimens of *N. melanostomus* were represented by four males and two females with total weight 4.21–87.69 g and total length 71–189 mm (Table 4.1). This size range comprises juvenile and adult round gobies. The body is elongate and round in cross section. Pelvic fin lengths of the specimens were 0.7–0.8 times the distance from the pelvic fin insertion to the anal fin origin. Pelvic fins do not reach the anus. Colour is brown to yellowish-grey, with several lateral blotches. The first dorsal fin has a large black spot in the posterior (except ZMH 26018). The predorsal area is scaled. Further characters are given in Table 4.1.

	Neogobius melanostomus					
Characters	ZMH 25592	ZMH 25914	ZMH 26002	ZMH 26003	ZMH 26008	ZMH 26018
Total length (TL)	189	121	118	151	71	140
Standard length (SL)	151	98	98	123	59	116
Total weight (TW)	87.69	22.63	23.37	65.33	4.21	42.27
First dorsal-fin spines (D1S)	VII	VI	VI	VI	VI	VI
Second dorsal-fin spines and rays (D2SR)	l,16	I,15	I,15	I,15	I,15	l,15
Anal-fin spines and rays (ASR)	I,12	I,12	I,12	I,12	I,12	I,12
Pelvic-fin spines and rays (VSR)	I,5	1,5	I,5	I,5	I,5	1,5
Pectoral-fin rays (PR)	18	18	18	18	17	18
Lateral line scales (LL)	55	54	55	54	55	55
Predorsal area scaled (PS)	+	+	+	+	+	+
Black Spot on posterior part of first dorsal-fin (BS)	+	+	+	+	+	-
Total vertebrae (Ve)	33	33	33	33	32	33
Sex	male	female	female	male	male	male
% SL						
Body depth (BD)	22.7	26.3	26.3	27.8	20.7	27.1
Head length (HL)	27.7	27.2	27.2	27.0	27.8	26.6
Head depth (HD)	21.7	19.8	19.8	24.4	18.0	21.7
Head width (HW)	25.6	21.1	21.1	25.4	25.6	21.9
Snout length (SnL)	8.8	9.8	9.8	9.6	9.7	9.1
Postorbital head length (PHL)	15.8	14.0	14.0	14.3	13.9	13.4
Orbit diameter (OD)	5.3	8.0	8.0	6.4	7.5	6.7
Interorbital width (IW)	5.7	5.3	5.3	6.7	5.4	5.9
Caudal peduncle depth (CPD)	12.4	11.6	11.6	12.4	12.5	11.6
Caudal peduncle length (CPL)	13.0	15.5	15.5	13.2	18.1	15.8
Prepectoral length (PPL)	29.4	30.3	30.3	28.9	29.5	29.7
Prepelvic length (PVL)	31.5	30.9	30.9	29.2	30.5	30.7
Predorsal length (PD1L)	32.4	37.2	37.2	33.5	35.1	33.8
Preanal length (PAL)	58.3	58.0	58.0	57.6	58.0	56.4
First dorsal-fin height (D1H)	12.1	13.6	13.6	14.8	14.4	12.2
Pectoral-fin length (PL)	30.3	28.3	28.3	27.0	29.2	28.1
Pelvic-fin length (VL)	19.5	21.6	21.6	22.2	22.9	20.4
Orbit diameter/Interorbital width (OD/IW)	0.93	1.50	1.50	0.96	1.38	1.15
Orbit diameter/head length (OD/HL)	0.19	0.29	0.29	0.24	0.27	0.25
Pelvic-fin insertion to anal-fin origin (VI-AO)	28.1	29.8	29.8	28.8	29.0	28.4

Table 4.1. Morphometric measurements and meristic counts for the new records of six specimens of Neogobius melanostomus stored in the ZMH fish collection. Total weight is given in grams. Total and standard lengths are in millimetres, other measurements are expressed as a percentage of standard length.

Identification

The reported specimens agree well with the coloration, body shape, counts and measurements presented by MILLER (1986) and KOTTELAT & FREYHOF (2007). The specimen (ZMH 26018) without a black spot on the posterior part of the first dorsal fin is reliable identified as N. melanostomus based on all other counts and measurements and an additional investigation of the structure of the rows of sub-orbital papillae (MILLER 1986).

Distribution

Nowadays the round goby is one of the most wide-ranging invasive fish species on earth with substantial introduced populations in several European rivers systems, the Baltic Sea and the Laurentian Great Lakes (KORNIS ET AL. 2012). The species is currently spreading in European rivers (KALCHHAUSER ET AL. 2013). Westwards of the Elbe River, the round goby was recorded for the first time in the Dutch Rhine delta (VAN BEEK 2006) in 2004, in the Scheldt River and the Albert Canal in 2010 (VERREYCKEN ET AL. 2011) and in the Weser River in 2012 (BRUNKEN ET AL. 2012). Eastwards, in the Baltic Sea, *N. melanostomus* has developed high abundance in many regions (KORNIS ET AL. 2012). The new records of *N. melanostomus* in the Elbe River (Figure 4.1) can close the distribution gap of round goby between the western and eastern records. The records of adult male and female individuals, the relatively high number of records in 2012 and 2013 and the occurrence of juvenile and adult specimens suggest the establishment of the species in the tidal Elbe River. One preferred substrate of the round goby, stone packing (BORCHERDING ET AL. 2011) and stony groynes, large man-made hydraulic structures directing the river current, are common at the banks of the Elbe. The presence of these man-made substrates could support further spreading into regions upstream and downstream from the recent records.

Both natural dispersal and transport via commercial shipping are possible ways of introduction of *N. melanostomus* into new areas (KORNIS ET AL. 2012). The port of the city of Hamburg in the Elbe River is the 15th-largest port worldwide and is visited by about 12,000 ships every year (HPA 2013). Therefore, round gobies are likely to have plentiful opportunities to reach the tidal Elbe River section around Hamburg within ballast water tanks of ships. Although it is not clear from which original localities the recorded specimens stem, the observation of an individual without a large black spot in the posterior part of the first dorsal fin maybe indicates that at least some of the specimens could originate from the Laurentian Great Lakes. There, specimens of *N. melanostomus* without a black spot on their first dorsal fin were reported from Lake Erie (ISSG 2013). Natural dispersal from the Baltic Sea, as another possible source, seems more improbable because of the lack of early records in the Elbe estuary down-stream of the city of Hamburg. Future genetic analysis could help to clarify the origin of the round gobies recorded in the Elbe River.

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Appendix

Appendix 4.1. Records of *Neogobius melanostomus* in the Elbe River.

Catalogue or	r Location -	Records coordinates		Decord Date	No.	Courses
record number		Latitude, N	Longitude, E	Record Date	specimens	Source
Specimens stored	in the ZMH fish collection					
ZMH 25592	Veddel channel, Hamburg	53°31'28"	9°59'11"	09/05/2008	1	commercial fisherman, Reinhard Diercking
ZMH 25914	Harburg, Hamburg	53°28'24"	9°59'17"	19/10/2012	1	angler
ZMH 26002	Harburg, Hamburg	53°28'22"	9°59'28"	04/05/2013	1	first author
ZMH 26003	Neßsand, Hamburg	53°33'37"	9°45'02"	22/05/2013	1	commercial fisherman
ZMH 26008	Veddeler Brückenstraße, Hamburg	53°31'58"	10°01'38"	14/06/2013	1	first author
ZMH 26018	Müggenburger Zollhafen, Hamburg	53°31'19"	10°01'39"	07/07/2013	1	angler
Additional recor	ds not stored in the ZMH fish c	ollection	_			
1	Harburg, Hamburg	53°28'24"	9°59'17"	23/09/2011	1	angler
2	Oortkaten, Hamburg	53°26'46"	10°05'37"	Oct. 2011	1	angler
3	Fischmarkt, Hamburg	53°32'37"	9°56'47"	24/03/2012	1	angler
4	Landungsbrücken, Hamburg	53°32'43"	9°58'12"	29/03/2012	1	angler
5	Harburg, Hamburg	53°28'22"	9°59'23"	13/05/2012	1	angler
6	Kattwykbrücke, Hamburg	53°29'36"	9°57'00"	14/06/2012	4	angler
7	Finkenried, Hamburg	53°28'23"	10°00'18"	16/07/2012	4	angler
8	Kattwykbrücke, Hamburg	53°29'34"	9°57'19"	15/08/2012	1	angler
9	near Geesthacht	53°24'48"	10°23'26"	Aug. 2012	1	commercial fisherman
10	Teufelsbrück, Hamburg	53°32'46"	9°51'48"	12/09/2012	1	angler
11	Kattwykbrücke, Hamburg	53°29'43"	9°57'10"	22/09/2012	2	angler
12	Harburg, Hamburg	53°28'24"	9°59'17"	11/10/2012	5	angler
13	Fischmarkt, Hamburg	53°32'38"	9°57'03"	Nov. 2012	2	angler
14	Köhlbrand, Hamburg	53°31'18"	9°56'12"	01/05/2013	2	angler
15	Landungsbrücken, Hamburg	53°32'44"	9°57'39"	22/05/2013	2	angler
16	Zweibrückenstraße, Hamburg	53°32'04"	10°01'20"	07/07/2013	1	angler

Contribution of authors to Chapter 4

Mattias Hempel collected the examined specimens of round goby except the first record, identified the species and took all measurements and counts. He made the map, prepared all Figures and Tables, wrote the initial draft of the manuscript, the comments to the editor and the reviewers and the revised version of the manuscript. Ralf Thiel provided the first record from 2008 (ZMH 25592) and assisted in the laboratory work and the writing process.

5 Effects of salinity on survival, daily food intake and growth of juvenile round goby *Neogobius melanostomus* (Pallas, 1814) from a brackish water system

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Abstract

The growth response of juvenile round gobies was tested in various salinities to assess the potential performance of the species in widely differing salinities in the Baltic Sea and adjacent waters. An experimental approach analyzed the survival, daily food intake and growth of juvenile Neogobius melanostomus from the brackish water region of the Kiel Canal, Germany. Round gobies (TL = 61.5±5.3 mm, *n* = 40) were fed *ad libitum* (diet = mysid shrimp, energy content = 4.7 cal per mg dry weight) at salinity levels of 0.1, 7.5, 15 and 30 over 12 weeks. Two replicate tanks (125 x 30 x 30 cm, 100 L) were maintained for each salinity level, each holding five specimens at a mean temperature of 19.90°C (±0.64°C, n = 136). Survival at all levels was 100%, except for 0.1 with 90%. Mean daily food intake (% of body weight) increased from salinity level 0.1 to 15 (from 17.76% to 19.95%); specimens at level 30 showed a more diversified and lower mean food intake (18.44%). Mean cumulative body weight gain appeared greatest at a salinity level of 7.5 (224.52%), closely followed by 0.1 (206.57%) and 15 (191.92%). In contrast, fish growth was significantly lower at a salinity level of 30 (100.57%). Results of the experiment indicate that salinity tolerance estimates of the round goby made by other authors are too low regarding specimens that stem from brackish habitats. Growth patterns of some specimens, even in the salinity 30 treatment, show that a further spread of this species into regions with higher salinities is quite possible.

Introduction

The round goby *Neogobius melanostomus* (Pallas, 1814) is a benthic fish reaching a maximum total length of 250 mm (SAPOTA 2006). It is a highly invasive Ponto-Caspian gobiid species with introductions into many ecosystems in Europe and North America (KORNIS ET AL. 2012).

Round goby populations inhabit ecosystems with a wide range of salinity levels. On the one hand they are known from freshwater, for example in the Rhine (BORCHERDING ET AL. 2011) and Danube (WIESNER 2005) rivers in Europe and from the Laurentian Great Lakes in North America (CHARLEBOIS ET AL. 2001). On the other hand the native distribution of the round goby is located in the area of the Black Sea with a mean salinity of 19, the Caspian Sea with a mean salinity of 12.8 (CHARLEBOIS ET AL. 1997), and the Sea of Azov with a maximum salinity of 13 (REID & ORLOVA 2002), and is also native to the tributaries of these waters (MILLER 1986). As described in the review paper of KORNIS ET AL. (2012), there is no established population of round goby under full marine salinity conditions and its salinity tolerance in marine waters is assumed to be below 30.

However, there is no published experiment dealing with survival, daily food intake and growth in different salinity levels with round goby that stems from brackish water. Thus, to determine this we conducted an experiment with four different salinity levels meant to simulate pure freshwater (0.1), brackish estuary waters (7.5), a salinity content comparable with the western part of the Baltic Sea (15), and a salinity nearly as high as full ocean salinity (30). The experimental design was planned partially following the procedure described by ONADEKO & KUSEMIJU (1991). The experiment was conducted to assess the potential performance of the species now found in very different salinities of the Baltic Sea and adjacent waters.

Materials and methods

Fish collection, transport, acclimatization and maintenance

Round goby specimens used in the experiments were caught by beach seining in autumn (20 September 2011) in the Kiel Canal, Germany, near the city of Rendsburg ($54^{\circ}20'39''$ N, $9^{\circ}44'58''$ E). Salinity at the sampling point was 3.2 and water temperature was 18.4°C. Fishes were transported in an aerated plastic tank with circa 100 L of water from the sampling point to the research aquarium, where they were acclimatized to a salinity of 0.1 over a one-day period and maintained in a glass tank (240 x 50 x 60 cm) containing about 600 L of water. Water temperature was 20.6°C ($\pm 0.22^{\circ}$ C, n = 9, Table 5.1). Bottom substrate was sand, with shelter structures for the fishes (plastic pipes and broken clay pots). An external biological filter-unit (Eheim[®] professionel 3 2075), cleaned every second week, was used to operate the tank in a closed-system mode. Round gobies were kept in a dark room at a
constant 12-hour-light: 12-hour-dark photoperiod with a fluorescent light (Philips[®] Master 58 W) installed 40 cm above the tank. The fish were fed six days a week with frozen mysid shrimp *Neomysis integer* (Leach, 1814) (energy content = 4.7 cal per mg dry weight (ASTTHORSSON 1980)) caught in the Kiel Canal near the sampling point and defrosted before use. To maintain adequate water quality the parameters of salinity, water temperature, pH and dissolved oxygen concentration were measured weekly (Table 5.1) using a multi-probe U50 (Horiba[®]). Additionally, nitrite, nitrate, ammonium/ammonia and phosphate contents were quantified with aqua-tests (JBL[®]). One-third of the water was changed weekly.

Experimental design

The experiments were performed in eight rectangular glass aquaria ($125 \times 30 \times 30$ cm, 100 L) with two replicate tanks for each salinity level (0.1, 7.5, 15 and 30). Salinity levels were prepared by mixing the supply water with Red Sea Salt (Red Sea[®]). Each tank was divided with mesh-slides (mesh size 1 mm) into five compartments with an area of 20 x 30 cm and a water volume of 16 L. Sand and the half of a clay pot were added to each compartment to provide natural substrate and shelter. A separate filter-system (Eheim[®] classic 2211), cleaned every second week, was used for each tank in the closed-system mode. Fishes were held as before in the same light regime (12/12, fluorescent light Philips[®] Master 58 W, 100 cm above tanks) at a water temperature of 19.9° C ($\pm 0.64^{\circ}$ C, n = 136). Salinity, water temperature, pH and dissolved oxygen concentration were measured at least once a week. At the same time, nitrite, nitrate, ammonium/ammonia and phosphate were quantified with aqua-tests (Table 5.1). To maintain adequate water quality, half of the water in the tanks was changed weekly. In the case of elevated nitrogen or phosphate values, additional water changes were performed to avoid any influence on the experimental results. This occurred five times during acclimatization (in the first four weeks in the experimental tanks).

Ten round goby specimens (40 total) were adapted to a salinity level (0.1, 7.5, 15 and 30) over time with a maximum change in salinity of 5.0 per day. Afterwards, each of the 40 individuals was stocked separately in a compartment of the tank and acclimatized over a four-week period before beginning the experiment. The four weeks were also used to ascertain an approximate food intake to determine the later amount of *ad libitum* feeding during the experiments. Weight was 2.76 g (±0.78 g, *n* = 40, range 1.53–4.60 g) and *TL* was 61.5 mm (±5.3 mm, *n* = 40, range 53–73 mm) at the beginning of the experiment. ANOVA (analysis of variance) in SPSS[®] (IBM[®] Statistics) was used to verify that mean fish weight did not differ among the salinity treatments. Once a week, all gobies were individually measured for body weight (to 0.01 g precision) and total length (nearest mm). Six days a week they were fed *ad libitum* with mysid shrimp equivalent to 25% of their body weight at the last weighing.

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Tank	Time (weeks)	n	Parameter							
			Salinity	Temperature (°C)	рН	O ₂ (mg L ⁻¹)	Nitrite (mg L ⁻¹)	Nitrate (mg L ⁻¹)	Ammonia (mg L ⁻¹)	Phosphate (mg L ⁻¹)
Acclimatization	9	9	0.14 (0.05)	20.6 (0.22)	7.27 (0.33)	7.98 (0.50)	0.05 (0)	5 (0)	0.5 (0)	0.1 (0)
(1) Salinity 0.1	16	17	0.12 (0.05)	19.74 (0.57)	6.89 (0.29)	8.74 (0.79)	0.07 (0.14)	3.01 (2.19)	0.09 (0.13)	0.48 (0.49)
(2) Salinity 0.1	16	17	0.11 (0.06)	19.76 (0.57)	6.88 (0.33)	8.66 (0.70)	0.08 (0.10)	2.83 (2.12)	0.09 (0.13)	0.40 (0.47)
(3) Salinity 7.5	16	17	7.75 (0.16)	19.62 (0.91)	7.54 (0.16)	8.85 (0.53)	0.24 (0.21)	3.06 (2.13)	0.16 (0.21)	0.41 (0.49)
(4) Salinity 7.5	16	17	7.65 (0.20)	19.81 (0.47)	7.50 (0.15)	8.85 (0.15)	0.24 (0.23)	3.01 (2.19)	0.16 (0.21)	0.41 (0.49)
(5) Salinity 15	16	17	15.19 (0.32)	20.23 (0.61)	7.66 (0.15)	8.76 (0.72)	0.28 (0.26)	3.24 (2.80)	0.20 (0.24)	0.31 (0.49)
(6) Salinity 15	16	17	15.17 (0.28)	19.86 (0.58)	7.69 (0.13)	8.71 (0.67)	0.24 (0.26)	3.48 (2.77)	0.20 (0.24)	0.34 (0.49)
(7) Salinity 30	16	17	30.19 (0.43)	20.27 (0.55)	7.91 (0.15)	8.77 (0.55)	0.17 (0.20)	2.95 (2.86)	0.27 (0.24)	0.29 (0.47)
(8) Salinity 30	16	17	30.15 (0.32)	19.89 (0.56)	7.91 (0.12)	8.78 (0.49)	0.16 (0.19)	2.48 (2.81)	0.27 (0.24)	0.30 (0.48)

Table 5.1. Mean water parameters, standard deviations (in brackets) measured in acclimatization (acclimatization) and experimental tanks (1–8).

Time (weeks) = time of keeping *Neogobius melanostomus* specimens in the tank, *n* = number of measurements. While faeces and uneaten mysid shrimp were removed daily, weight of the remaining mysids was taken one day per week (weeks 7–12) to determine the daily food intake of the fish. It was supposed that the daily food intake was more sensitive and variable than the weekly growth and survival. Additional, a drop in food intake is possible in the initial phase due to stress associated with the change in environment (see NORDGARDEN ET AL. 2003). Thus, acclimatization time for measuring daily food intake was extended until week seven of the experiment.

The experiment lasted 12 weeks (84 days). Statistical analyses of differences in growth and daily food intake between the salinity treatments were performed using ANOVA (comparable to KARSIOTIS ET AL. (2012)). When a significant difference occurred (p < 0.05), Fisher's least significant difference (LSD) post hoc tests were added to determine the source.

In addition, the (1) specific growth rate, (2) Fulton's condition factor, and (3) the food conversion efficiency of all individuals were calculated following ONADEKO & KUSEMIJU (1991):

(1) Specific growth rate (%) =
$$\frac{\ln \text{ final body weight (g)} - \ln \text{ initial body weight (g)}}{\text{Time (days)}} \times 100$$

(2) Fulton's condition factor = $\frac{\text{Total body weight (g)}}{\text{Total length (cm)}} \times 100$

(3) Food conversion efficiency (%) = $\frac{\text{Total weight gain (g)}}{\text{Total feed intake (g)}} \times 100$

Results

Survival and condition

Acclimatization of the round gobies to the selected salinities was performed with no obvious problems. Survival in the experiment was high, with only one dead specimen (salinity level 0.1). At salinity level 7.5, one female specimen showed spawning activity associated with weight loss. Both aforementioned specimens were excluded from further analysis. One specimen in salinity treatment 30 was in poor condition at the end of the experiment, but was still considered for further analysis.

Daily food intake

Mean daily food intake at different salinity levels (weeks 7–12 of the experiment) increased from salinity 0.1 over salinity 7.5 to salinity 15. Mean daily food intake of specimens at salinity level 30 was lower, with a wide range among the ten specimens (Figure 5.1). We found significant differences between the salinity levels (ANOVA, F(3, 37) = 3.286, p = 0.032), with daily food intake at salinity 15 higher than at 0.1 (p = 0.005) and 30 (p = 0.042).



Figure 5.1. Daily food intake (% of body weight) of round goby (*Neogobius melanostomus*) at different salinity levels. Experimental weeks 7–12, n = 9 at salinity levels 0.1 and 7.5, n = 10 at salinity levels 15 and 30.

Growth

Mean weights at the beginning of the experiment did not differ among the investigated salinity levels (F(3, 37) = 1.594, p = 0.209). During the 12-week experiment, growth of individual specimens showed a wide variation in weight gain, ranging from -16.60 to 299.63%. Best mean weight gain was at salinity level 7.5 (Figure 5.2, Table 5.2), closely followed by the salinity levels of 0.1 and 15. Mean weight gain at salinity level 30 was much lower than in all other salinities.



Figure 5.2. Mean cumulative weight gain (%) of round goby at different salinity levels during 12 experimental weeks (84 days). n = 9 at salinity levels 0.1 and 7.5, n = 10 at salinity levels 15 and 30.

Analysis of variance (ANOVA) showed that differences in weight gain at the investigated salinities were highly significant at the 5% level (F (3, 37) = 8.536, p = 0.000) after 84 days. Results of the Fisher LSD post-hoc tests revealed that mean weight gain at salinity level 30 differed significantly from all other treatments by week 6, with increasing differences until the end of the experiment (week 12). Growth of gobies at salinity level 30 was significantly slower than at the 0.1 (p = 0.000), 7.5 (p = 0.000) and 15 (p = 0.001) levels. Among the other salinity treatments, no significant differences in weight gain were observed at any time during the experiment.

Highest specific growth rate was at salinity level 7.5, with level 30 the lowest (Table 5.2). Fulton's condition factor showed the same characteristics, with the best mean condition factor at salinity level 7.5; salinity level 30 had the worst mean condition factor. Food conversion efficiency showed a slightly different trend, with the highest food conversion efficiency achieved in the freshwater treatment followed by salinity levels 7.5, 15, and 30 (Table 5.2).

	Salinity level							
Parameter	0.1	7.5	15	30				
	(<i>n</i> = 9)	(<i>n</i> = 9)	(<i>n</i> = 10)	(<i>n</i> = 10)				
Initial body weight (g)	3.01 (0.89)	2.99 (0.76)	2.76 (0.65)	2.34 (0.76)				
	<i>1.97–4.60</i>	2.04–4.35	<i>1.94–</i> 3.77	<i>1.53–3.96</i>				
Final body weight (g)	9.18 (2.95)	9.82 (3.06)	8.09 (2.19)	4.52 (1.95)				
	6.07–13.34	5.21–13.48	5.09–10.82	<i>1.96–</i> 8.04				
Total weight gain (g)	6.16 (2.25)	6.83 (2.39)	5.32 (1.66)	2.18 (1.88)				
	<i>3.64–</i> 9.19	<i>3.17–9.61</i>	<i>2.97</i> –7.66	-0.39 to 4.68				
Total weight gain (%)	206.57 (56.55)	224.52 (48.30)	191.92 (41.54)	100.57 (80.26)				
	134.81–288.09	155.39–299.63	<i>135.62–268.77</i>	-16.60 to 238.78				
Specific growth rate (% / day)	1.32 (0.22)	1.39 (0.18)	1.27 (0.16)	0.73 (0.55)				
	<i>1.02–1.61</i>	<i>1.12–1.65</i>	<i>1.02–1.55</i>	-0.22 to 1.45				
Fulton's condition factor	1.30 (0.11)	1.33 (0.13)	1.28 (0.09)	1.12 (0.12)				
	<i>1.15–1.42</i>	<i>1.11–1.52</i>	<i>1.16–1.44</i>	<i>0.86–1.26</i>				
Food conversion efficiency (%)	7.47 (1.16)	7.42 (0.77)	6.75 (0.90)	4.12 (3.15)				
	<i>6.19–9.15</i>	<i>6.13–8.61</i>	<i>5.43–8.12</i>	-1.65 to 8.04				

Table 5.2. Mean growth parameters of round goby at different salinity levels during 12 experimental weeks (84 days). Mean values given together with standard deviation (in brackets) and related range (in italics).

Discussion

The wide salinity tolerance (e.g. CHARLEBOIS ET AL. 1997) of the round goby was confirmed in our study by the high survival rate over a broad range of salinities (0–30). Survival of all specimens at salinity level 30 is in disagreement with other published experiments on the salinity tolerance of round goby. In ballast water exchange experiments (ELLIS & MACISAAC 2009) representing a pure freshwater population, no N. melanostomus from the Detroit River, Canada, survived (5-h stepwise rise in salinity from freshwater to a salinity of 30) after 48 h. A freshwater round goby population from Maumee Bay, Lake Erie, OH, USA, was also used in an experiment dealing with ballast-water exchange (KARSIOTIS ET AL. 2012). In their 4-month approach, all gobies died within one month at salinities of 25 and 30, and only 5% survived at a salinity level of 20. Discrepancies in the results of ELLIS & MACISAAC (2009) and KARSIOTIS ET AL. (2012) and our study might be caused by the different origins of the specimens. In contrast to the pure freshwater Lake Erie area populations, the Kiel Canal gobies used in our study represent a brackish water population. In another species, the Adriatic sturgeon Acipenser naccarii Bonaparte, 1836, MCKENZIE ET AL. (2001) described a better tolerance for salinity changes in individuals acclimatized to brackish water by comparison to specimens stocked in freshwater. VETEMAA & SAAT (1996) found a higher salinity tolerance for ruffe Gymnocephalus cernua (Linnaeus, 1758) embryos from a brackish water population in comparison to those from freshwater. In the sampling point area we measured salinities of up to ~7 in 2011 and 2012. In addition, KARSIOTIS ET AL. (2012) noted that different genotypes of round gobies stemming from the Black Sea or the Caspian Sea could lead to differences in salinity tolerance. Results of our study indicate that a ballast water exchange to a salinity

level of 30 for \geq 24 h (as appears necessary in KARSIOTIS ET AL. (2012)) is not effective in eliminating further spread or introductions of the round goby.

A possible explanation for the increasing daily food intake with increasing salinity (Figure 5.1) could be the larger energy budget needed for osmoregulation. The review study of BŒUF & PAYAN (2001) reported that approximately 10 to >50% of the energy budget of fish is dedicated to osmoregulation and that better growth at intermediate salinities (8–20) is often correlated with a lower standard metabolic rate. Most teleost fishes have plasma solute concentrations of about 10.5 (KARSIOTIS ET AL. 2012). Accordingly, the salinity optimum of the Kiel Canal round gobies seems to be somewhere below 15. Up to a salinity level of 15, round gobies could balance the higher metabolic rate with a higher food intake. At a salinity level of 30, only some of the fish were able to balance their metabolism, while others showed a depressed food intake and growth.

Growth of fishes is linked to appropriate conditions of the inhabited environment in food availability and water temperature (STARRS ET AL. 2013) and salinity (BŒUF & PAYAN 2001). In our experimental design the salinity level was the only difference among treatments, while food availability and temperature were held constantly. Thus, growth at the different salinity levels was a direct hint of habitat suitability in relation to salinity. A salinity level of at least up to 15 seems to have provided good conditions for all specimens. In the 30 salinity level treatment, specimen growth was very diverse, with weight gains of -16.6 to 238.78% in body weight. Six of ten fishes showed a weight gain of more than 100%. With regard to this result it seems possible that a part of the Kiel Canal population has the ability to live in regions with higher salinities (30), or maybe even in full salinity of the North Sea. This must be more challenging to the gobies, but at least a temporary life in this higher salinity cannot be ruled out. A possible barrier could be from an earlier life stage of the round goby, whereby it is not fully clarified whether egg development is possible at high salinity levels. KARSIOTIS ET AL. (2012) described the Black Sea round goby population as having reproduced successfully only in salinities <19; however, due to possible differences between populations, further experiments are needed to rule out the possibility of successful reproduction at higher salinities.

Results of our experiment yielded new findings on the salinity tolerance of the invasive *N. melanostomus*. Tolerance of populations from brackish water environments can be higher than those estimated by the different authors for specimens from freshwater populations. A spread of the round goby population from the Kiel Canal to the coastal areas of the North Sea or to other marine areas with comparable high salinities cannot be ruled out.

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Contribution of authors to Chapter 5

Mattias Hempel collected the involved specimens, conducted the aquaria experiment and made all calculations and statistics. He prepared all Figures and Tables, wrote the initial draft of the manuscript, the comments to the editor and the reviewers and the revised version of the manuscript. Ralf Thiel assisted in the work with the aquaria and the writing process.

6 Feeding ecology, growth and reproductive biology of round goby *Neogobius melanostomus* (Pallas, 1814) in the brackish Kiel Canal

A similar version of this Chapter was prepared in collaboration with Victoria Magath, Rüdiger Neukamm and Ralf Thiel and submitted to Marine Biodiversity, a peer-reviewed journal.

Abstract

We inspected diet, growth and reproductive biology of round goby *Neogobius melanostomus* (Pallas, 1814) from the Kiel Canal, northern Germany, to give an insight into the life cycle of this species in a brackish water canal. Crustaceans had the highest importance in the diet of all inspected size classes of round goby. Other major food taxa were annelids and chironomids. Mussels and fish were consumed almost exclusively by medium- and large-sized *N. melanostomus* and contributed to the diet to a minor extent. In addition to the variability between size classes, we found a high interannual variability in the prey species. Round goby in the Kiel Canal was characterized by relatively low asymptotic total length and high relative fecundity. An almost balanced sex ratio indicates the established status of the population. The highest spawning activity took place between May and the end of June. From our data, negative effects of round goby on native species due to competition for prey and habitat are more likely than due to direct predation on eggs or fry. Affected species with similar habitat requirements are for example small gobiids from the genus *Pomatoschistus*. As round goby occurs in high abundance, radical changes of the Kiel Canal ecosystem seem possible and likely.

Key words: Gobiidae, invasive species, diet analyses, fecundity, sex ratio

Introduction

The round goby *Neogobius melanostomus* (Pallas, 1814) is a highly invasive fish species of our time (see e.g. BROWNSCOMBE & FOX 2012). In many cases it spreads fast into new regions and reaches high densities shortly after its first occurrence (e.g. BORCHERDING ET AL. 2011, HEMPEL ET AL. 2016). When occurring in high numbers, the impact of the non-indigenous *N. melanostomus* on the invaded ecosystem and its native inhabitants can be fundamental (see JANSSEN & JUDE 2001, VAN KESSEL ET AL. 2016).

Round goby can directly compete with native fish species for resources, habitat and spawning places. Therefore, it is able to affect those species strongly and even displace them locally (see KORNIS ET AL. 2012). In Lake Michigan, North America, abundance of the fish species mottled sculpin Cottus bairdii Girard, 1850 and Johnny darter Etheostoma nigrum Rafinesque, 1820 have strongly declined after the establishment of round goby (LAUER ET AL. 2004). In the area of Calumet Harbor, southern Lake Michigan, mottled sculpin had nearly disappeared four years after the first occurrence of N. melanostomus (JANSSEN & JUDE 2001). In many large European streams, round goby and other invasive gobiid species such as bighead goby Ponticola kessleri (Günther, 1861) and monkey goby Neogobius fluviatilis (Pallas, 1814) have built up high densities in the last two decades. Often the majority of fishes captured in nearshore habitats are non-indigenous gobiids (e.g. BORCHERDING ET AL. 2011, BRANDNER ET AL. 2013b). This represents a high potential for competition with native species. In the Meuse River, the Netherlands, a rapid decline in the densities of native river bullhead Cottus perifretum Freyhof, Kottelat & Nolte, 2005 after the occurrence of N. melanostomus in 2011 was detected (VAN KESSEL ET AL. 2016). At sites where only other invasive gobiids (bighead goby and western tubenose goby Proterorhinus semilunaris (Heckel, 1837)) were present, river bullhead density remained relatively high. At sites with established round goby populations it decreased to nearly zero. For the Bay of Gdańsk, Baltic Sea, a high overlap in the diets of European flounder Platichthys flesus (Linnaeus, 1758) and N. melanostomus was reported (KARLSON ET AL. 2007). Flounder seems to be displaced to suboptimal habitat by round goby as densities in flat nearshore areas have declined since the establishment.

Due to direct predation, round goby can alter invertebrate communities (e.g. KIPP & RICCIARDI 2012) and threaten invertebrate species at risk (POOS ET AL. 2010). In Lake Michigan, reduced densities of dreissenid mussels, zebra mussel *Dreissena polymorpha* (Pallas, 1771) and quagga mussel *Dreissena bugensis* (Andrusov, 1897), as well as other macroinvertebrates were found, e.g. by KUHNS & BERG (1999) and LEDERER ET AL. (2008). The biomass of benthic algae increased with round goby density because of its predation on gastropods and other algivores (KUHNS & BERG 1999, KIPP & RICCIARDI 2012). This was assumed to have extensive impacts on the trophic levels in areas with established populations of round goby.

As confirmed for birds (JAKUBAS 2004), snakes (KING ET AL. 2008) and many fish species (e.g. STEINHART ET AL. 2004, MADENJIAN ET AL. 2011, CRANE ET AL. 2015), round goby itself can contribute strongly to the diet of predators. In some cases, this can enhance the situation of the predator as shown for example with better condition and growth (e.g. CRANE ET AL. 2015, HEMPEL ET AL. 2016). On the other hand, incorporating *N. melanostomus* into the diet of birds could be linked with outbreaks of botulism in Lake Michigan due to the transfer of toxins (YULE ET AL. 2006).

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Growth and maximum size of round goby differ strongly between populations in different ecosystems. In its natural distribution area, the Black, Azov and Caspian Seas and their tributaries, round goby reaches a total length (*TL*) of 220 mm (MILLER 1986). In many introduced populations it is described to stay smaller and dwarf forms of *N. melanostomus* can occur in freshwater and slightly brackish water (MACINNIS & CORKUM 2000, CORKUM ET AL. 2004). On the other hand, the introduced population in the Bay of Gdańsk under slightly brackish conditions originates the largest specimens reported for the species. SOKOŁOWSKA & FEY (2011) sampled specimens with up to 235 mm total length.

Range expansion of round goby is also promoted by its spawning behaviour. As a batch spawner it can repeat spawning from April to October at temperatures of 9–26°C and eggs are guarded by the males (e.g. CHARLEBOIS ET AL. 1997, KORNIS ET AL. 2012). Age at maturity in the native Pontocaspian area is 2–3 years for females and 3–4 years for males (MILLER 1986). Specimens in introduced populations are described to mature earlier (e.g. MACINNIS & CORKUM 2000). Studies from different ecosystems confirm different strategies in spawning behaviour of round goby. In the Lower Rhine, Germany, the spawning intensity of round goby is usually highest at the beginning of the season (April/May) but behaviour varies strongly between years (GERTZEN ET AL. 2016). In the Bay of Gdańsk, the most advanced gonad stages were detected in April and July, indicating two peaks in spawning activity (TOMCZAK & SAPOTA 2006). Sex ratio in stable native populations is nearly 1:1 while in introduced populations the percentage of males can be considerably higher (TOMCZAK & SAPOTA 2006), with up to six males to one female (CORKUM ET AL. 2004).

Since the occurrence of *N. melanostomus* can cause extensive changes in ecosystems and since it is one of the most prominent invasive species of our time, it is a popular study subject. Numerous scientific papers have been published describing the role of round goby in large freshwater streams and lakes and in brackish areas of the Baltic Sea and the Pontocaspian area. In spite of all the studies mentioned, no details about round goby are available from the ecosystems of brackish artificial waters. Such systems differ strongly from natural waters in their morphology and in the level of anthropogenic impacts, for example due to changed hydrodynamics and highly variable flow velocities induced by shipping traffic (see ARLINGHAUS ET AL. 2002). To close this gap in knowledge, we performed our study in the brackish Kiel Canal, northern Germany, which is a major habitat of round goby in that area and inhabited by a dense population of this species (see HEMPEL ET AL. 2016). We provide a comprehensive insight by describing the diet, growth and reproductive biology of *N. melanostomus* combined in one study.

Materials and methods

Study area

The Kiel Canal in northern Germany, which connects the North Sea with the Baltic Sea, is a highly frequented artificial waterway (see HEITMANN ET AL. 2013). The canal, with a length of about 100 km and a width that varies between 162 and 240 m (KAFEMANN ET AL. 2000), was built in the late 19th century. The guaranteed depth of its navigation channel is 11 m (WSD NORD 2015). The banks of the canal are commonly stabilised by stone packaging to avoid damage due to shipping traffic. Mainly due to the connection with the Baltic Sea, the canal shows a salinity gradient with highly brackish water at the eastern end and westwards decreasing salinity. Mean salinity was ~3 near the western end and ~10 at the eastern end in 2011 and 2012, at the study site it was ~7. These conditions allow freshwater, euryhaline and marine taxa to live in the Kiel Canal. KAFEMANN (2000) recorded 46 fish species in 1995/1996. Some of these occur in high numbers depending on the season, for example, the Atlantic herring *Clupea harengus* Linnaeus, 1758. Adults migrate into the canal in spring for spawning, whereas the juveniles stay in the canal during summer. The study site (54°19′19″ N, 9°42′49″ E) was situated east of the city of Rendsburg (Figure 6.1). In this area, the Kiel Canal cuts through a former lake. Thus, the banks show a relatively natural morphology with enlarged flats in comparison to other sections of the canal.



Figure 6.1. Location of the study site.

Sampling

Sampling of *N. melanostomus* was performed with a beach seine in bankside areas (depth <2 m) and with a demersal trawl in deeper areas more distant from the banks (depth >10 m). The used seine net (length 30 m, width 3 m, mesh size 4 mm) was deployed vertically to the banks by a small motor operated boat and dragged back to it trying to cover the largest area possible. Bottom material was mainly sand with only a few macrophytes. In 2011, monthly sampling by seine took place from May to October while in 2012 it was used from April to October. Demersal trawling was realized in collaboration with a resident commercial fishery. The net had an opening width of 14 m and a mesh size of 10 mm in the cod end. It was trawled along the bottom with a speed of ~2.5 knots (see HEMPEL ET AL. 2016, Chapter 7). Bottom substrate in this depth was mainly sand and mud. In 2011 and 2012, monthly sampling took place with 10 km trawling distance in total (5 hauls, each 1–3 km in length) from June to October. Immediately after catching, all round goby specimens were frozen at -20 °C. Afterwards, they were preserved in formaldehyde solution in the laboratory (4%, buffered with sodium tetraborate) until further analyses took place.

Diet analysis

We examined the digestive tracts of 533 round gobies for prey consumed. In order to investigate possible diet shifts or seasonal influences on diet, we grouped fish by three length classes (TL: small <50 mm (*n* = 138), medium 50–99 mm (*n* = 242) and large ≥100 mm (*n* = 153)) and studied month, respectively. Specimens designated for analysis of gut contents were watered overnight to wash out formaldehyde solution. Afterwards, we measured total length (mm below) and total weight (nearest 0.01 g). As round goby lacks a real stomach, the whole digestive tracts from oesophagus to rectal sphincter were analysed (see GERTZEN 2016). Prey items from the tracts were determined to the lowest taxon possible, counted and their relevant structures (bones of fishes or body parts of invertebrates; e.g. dentary bones, mussel shells, jaws) measured for subsequent back calculation of ingested biomass. Biomass was calculated using published regression equations between measurements and fresh weight of the species or by using standard biomasses (e.g. ÓLAFSSON & PERSSON 1986, DEBUS 1987, DEBUS & WINKLER 1996). Whenever no data were available, we calculated own regression equations in order to determine fresh weight of prey items. For lagoon cockle Cerastoderma glaucum (Bruguière, 1789) we used: $W_t = 0.00009 \times D_{max}^{3.3380}$ (freshly swallowed specimens from the digestive tracts of round goby: n = 7, range 8.6–12.5 mm (D_{max}), $r^2 = 0.7283$, p = 0.015) to calculate total weight of the prey from its largest diameter (D_{max}). For sea squirt Ascidiacea we used: $W_t = 0.0002 \times D_{max}^{2.6093}$ (freshly swallowed specimens from the digestive tracts: n = 9, range 2.8–9.0 mm (D_{max}), $r^2 = 0.9568$, p < 0.001). For Harris mud crab Rhithropanopeus harrisii (Gould, 1841) we used: $W_t = 0.0001 \times W_c^{3.2245}$ (freshly swallowed specimens from the digestive tracts: n = 9, range 6.4–13.0 mm (W_c), $r^2 = 0.8321$, p < 0.001) to calculate total weight of the prey from its carapace width (W_c). For bay barnacle Amphibalanus improvisus (Darwin, 1854) we used: $W_t = 0.0001 \times D_{max}^{3.2618}$ (specimens freshly sampled in the lower Trave River, Germany: n = 35, range 4.0–10.1 mm (D_{max}), $r^2 = 0.925$, p < 0.001) to calculate total weight of the prey from its largest diameter at the base of the shell (D_{max}). All calculated fresh weights included the shells.

In order to analyse the importance of different prey items, we used the relative importance index (*RI*) of prey taxa following GEORGE & HADLEY (1979). It was calculated as follows: RI_i (%) = ($N_i + B_i + F_i / \sum (N_i B_i F_i)$) × 100, where N_i (%) is the number, B_i (%) is the biomass and F_i (%) is the frequency of occurrence of the prey component *i* in the sample.

Statistical analyses on diet composition were conducted in R version 3.2.2 (R Core Team 2015). For temporal diet analysis functions metaMDS, simper and anosim from the package vegan (OKSANEN ET AL. 2015) were used. Nonmetric multidimensional scaling (nMDS) was applied to compare the diet per month. Bray-Curtis dissimilarity index was used. Maximum number of random starts was set to 100. A scree plot was used to determine the optimal number of dimensions (2). Stress was calculated as sum of squared differences between fitted and predicted distances from the regression function. A cluster dendrogram was used to analyse differences in diet between length classes of gobies.

Growth estimation

For growth estimation of round goby we investigated 133 specimens from the study area. These fish were measured in the same manner as for diet analysis. Afterwards, ten scales from the left side between the first dorsal fin and the lateral line were removed for determination of age. Otoliths (sagittae and lapilli) were inspected as well, but scales were much better to read. We cleaned the removed scales from adherent epidermis and stored them between two glass slides (see GRULA ET AL. 2012). Counting of annuli took place using a stereomicroscope (Leica MZ 9.5) and under transmitted light. We excluded regenerated and not clearly readable scales from analysis. A second person read all scales. From the 133 specimens inspected, five were excluded from further analysis due to poorly developed annuli. 128 were used for growth estimation (*TL* <50 mm: n = 8, 50–99 mm: n = 72, 100–149 mm: n = 45, ≥150 mm: n = 3).

To define the exact age of round goby specimens, we used the 15th of June as a hypothetical date of birth. This was defined without consideration of the batch spawning behaviour (repeated spawning events). The decrease in the gonadosomatic index of females after the beginning of June indicates that the highest spawning activity takes place at that time of the year.

The relationship between total weight (W_t) and TL was characterized by the equation: $W_t = a \times TL^b$, where a is a coefficient for the weight at length and b is a shape parameter for the body form. Growth

in length was calculated using FISAT II version 1.2.2 (FAO, Rome, Italy) by the von Bertalanffy growth equation: $L_t = L_{\infty} \times (1 - e^{-k(t - t_0)})$, where L_t is the length at time t (*TL*, mm), L_{∞} is the asymptotic length (*TL*, mm), *k* is a growth coefficient and t_o is the prenatal age (years).

Determination of fecundity, spawning period and sex ratio

We identified the sex of 3,049 specimens caught during demersal trawling in 2011 and 2012 by examination of the urogenital papillae that is long and pointed in males but broad and blunt in females (see KORNIS ET AL. 2012). To test if there is an influence of the sampling date on the results these fish were grouped by month (June: n = 880, July: n = 767, August: n = 733, September: n = 636, October: n = 33). To identify possible differences in sex ratio between months, a Pearson's chi-square test with subsequent Fisher's exact test was conducted in R version 3.2.2.

To analyse the fecundity of round goby from the Kiel Canal we counted the eggs of 50 female specimens using a stereomicroscope. As round goby is a batch spawner, eggs in different developmental stages are present in the ovaries simultaneously. Therefore, eggs were divided into two fractions as done by TOMCZAK & SAPOTA (2006): the IR fraction with large eggs in a high development stage (diameter typically ≥ 1 mm) and the IIR fraction with smaller eggs in an earlier development stage. Regression analyses between absolute fecundity and both *TL* and *W_t* were performed in R version 3.2.2.

To determine the spawning period of round goby in the inspected area of the Kiel Canal the standard gonadosomatic index (*GSI*) of 348 mature individuals (183 females and 165 males, *TL* >60 mm) was calculated using the equation: *GSI* (%) = *B* / *W* × 100 where *B* was the gonad weight and *W* was the fish weight in gram (see ZEYLET AL. 2014). To determine possible differences in the *GSI* of females and males between the different inspected periods, a Kruskal-Wallis H test was conducted in R version 3.2.2. Subsequent post hoc tests using Bonferroni correction were taken from the package agricolae (MENDIBURU 2015). Due to the low number of available specimens in the first period (25th of April till 2nd of May), we combined *GSI* Values from 2011 and 2012.

Results

Diet composition

Crustaceans (mainly the corophiid *Corophium volutator* (Pallas, 1766), the gammarid *Gammarus tigrinus* Sexton, 1939 and the isopod *Cyathura carinata* (Krøyer, 1847)) had the highest relative importance (*RI*) for all three length classes (Figure 6.2). With increasing *TL*, the *RI* of crustaceans decreased, from 71.5% in small specimens <50 mm to 46.9% in large specimens of \geq 100 mm. In contrast, the percentage of annelids, the taxa with the second highest *RI*, increased from 9.6% in small

to 29.5% in large round gobies. The importance of chironomids decreased from small (14.6%) to large (1.9%) round gobies. Mussels and especially fish were found almost exclusively in medium-sized (2.9 and 1.7%, respectively) and in large round gobies (8.2 and 4.4%, respectively). Major differences in prey composition were estimated between the smallest and the other two length classes, whereas prey composition of medium-sized and large specimens was relatively similar. This was confirmed by cluster analysis and is shown in the corresponding dendrogram (Figure 6.3). While the dissimilarity between the prey compositions of the medium and large length classes was low (~13%), the small length class was separated at a considerably higher level (~38%).



Figure 6.2. Relative importance index (*RI*) of prey taxa (2011 and 2012) divided into three length classes (*TL*) of round goby. Numbers of non-empty guts examined are given above bars.



Figure 6.3. Cluster dendrogram showing the dissimilarity in prey composition (% of relative importance index) between the inspected length classes.

Analysis of similarities (ANOSIM) revealed that the diet composition of round goby in the Kiel Canal differed between the inspected years (r = 0.296, p < 0.01). Similarity percentages (SIMPER) suggested

that crustaceans were the most influential species (followed by chironomids, annelids and fish). Additionally, the diet varied seasonally from April/May to October (Figure 6.4). In 2011, the *RI* of crustaceans, the most important food taxon for round goby in our findings (mean 46.3%, range 27.2–68.1%, SD ±14.3%), was highest in May, relatively low from June to September and slightly increased again in October. In 2012, crustaceans had a high *RI* throughout the whole year (mean 61.5%, 54.0–69.6%, ±5.3%). In contrast to the *RI* of crustaceans, the taxon annelids showed its highest importance from June to September in 2011 (mean 26.3%, 10.7–38.6%, ±10.3%).



Figure 6.4. Relative importance index (*RI*) of prey taxa of round goby for the inspected months of the years 2011 and 2012. Numbers of non-empty guts examined are given above bars.

In 2012, annelids had a constant *RI* during all inspected months (mean 23.6%, 20.8–28.6%, ±2.8%). Chironomids were a relevant taxon throughout all months in 2011 (except for July), whereas in 2012 they were relevant only in April. Fish was present in the prey of round goby in July and August in 2011 (18.3 and 8.1%, respectively; mainly young-of-the-year Atlantic herring), whereas in 2012 it was present in all months except for April and July but only in low percentages. Fish eggs were only relevant in the prey in June 2012 (6.7%), where one specimen of round goby had eaten 240 gobiid eggs (probably *Pomatoschistus* spp.). As visible in the nMDS-plot (Figure 6.5), the prey composition of specimens caught in April 2012 seemed to be more similar to that in the different months of 2011 than to other months of 2012. This was mainly caused by the high proportion of chironomids.



Figure 6.5. NMDS-plot showing the dissimilarity in prey composition (% of relative importance index) between the inspected months of the years 2011 (grey) and 2012 (black) (stress = 5.0%).

Growth

The round goby from the Kiel Canal showed positive allometric growth characteristics (Figure 6.6), i.e. specimens grow faster in weight than in length during their lifetime.



Figure 6.6. Length-weight relationship of the studied round gobies from the Kiel Canal (n = 133, $r^2 = 0.9883$, years 2011 and 2012).

Calculated by the von Bertalanffy growth equation, round goby reached an asymptotic length (*TL*) of 133 mm (Figure 6.7). Growth was fastest in the first two years of life and much slower afterwards.



Figure 6.7. Length-growth curve estimated for round goby from the Kiel Canal (*n* = 133, years 2011 and 2012).

Sex ratio

The overall sex ratio (females to males) for *N. melanostomus* specimens caught during demersal trawling was 1.3:1. In June, the percentage of females was highest with ~69%, while afterwards it decreased down to ~45% in September. In October, the percentage of females was almost equivalent to the overall mean value with ~55% (Figure 6.8). The differences in sex ratio between the sampled months were significant (Pearson's chi-square test, χ^2 (4, *n* = 3,049) = 121.46, *p* <0.001). The results of the post-hoc Fisher's exact test are shown in Figure 6.8 as alphabetic characters.



Figure 6.8. Sex ratio of round goby caught while trawling in the Kiel Canal (years 2011 and 2012). Numbers of examined specimens are given above bars. Alphabetic characters (abc) indicate differences between inspected months.

Fecundity

We calculated absolute and relative fecundity for 50 specimens of round goby. Absolute fecundity was in the range from 457 (specimen of 76 mm *TL*) to 3,203 eggs (specimen of 145 mm *TL*). Relative fecundity (eggs per gram of female W_t) was in the range from 45 to 118 (mean 80 ±13). The relationship

between absolute fecundity and *TL* was expressed by a power function (Figure 6.9), like also the relationships between IR and *TL* (large eggs in a high stage of development) and IIR and *TL* (small eggs in a lower stage of development). All relationships between fecundity and *TL* were highly significant (both egg fractions: $r^2 = 0.8813$, p < 0.001, IIR egg fraction: $r^2 = 0.8107$, p < 0.001, IR egg fraction: $r^2 = 0.8209$, p < 0.001).





A linear equation was used to express the relationship between fecundity and W_t (Figure 6.10). All relationships between fecundity and W_t were highly significant (both egg fractions: $r^2 = 0.8793$, p < 0.001, IIR egg fraction: $r^2 = 0.8487$, p < 0.001, IR egg fraction: $r^2 = 0.7891$, p < 0.001).



Figure 6.10. Relationship between total weight and absolute fecundity (F) of females.

Spawning period

In order to determine the spawning activity of round goby in the Kiel Canal we inspected the standard gonadosomatic index (*GSI*). It was low in the first inspected period (25^{th} of April to 2^{nd} of May), especially for females. In the second period (1^{st} to 11^{th} of June), it was highest with values of 8.13 ±5.44 for females and 1.46 ±2.23 for males (Figure 6.11). After this peak in *GSI*, it decreased slowly until it reached the lowest values in the last inspected period (2^{nd} of September to 11^{th} of October). Differences in the *GSI* of females between the inspected periods were highly significant (Kruskal-Wallis H test, *H* (4) = 94.283, *p* <0.001). In males, no significant difference was detected between the inspected periods (*H* (4) = 9.8732, *p* >0.05).



Figure 6.11. Development of the standard gonadosomatic index (*GSI*). Females white and males grey boxes. Numbers of examined specimens given below boxes. Alphabetic characters (abc) indicate differences between inspected periods.

Discussion

Diet composition

Regarding the results of our analyses, a diet shift seems to occur between the length classes small (<50 mm) and medium (50–99 mm). While smaller individuals consume high percentages of small-sized chironomids and crustaceans, larger individuals incorporate high percentages of relatively large annelids and crustaceans into their diets. In comparison to other studies on the diet of *N. melanostomus,* this diet shift is less distinct in our findings and occurs in smaller individuals. This can be explained by the fact that mussels are a less important part of the diet in the Kiel Canal than in many other systems. In the Lower Rhine, molluscs (mainly invasive *Dreissena* sp. and *Corbicula* sp.) were the most important food taxon for round goby \geq 125 mm *TL*, while for smaller individuals they were not relevant (BORCHERDING ET AL. 2013). BARTON ET AL. (2005) found a shift to high percentages of dreissenid mussels in large-sized round goby (*TL* <100 mm) in the eastern basin of Lake Erie, Great Lakes region. The same was assumed from laboratory experiments inspecting round goby from the Detroit River, Great Lakes region (RAY & CORKUM 1997). If mussels were consumed by large round gobies in the Kiel Canal, it was mainly the sand gaper *Mya arenaria* Linnaeus, 1758 with shell lengths of up to 19 mm. Maybe due to the brackish conditions (see ORLOVA ET AL. 2004), dreissenid mussels are almost not existent at the study site and, in addition, soft-bodied prey seems available in relatively

high density. Thereby, the artificial ecosystem of the Kiel Canal differs strongly from freshwater ecosystems invaded by round goby, where dreissenids can often dominate the macroinvertebrate community (e.g. BARTON ET AL. 2005).

Low percentages of fish prey seem to be common for *N. melanostomus* as many studies are available from introduced populations in Europe (e.g. AZOUR 2011, BORCHERDING ET AL. 2013, BRANDNER ET AL. 2013a) and North America (e.g. CARMAN ET AL. 2006, TARABORELLI ET AL. 2010) with similar results. Similarly, in the Kiel Canal, fish only contributed to a minor extent to the diet of medium-sized and large round goby. This was highly correlated with the seasonal occurrence of young-of-the-year Atlantic herring in the system. The small-sized herring are available in high quantities in summer and eaten by many fish species, e.g. European flounder, European eel *Anguilla anguilla* (Linnaeus, 1758), European perch *Perca fluviatilis* Linnaeus, 1758 and zander *Sander lucioperca* (Linnaeus, 1758) (HEMPEL ET AL. 2016, personal communications from fishermen). Higher percentages of fish in the diet of invasive gobiids were recorded especially for bighead goby (BORCHERDING ET AL. 2013, BRANDNER ET AL. 2013a).

The differences we found in the prey of round goby from the Kiel Canal between 2011 and 2012 may be explained by variability in the abundance of important prey species. The density of the isopod *C. carinata*, which represented a major part of all crustaceans consumed, e.g. was found to vary strongly between years (ÓLAFSSON & PERSSON 1986). *N. melanostomus* is described to have an opportunistic feeding behaviour (e.g. RABY ET AL. 2010, BORCHERDING ET AL. 2013). Therefore, fluctuations in the availability of prey species can directly influence the composition of the ingested food.

The food composition of round goby in the Kiel Canal indicates competition for food with other fish species. Especially small-sized native gobiid species of the genus *Pomatoschistus* are vulnerable to competition with *N. melanostomus* as they inhabit the shallows of the inspected area in high abundance and prey at least partially on the same taxa (KNÖRR 2013). Additionally, food competition with European flounder, European perch and several cyprinid species seems likely as these species consume similar prey organisms (see KAFEMANN 2000, LAPPALAINEN ET AL. 2001, KARLSON ET AL. 2007). It appears that due to the occurrence of *N. melanostomus* in high numbers (see HEMPEL ET AL. 2016) a strong effect on the ecosystem is inevitable.

Growth

With an asymptotic *TL* of 133 mm, round goby in the Kiel Canal reached a relatively low value for the species. This is comparable to growth patterns from pure freshwater habitats in the introduced range of *N. melanostomus*. For example in Lake Michigan, round goby reaches a *TL* of about 100 mm in its fourth year (HUO ET AL. 2014). CORKUM ET AL. (2004) described that dwarf forms of the species can occur

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in freshwater and slightly brackish water. In its native area, round goby was found to grow larger in marine conditions (Black Sea) than in rivers or lakes (VELKOV ET AL. 2014). Salinity is not the only parameter determining the growth potential of round goby. This becomes apparent in the population of the Bay of Gdańsk, where a salinity comparable to that of our study area exits (see ZAKOWSKI ET AL. 2014). In that ecosystem round goby grew much bigger, reaching the highest *TL* stated for that species (CORKUM ET AL. 2004). The availability of food resources and the length of the growth period seem to play important roles for reaching large sizes (SOKOŁOWSKA & FEY 2011). The growth period was comparable between the Bay of Gdańsk and our study area. In the case of the Kiel Canal growth of round goby was possibly inhibited by high abundance (see HEMPEL ET AL. 2016) and consequential intraspecific competition for food resources.

Sex ratio

Sex ratio of round goby in our study was almost balanced with 1.3:1 (females to males). It varied throughout the inspected period, which was probably caused by the spawning behaviour of the species. In June and July the proportion of males could have been underestimated when many of them were guarding nests in the stone packaging of bankside areas. This corresponds well to our findings from the determination of the spawning period. Typically, introduced populations of *N. melanostomus* are strongly male biased while in stable native populations, the sex ratio is nearly 1:1 (e.g. CORKUM ET AL. 2004, TOMCZAK & SAPOTA 2006). The balanced sex ratio of the population in the Kiel Canal could indicate its established status.

Fecundity

With a relative fecundity of 80 ±13 eggs per gram of female W_t , round goby from the Kiel Canal showed a high value for the species in its introduced range. In the Bay of Gdańsk, a relative fecundity of 34 ±24 was calculated by TOMCZAK & SAPOTA (2006). In the upper Detroit River a hypothetical specimen with the standard length (*SL*) of 80 mm was calculated to have an absolute fecundity of 384 eggs (MACINNIS & CORKUM 2000) which was also considerably lower than for specimens of the same length in the Kiel Canal. Differences between fecundity in the Bay of Gdańsk and the Kiel Canal might be the expression of different reproduction strategies. Individuals from the Kiel Canal are smaller in size but on average older at a given length so that they may be able to produce a relatively higher number of eggs. The specimens inspected in the case of the Detroit River are possibly too small (only two specimens >75 mm *SL*) to produce high numbers of eggs. However, for the native distribution area of *N. melanostomus*, MILLER (1986) reports a fecundity of 328–5,221 at a *TL* of 70–130 mm, which is considerably higher than in our findings despite a high maximum length.

Spawning period

From our data it is apparent that the highest spawning activity of *N. melanostomus* in the Kiel Canal takes place between the beginning of May and the end of June. Afterwards, specimens with a high *GSI* are found only sporadically. The assumed period was comparable to that in the Romanian part of the native distribution area. In the area of Varna, Bulgaria, the reproductive season was reported to last from April till the end of September and in the Sea of Azov from April till July (MILLER 1986). Thus, the temporal limitation of the species spawning period differs strongly in the native area. Regarding introduced populations of round goby, the spawning behaviour in the Kiel Canal is comparable with the findings by MACINNIS & CORKUM (2000) for the Detroit River, where a strong decrease in the *GSI* of females was observed after the beginning of July. In the Lower Rhine, spawning mainly took place from April to July (GERTZEN ET AL. 2016). There, the spawning season varies strongly between the inspected years depending on water temperature and river discharge. The differences in the spawning season between ecosystems, regions and inspected years highlight once more the high adaptability of round goby.

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Contribution of authors to Chapter 6

Mattias Hempel collected the examined material of round goby in cooperation with co-author Rüdiger Neukamm, conducted the laboratory work for diet analysis and age determination and made all calculations. He made the map, prepared all Figures (except 6.5 and 6.11) and wrote the initial draft of the manuscript. Victoria Magath conducted the statistics and prepared Figures 6.5 and 6.11. Ralf Thiel assisted in the laboratory work, the calculations and the writing process. 7 Effects of introduced round goby (*Neogobius melanostomus*) on diet composition and growth of zander (*Sander lucioperca*), a main predator in European brackish waters

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Abstract

We inspected diet and growth of zander Sander lucioperca, a main predator in North European brackish waters, from the Kiel Canal, northern Germany, to analyse effects of the introduced round goby Neogobius melanostomus as possible prey. Round goby represented the most important fish prey taxon in the zander diet in 2011–2013 with 21.8% of relative importance index (RI) over all inspected specimens. In particular zander of total length ≥200 mm fed heavily on round goby, at least in the warmer quarters (2nd and 3rd) of the years under study. *RI* for these quarters was ranging from 10.4% to 52.1% for predator specimens of 200–399 mm and from 45.6% to 79.7% of *RI* for specimens of \geq 400 mm. In the colder quarters of the inspected years (1st and 4th), zander ≥400 mm showed a high amount of cannibalism, with R1 of young-of-the-year zander as prey ranging from 23.9% to 38.9%. The asymptotic total length (L_{∞}) of the zander, calculated by the von Bertalanffy growth equation, was 937 mm. In comparison with data from 1995/1996, before the round goby invasion in 2006, zander from our study showed considerable differences in diet composition. RI of the prey organisms herring and Pomatoschistus spp. decreased from 26% to 13.4% and 14.2% to 4.4%, respectively. RI of young-of-theyear zander as prey for larger zander increased from 1.2% to 7.7% and round goby occurred as newly established prey taxon. Zander also showed faster growth and a better condition factor. We assume that this development has the potential to enhance the situation of the local fisheries that are strongly related to the catch of zander. In addition, abundance control of round goby seems to result from the high predator abundance.

Key words: Gobiidae, invasive species, diet shift, predator-prey relationships, Kiel Canal

Introduction

The zander *Sander lucioperca* (Linnaeus, 1758) is an important top predator (in terms of ecosystems and fisheries) of many brackish water ecosystems in northern Europe (LEHTONEN ET AL. 1996, GRÖGER ET AL. 2007). Total catch from European inland fisheries reported to FAO was 17,892 t in 1999 (FAO 2014). The non-indigenous round goby *Neogobius melanostomus* (Pallas, 1814) has been introduced into many ecosystems in the northern hemisphere and continues to spread (KORNIS ET AL. 2012, KALCHHAUSER ET AL. 2013). It is assumed that round goby became established in the eastern parts of the Kiel Canal in 2006 (NEUKAMM 2009a). It has been abundant in the studied middle region of the Canal since 2009 (see result section).

The establishment of the non-indigenous goby is described as having strong negative impacts on the invaded ecosystems and its inhabitants. KORNIS ET AL. (2012) summarize that round goby competes with native species by resource competition, spawning interference and displacement to sub-optimal habitats. In addition, the round goby seems to have a strong predation impact on invertebrate communities (e.g. KUHNS & BERG 1999, LEDERER ET AL. 2008).

In contrast to the above mentioned impacts on native fish species of similar trophic level and invertebrates, non-indigenous round goby provide an important food resource for predatory fishes (e.g. STEINHART ET AL. 2004b, ALMQVIST ET AL. 2010, REYJOL ET AL. 2010, TARABORELLI ET AL. 2010, POTHOVEN & MADENJIAN 2013, RAKAUSKAS ET AL. 2013). The newly available food can enhance growth and have positive effects e.g. on survival and reproduction. The effect of incorporating *N. melanostomus* in diets seems to differ strongly between species and locations (see CRANE ET AL. 2015). Predator species in North America from the same genus as the studied zander are found to consume high proportions of round goby (REYJOL ET AL. 2010, CRANE ET AL. 2015). There are also descriptions of important changes in prey composition following round goby invasion for fish eating birds and snakes (e.g. JAKUBAS 2004, KING ET AL. 2008).

Round goby is not only a potential prey for predatory fish species but can also act as a predator on fish eggs and larvae (e.g. STEINHART ET AL. 2004a). KORNIS ET AL. (2012) gave multiple examples for species suffering from egg depredation and concluded that benthic eggs of any species were probably vulnerable to predation by round gobies.

Although many predators of round goby have been identified, no study has examined the effects of round goby on zander diet and growth. To fill this gap in knowledge, we investigated these aspects in the framework of a case study in the brackish Kiel Canal. Supported by reports of local fishermen, we hypothesized that the zander in the Kiel Canal showed faster growth due to predation on round goby than before the round goby invasion. To test this hypothesis, we compared

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the results of our current research from the years 2011–2013 with data from 1995/1996 (KAFEMANN & THIEL 1998, KAFEMANN 2000). These analyses were complemented by the inspection of the development of biomasses of zander, round goby and other dominant fish species in the study area in 2008–2013.

Materials and methods

Study site

The Kiel Canal in northern Germany is the most commonly used artificial waterway worldwide (HEITMANN ET AL. 2013). In 2012, about 35,000 vessels transporting 104 million tonnes of cargo passed the Kiel Canal (WSD NORD 2015). It was built in the late 19th century and connects the North Sea with the western Baltic Sea over a length of about 100 km.





Maximum depth of the Kiel Canal is 11 m and width varies between 162 m and 240 m (KAFEMANN ET AL. 2000). The shoreline of the Kiel Canal is mostly lined with stone packaging to avoid damage due to shipping traffic. The stone packaging represents a common way to ensure stability of shorelines, not only in artificial waters but also in large streams and lakes. The canal shows a salinity gradient, caused by water exchange with the Elbe River at the western end and with the

Baltic Sea at the eastern end. Throughout the investigated period (2011–2013), mean salinity near the western end of the canal was 2.7 and 10.4 near the eastern end. The study site (54°19'19" N, 9°42'49" E) was situated close to the city of Rendsburg (Figure 7.1), where mean salinity was ~7. The fish fauna of the Kiel Canal comprises numerous limnic, euryhaline and marine taxa. In 1995/1996, 46 fish species were recorded in the Kiel Canal by KAFEMANN (2000), whereby abundance of some fish species in the canal was highly correlated with the seasons.

Sampling

Sampling was performed in collaboration with a resident commercial fishery. The net (opening width 14 m, 10 mm mesh size in the cod end) was trawled along the bottom with a speed of ~2.5 knots, mostly in areas with depth >10 m (verified by echo sounding). Bottom material in this depth was characterised mostly by sand and mud (verified by bottom grab sampling). In 2011 and 2012, monthly sampling from June to October took place with 10 km trawling distance in total (5 hauls, each 1–3 km in length) in the framework of a monitoring program on European eel *Anguilla anguilla* (Linnaeus, 1758). That program has taken place annually since 2007 (see NEUKAMM 2009b). Biomass data from the years 2008–2010, later presented in the results section, have been assessed within the above mentioned monitoring program as well. Zander specimens for the months November to March of the years 2011–2013 were provided by fishermen for the same area. The specimens were caught using the same fishing technique and the same net. Thus, an influence of different sampling techniques on the food composition in the digestive tract can be ruled out. Fish were frozen and kept at -20°C until analysis. In large specimens with total length (*TL*) >350 mm, the digestive tracts and complete heads were frozen after determining weight (0.1 g precision) and *TL* (to the nearest mm) of the whole individuals.

Biomass calculation

Biomass (*B*) of zander, round goby and other important fish species in the region under study was calculated as kg ha⁻¹ according to the equation: $B = B_T / A_T \times 10,000$, where B_T was the determined biomass of a particular species in the trawled area in kg and A_T was the size of the trawled area in m². For biomass calculations, only results from the trawling procedures from June to October of the years 2011 and 2012 were considered. That was because exact biomass was measured only in the monitoring program described above and not during conventional fisheries.

Diet analysis

Digestive tracts of 504 zander were examined for prey consumed. Investigated fish were grouped into three length classes (*TL*: juveniles <200 mm ($Z_{<200}$), sub-adults 200–399 mm

 $(Z_{200-399})$ and adults \geq 400 mm ($Z_{\geq400}$) and into quarters of the years (1 = January–March, 2 = April– June, 3 = July–September, 4 = October–December). Used length classes were chosen due to an assumed diet shift with dominance of a piscivorous feeding behaviour from the second year of life (see LEHTONEN ET AL. 1996). In our study a *TL* of about 200 mm was reached by the zander at the beginning of their second year (see results section). To determine if adult zander possibly preyed on different taxa than juveniles and sub-adults, we included the third length class.

Digestive tracts were defrosted and prey items (from oesophagus and stomach) were determined to the lowest taxon possible (generally to species level, except for few prey items with high rate of digestion). Prey items in different stages of digestion were counted and their relevant structures (bones or body parts; e.g. otoliths, dentary bones, vertebrae, maxillary bones) were measured for subsequent back calculation of ingested biomass.

Back calculation of biomass was performed using published regression equations between length of structures and total fresh weight of the species (e.g. HÄRKÖNEN 1986, DEBUS & WINKLER 1996, DIETRICH ET AL. 2006, TARKAN ET AL. 2007, VERREYCKEN ET AL. 2011). In cases where no published data were available, we calculated regression equations of prey items based on fresh material from our own trawl net catches. In this regard, we used: $W_t = 0.000005 \times TL^{3.2127}$ (specimens from trawling at the study site 2011: n = 2,170, range 41–172 mm (*TL*), $r^2 = 0.9766$, p < 0.001) to calculate round goby total weight (W_t) in g from *TL* in mm (measured directly in intact prey or calculated from the length of round goby bones). To calculate *TL* of young-of-theyear zander in mm from the maximal length of its dentary bone (*LD*) in mm we used: *TL* = 9.6284 × *LD* + 7.3369 (specimens from trawling at the study site 2012: n = 13, range 117–174 mm (*TL*), $r^2 = 0.9696$, p < 0.001).

The relative importance index (*RI*) of prey taxa was calculated following GEORGE & HADLEY (1979): RI_i (%) = ($N_i + B_i + F_i / \sum (N_i B_i F_i)$) × 100, where N_i (%) is the number, B_i (%) is the biomass and F_i (%) is the frequency of occurrence of the prey component *i* in the sample.

One-way ANOVA was performed, using SPSS version 20 (SPSS Inc., Chicago, Illinois) software, to analyse differences in round goby *RI* between the three zander size classes, as well as between the four yearly quarters. Fishers LSD post-hoc tests were conducted to assess pairwise differences, where needed. Cluster analysis was performed using PRIMER version 6 (Primer-E Ltd., Plymouth, UK) to compare the dietary composition of zander from our study with that of zander from 1995/1996 inspected by KAFEMANN (2000). Therefore the *RI* of different food taxa was used for each year. Prior to analyses, the raw data were fourth root transformed in order to minimize any effect due to oversized replicates. A dendrogram was used to visualize the percentage of Bray-Curtis similarity between samples, calculated by PRIMER 6. This takes into account both the number of shared taxa between years and their *RI* to calculate the percent similarity between pairs of samples.

Growth estimation

Age determination of 961 zander (*TL* 65–820 mm) was performed using both sagittae otoliths and opercular bones for all specimens. Scales were inspected as well, but annuli were often difficult to read. Otoliths and opercular bones were removed from the defrosted fish or fish heads and cleaned of adherent tissue with water. The material was stored dry until age determination took place, where it was handled under magnification with reflected light in a black dish filled with alcohol (Figure 7.2) following the procedure of BOSTANCI (2008).



Figure 7.2. Otoliths of zander: A. age 1+, B. age 2+, C. age 3+ and D. age 4+. Arrows show the location of annuli. Photomicrograph by M. Hempel.

As the initial annulus was difficult to detect in some older individuals (see BUCKMEIER ET AL. 2002), we compared otoliths and opercular bones from young-of-the-year zander from the time of first annulus formation. Fish that showed unclear annuli or different age estimates in otoliths and opercular bones (14 specimens) were excluded from the analysis. Table 7.1 shows the number of specimens used for age determination per length class.

Γable 7.1. Number of age determinations f	or zander from the Kiel (Canal per length classes (TL)
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Length class (mm)	Number
0-100	43
>100-200	494
>200-300	156
>300-400	113
>400-500	103
>500-600	45
>600-700	4
>700-800	2
>800–900	1
All sizes	961

To define an exact age (days since hatching), we used 1st of May as a hypothetical date of birth for the zander (see KAFEMANN & THIEL 1998). This is a common time of hatching for zander in the inspected geographical region. To ensure comparability of this study with the data from previous investigations before round goby occurred in the Kiel Canal, we excluded two fish older than eight years from growth analysis as done by KAFEMANN & THIEL (1998).

The relationship between W_t and TL was characterized by the length-weight equation: $W_t = a \times TL^b$, where the parameter a is a coefficient for the weight at length and b is a shape parameter for the body form. Growth in length was calculated by the von Bertalanffy growth equation using FISAT II version 1.2.2 (FAO, Rome, Italy): $L_t = L_{\infty} \times (1 - e^{-k(t-t_0)})$, where L_t is the length at time t (TL, mm), L_{∞} is the asymptotic length (TL, mm), k is a growth coefficient and t_o is the prenatal age (years). The 95% confidence interval for L_{∞} was calculated from the standard error given by the FISAT II output, in order to compare growth of zander from the Kiel Canal 1995/1996 and the tidal Elbe River 1992/1993 (KAFEMANN & THIEL 1998) with those of zander in our study. Additionally, the mean total length for zander at an age of 1–6 years (1995/1996 and 2011–2013 Kiel Canal), calculated by the von Bertalanffy growth equation, was compared using the Wilcoxon signed-rank test in SPSS. Furthermore, the index of growth performance (P), after PAULY (1979), was calculated: $P = \log k \times W_{\infty}$, where k is a growth coefficient (von Bertalanffy growth equation) and W_{∞} is the asymptotic weight in g calculated by the length-weight equation and the asymptotic length (L_{∞}). To compare condition factor (K), we calculated it for zander of $TL \ge 200$ mm as done by KAFEMANN & THIEL (1998): $K = (W_t / TL^3) \times 100$, where TL was used in cm.

Results

Trends in biomass

From 2008 to 2012, we found similar trends in the biomasses of round goby and zander in catches from trawling at the study site (Figure 7.3). After the first appearance of round goby in 2008, its biomass increased rapidly to a maximum of 1.7 \pm 1.8 kg ha⁻¹ (mean \pm SD) in 2011. In 2012, round goby catches decreased to 0.6 \pm 0.4 kg ha⁻¹. Biomass of zander increased to a maximum of 8.1 \pm 8.7 kg ha⁻¹ in 2011 and decreased to 5.8 \pm 3.8 kg ha⁻¹ in 2012. Biomass of other fish species in the trawl net catches developed at least slightly differently. The high biomass of bream *Abramis brama* (Linnaeus, 1758) in trawl net catches from the inspected area, seems to follow the same trend but showed an increase in 2012 contrasting with the decrease of zander and round goby. Biomass development of flounder *Platichthys flesus* (Linnaeus, 1758) was relatively similar to that of round goby but showed two maxima with ~2 kg ha⁻¹ in 2009 and 2011. Biomass of European eel was high in 2009 (~2 kg ha⁻¹), lowest in 2011 and increased again in 2012. Roach *Rutilus rutilus* (Linnaeus, 1758) occurred with relatively constant biomass of ~ 1 kg ha⁻¹ since 2010. Thus, the similar trends in biomasses of round goby and zander were not followed by all fish species inspected and possibly did not represent a general productivity pattern.



Figure 7.3. Average biomass of zander, round goby and of all other fish species, reaching a biomass of >1 kg ha⁻¹ in at least one year, in trawl net catches at the sampling site (years 2008–2012, June–October).

Diet composition

Differences in zander diet were detected between length classes and seasons (Figure 7.4). For juvenile zander ($Z_{<200}$), mysid shrimp *Neomysis integer* (Leach, 1814) had the highest relative importance (*RI*) throughout the studied period (62.7 ±31.2% (mean ±SD), range 13.6–100%). In the warmer 2nd and 3rd quarters of 2011 and 2012, when young-of-the-year herring *Clupea harengus* Linnaeus, 1758 was abundant at the study site (especially 2nd quarter of 2011), this species comprised considerable percentages of *RI* (15.5 ±22.6%, 0–64%). Other frequent prey taxa of Z_{<200} were decapod shrimp (5.6 ±8.2%, 0–22.4%), gobies of the genus *Pomatoschistus* (5.3 ±8.4%, 0–22.8%) and young-of-the-year round goby (4.6 ±6.6%, 0–16.4%). *RI* of round goby in the diet of Z_{<200} did not vary significantly between the quarters of the inspected years (ANOVA, *F* (3, 7) = 1.879, *p* = 0.274).

For sub-adult zander ($Z_{200-399}$), the *RI* of mysid shrimp was lower than for $Z_{<200}$ (31.1 ±13.6%, 8.4– 50.4%). In contrast, the *RI* of round goby as prey was considerably higher (27.8 ±17.6%, 9–52.1%), reaching values of about 50% in the 2nd and 3rd quarters of 2011. *RI* of young-of-the-year herring as prey reached values comparable with $Z_{<200}$ (11.8 ±12.9%, 0–37.4%). As for $Z_{<200}$, *RI* of round goby in the diet of $Z_{200-399}$ did not vary significantly between the quarters of the inspected years (*F* (3, 7) = 1.633, *p* = 0.316).





For adult zander ($Z_{\geq 400}$), mysid shrimp did not represent an important prey item (3.7 ±5.9%, 0– 17.5%). Major prey taxa of this size group were round goby (31.9 ±31.7%, 0–79.7%) and youngof-the-year zander (20.5 ±15.7%, 0–38.9%). Other frequent prey taxa of $Z_{\geq 400}$ were decaped shrimp (15.2 ±10.8%, 0–33.8%) and herring (11.4 ±7.1%, 0–19.8%). Diet of $Z_{\geq 400}$ strongly depended on the season, with high importance of round goby only in the 2nd and the 3rd quarters of 2011 and 2012. Accordingly, we found a significant difference in *RI* of round goby as prey among the inspected quarters (*F* (3, 7) = 49.901, *p* = 0.001). In the 2nd and the 3rd quarter, *RI* of round goby in the diet was significantly higher than in the 1st and the 4th quarter (Fishers LSD post-hoc test, p = 0.001 and p = 0.003, respectively). Additionally, it was significantly higher in the 2nd quarter than in the 3rd quarter (*p* = 0.022). No difference was detected between the 1st and the 4th quarter (*p* = 0.831). Cannibalism (predation on young-of-the-year individuals of zander) was most crucial in the 1st and 4th quarters of the studied years.

The *RI* of round goby in the prey was different between the three size classes (*F* (2, 23) = 3.552, p = 0.001). It was significantly higher for $Z_{\geq 400}$ than for $Z_{<200}$ (Fishers LSD post-hoc test, p = 0.018). No significant difference was found between $Z_{200-399}$ and $Z_{<200}$ (p = 0.075) and $Z_{200-399}$ and $Z_{\geq 400}$ (p = 0.489). Regarding the number *N* (%), biomass *B* (%) and frequency of occurrence *F* (%) of prey taxa (supplementary Table 7.S1), it is most notable that round goby comprised high percentages of ingested biomass for $Z_{200-399}$ (52.8 ±28.5%, 19.3–90.1%) and $Z_{\geq 400}$ (42.4 ±44.2, 0–99.2%). In the warmer 2nd and 3rd quarters of both years, it composed 24.6–90.1% and 52.3–99.2% of ingested biomass of $Z_{200-399}$ and $Z_{\geq 400}$, respectively. In the colder 1st and 4th quarters, percentages of round goby biomass in the diet was much lower (16.2–42.9% for $Z_{200-399}$ and 0–8.8% for $Z_{\geq 400}$).

Cluster analysis was carried out to show similarities in prey composition of zander between the years 1995/1996 and the two years of our study (2/2011–1/2012 and 2/2012–1/2013). The related dendrogram (Figure 7.5) shows relatively high similarities in the prey composition of all considered years. However, the years 1995 and 1996 on the one side and the 1st and 2nd years of our study on the other side formed two clearly differentiable clusters separating at 78.1% of Bray-Curtis similarity.



Figure 7.5. Dendrogram showing the similarity in prey composition (% of relative importance index) between years before round goby invasion (1995/1996) and years of our study (exact sample period 2011: 2/2011–1/2012 and 2012: 2/2012–1/2013), when round goby occurred in the system. Fourth-root transformed data.

Growth patterns

Calculated by the von Bertalanffy growth equation, zander reached an asymptotic length (*TL*) of 937 mm (Appendix 7.1). In the first autumn, some of the investigated young-of-the-year zander reached more than 200 mm *TL*. After four years of growth, the mean calculated *TL* was 531 mm. Zander from the Kiel Canal showed an isometric growth characteristic (Appendix 7.2). Length and weight proportions remained the same during their lifetime. Mean condition factor (*K*) for zander of *TL* ≥200 mm was 0.81 ±0.07. The index of growth performance (*P*) for all investigated fish was 3.13.

In comparison with zander from other typical brackish water bodies in northern Germany (KAFEMANN & THIEL 1998), where the asymptotic length (L_{∞}) ranged from 792–987 mm (Table 7.2), specimens of our study showed a high value.

Table 7.2. Parameters calculated by the von Bertalanffy growth equation (t_0 , k, L_{∞} and 95% confidence interval, W_{∞}) and index of growth performance (P) for zander from the Kiel Canal 2011–2013 (this study), the Kiel Canal 1995/1996 (KAFEMANN & THIEL 1998) and other brackish water habitats in northern Germany (KAFEMANN & THIEL 1998), n.i. = parameter not indicated.

Parameter	t ₀ (years)	k	<i>L</i> ∞ (mm)	<i>L</i> ∞ (95% CI)	<i>W</i> ∞ (g)	$1/k + t_0$	Ρ
Kiel Canal 2011–2013	-0.40	0.19	937	845-1.029	7085	4.86	3.13
Kiel Canal 1995/1996	-0.36	0.22	685	615–754	2550	4.19	2.75
Tidal Elbe River 1992/1993	-0.88	0.17	987	690–1.285	8942	4.99	3.18
Greifswalder Bodden	0.21	0.27	831	n.i.	4011	3.90	2.93
Schlei	0.08	0.22	792	n.i.	4351	4.63	2.98

There is also an obvious difference between the L_{∞} calculated for the Kiel Canal for the years 1995 and 1996 (L_{∞} = 685 mm, 95% confidence interval = 615–754 mm, KAFEMANN & THIEL 1998) and the higher L_{∞} of our study (L_{∞} = 937 mm, 845–1.029 mm, 2011–2013).

Additionally, the mean total length at the age of 1–6 years (Table 7.3) was significantly higher in our study than in 1995/1996 (Wilcoxon signed-rank test, Z = 21.000, p = 0.028). The estimated asymptotic length for zander from the Kiel Canal from 2011–2013 is also higher than the calculated values for the Greifswalder Bodden and the Schlei, which both represent large brackish waters adjacent to the Baltic Sea with relatively low depths. Growth parameters for zander in our study are close to the parameters calculated for zander from the highly productive tidal Elbe River in 1992/1993 (KAFEMANN & THIEL 1998). In our study, zander realized two thirds of their growth capacity (1 / $k + t_o$) after 4.86 years (Table 7.2). This growth capacity (see HOHENDORF 1966) is again comparable to the value for the tidal Elbe River in 1992/1993 (4.99 years). The index of growth performance (P) after PAULY (1979) has the same characteristic with 3.13 (Kiel Canal) and 3.18 (Elbe River). A higher Fulton's condition factor of the specimens in our study

(0.81 ±0.07) indicates a better constitution than in 1995/1996 (0.75 ±0.11), with nearly as high value as for the tidal Elbe River in 1992/1993 (0.84 ±0.12).

Ago (voors)	Mean total	length (mm)
Age (years)	Kiel Canal 1995/1996	Kiel Canal 2011–2013
1	141	219
2	266	343
3	353	446
4	424	531
5	474	601
6	524	659

Table 7.3. Mean total length at age (mm) calculated for the age of 1–6 years by the von Bertalanffy growth equation. Data of 1995/1996 by KAFEMANN & THIEL (1998).

Discussion

Trends and interrelations in biomasses of fish species

Due to the similar trends in the development of the biomass of zander and round goby, we hypothesize that the increase in zander biomass 2008–2011 could be related to the high availability of round goby. While the biomasses of both species were highest in 2011, they declined in 2012. The biomass of round goby dropped to about one third within one year whereas the biomass of zander slightly decreased (Figure 7.3). We hypothesize the decrease in round goby biomass to be caused by the high zander abundance.

Additionally, in 2013 and 2014 low round goby catches were common in the deeper parts of the Kiel Canal, whereas zander catches remained on a high level (own observations and personal communication fishermen). High abundance of round goby was still present in the shallow, rocky areas near to the shore (own observations and information by anglers).

According to KORNIS ET AL. (2012), there is some evidence that high predation by piscivorous fish can lead to a control of round goby abundance. As an example, a population of round goby in Lake Erie, USA, declined during 2004–2008 after burbot *Lota lota* (Linnaeus, 1758) had started to use it as a major prey organism in 2003 (MADENJIAN ET AL. 2011). We hypothesize this effect of population control to take place in the Kiel Canal as well. Predation of round goby by zander may contribute to long term stabilization of the round goby population in the Kiel Canal at relatively low abundance. Long-term monitoring will show if a permanent relationship really exists between the biomasses of round goby and zander as its predator.

However, our data from trawling can only reflect trends in biomass in the deeper water area away from the shore. In the shorelines of the Kiel Canal, covered by stone packaging, the biomass of round goby is supposed to be higher than in the sandy/muddy deeper areas (see RAY & CORKUM 2001). In addition, we suppose that the trawl net used may have caused different catchabilities for zander and round goby, e.g. due to differences in escape behaviour of both species. Overall, we hypothesize that the real biomass of round goby was much higher than determined.

Diet composition and shift

The importance of round goby as prey, identified in our study, is the highest reported for the species zander. In the Curonian Lagoon, a large, shallow freshwater basin connected to the south-eastern Baltic Sea, RAKAUSKAS ET AL. (2013) calculated by stable isotope analyses that round goby made up 17% (by biomass) of zander diet (August–September 2010, specimens of *TL* 283 ±54 mm). In contrast, during the same season (3^{rd} quarters of 2011 and 2012), the zander diet from individuals of a comparable length ($Z_{200-399}$) consisted of 82.8 and 24.6% round goby (by biomass) in our study (supplementary Table 7.S1).

Before round goby established in the Kiel Canal, KAFEMANN (2000) analysed the diet of small to large zander in 1995/1996. In that study, mysid shrimp had the highest relative importance (*RI*, 32.2 \pm 1.4%) comparable to that in our study (31.4 \pm 9.6%) (Figure 7.6). Herring had the second highest *RI* in 1995/1996 with 26 \pm 8%, while in our study it was only half that value (13.4 \pm 5.8%). Small gobies (*Pomatoschistus* spp.) had a *RI* of 14.2 \pm 11.6% in 1995/1996, which was considerable higher than in our study (4.4 \pm 1.6%). *RI* of European smelt *Osmerus eperlanus* (Linnaeus, 1758) decreased from 6.7 \pm 5.6% in 1995/1996 to 2.9 \pm 0.1% in 2011–2013. Important prey items in our study were young-of-the-year zander with 7.7 \pm 3.3% of *RI* in comparison to 1.2 \pm 1.7% in 1995/1996.

As in our study, the consumption of prey species in 1995/1996 was strongly related to their seasonal availability. KAFEMANN (2000) described that zander suffered from the absence of adequate prey during summer when it fed mainly on small native gobies (*Pomatoschistus* spp.). These small gobiid species reach a maximum *TL* of only 95 mm (MILLER 1986), while round goby reaches a maximum *TL* of about 250 mm (SAPOTA 2006). In our study, mean fresh weight calculated for round goby found in the stomachs of inspected zander specimens (6.36 ±7.92 g) was about ten times higher than for *Pomatoschistus* spp. (0.57 ±0.46 g). Round goby, which was not existent in the prey composition in 1995/1996, accomplished 21.8 ±3.2% of *RI* throughout all length classes of zander and all seasons within our study.



Figure 7.6. Relative importance index (*RI*) of prey taxa for zander in 1995/1996 and in the 1st (2/2011–1/2012) and 2nd (2/2012–1/2013) years of our study. The category "other gobies" includes sand goby and common goby. Numbers of examined specimens are given above bars.

The comparison of the diet of zander analysed in 1995/1996 with that of our study is limited due to different sizes of the study sites. Zander was taken along the entire Kiel Canal (~100 km) in 1995/1996, while our fish were taken from a smaller section (~6.5 km). Additionally, adult zander ($Z_{\geq 400}$) represented a smaller percentage of examined specimens in 1995/1996 (n ($Z_{<200}$) = 508, n ($Z_{200-399}$) = 512, n ($Z_{\geq 400}$) = 205) than in 2011–2013 ($Z_{<200}$ = 150, $Z_{200-399}$ = 193, $Z_{\geq 400}$ = 161). Nevertheless, the establishment of round goby clearly resulted in a change of zander prey composition.

Similar to the results of our study on the zander in the Kiel Canal, round goby invasion resulted in important diet shifts in numerous other predatory species in invaded ecosystems. For instance in the Baltic Sea area, important predators include the fish species cod *Gadus morhua* Linnaeus, 1758 and perch *Perca fluviatilis* Linnaeus, 1758 and the bird species great cormorant *Phalacrocorax carbo* (Linnaeus, 1758) and grey heron *Ardea cinerea* Linnaeus, 1758 (JAKUBAS 2004, ALMQVIST ET AL. 2010, RAKAUSKAS ET AL. 2013). In Lake St. Pierre, St. Lawrence River, North America, REYJOL ET AL. (2010) found round goby as a prey item in 64.5% of non-empty stomachs of sauger *Sander canadensis* (Griffith & Smith, 1834), a species closely related to the zander.

The high seasonal variability in the prey composition of zander in the Kiel Canal might be a result of different habitat occupancy of zander and round goby. It seems that in the Kiel Canal, in contrast to that described for the Baltic Sea and the Laurentian Great Lakes (see CORKUM ET AL. 2004), round goby occupies the stone packaging throughout the year, with part of the population migrating into the deeper, muddy and sandy areas for the summer.

The water temperature is almost stable throughout the entire water column due to ship introduced turbulences. A higher energy requirement in summer may force a part of the population away from the stone packaging to feed. The described migration pattern was supported by data from trawling (commercial and monitoring program), showing an increase in round goby abundance at the bottom in deeper parts of the channel in spring which seems to be related to the increasing water temperature. In contrast to round goby, zander concentrate in the deeper areas throughout the year (personal communication fishermen). Therefore, while there is low potential for zander encountering round goby in winter, the round goby summer bottom-migration exposes them to the predator.

Another reason could be a high availability of other prey species in winter. Spring migration of herring, into the Kiel Canal, normally results in higher percentages of this species in the diet and additionally young-of-the-year zander are probably easy to capture when using the same areas as adult zander for wintering. In terms of energy density, with all likelihood, round goby do not generally provide a preferable food item. RUETZ ET AL. (2009) report the average energy content of round goby in the Muskegon Lake region, North America, to be 4.326 J per g of wet mass. This is comparable to that of *Pomatoschistus* spp. and herring from the German Wadden Sea (4.62 and 4.54 J per g, respectively) and smelt from the Elbe River, Germany (4.16 J per g) found by TEMMING & HERRMANN (2003). However, JAKUBAS (2004) reports that the calorific value of round goby in the Gulf of Gdańsk can reach 6.3 J per g of wet mass during its spawning period. This can make the round goby a preferable food in this time of the year. Our observations indicated that the spawning period of round goby almost coincided with the warmer quarters in which zander consumed high amounts of this prey.

Due to the importance of round goby in the diet of zander, the predatory pressure on other previously more intensively used prey species may decline. In the Kiel Canal, this possibly especially affects native gobiids (*Pomatoschistus* spp.) which represented a major prey item in zander diet before the round goby invasion. We hypothesize that this can partially compensate for negative effects, such as competition for nesting sites and food. Otherwise, the assumed increased biomass of zander can reduce or even eliminate the effect of reduced predatory pressure.

Growth patterns

We suppose that better growth and condition factor of zander was caused by the occurrence of high round goby biomass in the Kiel Canal. The high availability of this prey, at least during

summer, seemed to be an important stimulator for growth of zander. This is clearly supported by the fact that, before the round goby invasion, zander suffered from the absence of adequate prey in summer while feeding mainly on small native gobies (*Pomatoschistus* spp.) (KAFEMANN 2000).

Fish growth in general is correlated with the availability of adequate food resources (see BOISCLAIR & LEGGETT 1989). For zander, a limitation of growth by food availability was described by THIEL (1989) for young-of-the-year and by WINKLER (1991) for older individuals. As round goby often appears in high abundance, it can represent a profitable food source.

The low asymptotic length (L_{∞}) of zander in the Kiel Canal before round goby invasion (Table 7.2) indicates the low productivity (for zander) of this ecosystem at that time (1995/1996). Therefore, the high profit from the establishment of round goby (L_{∞} almost one third higher in 2011–2013) is possible. Zander populations in other low-productive ecosystems such as the Greifswalder Bodden and the Schlei are more likely to profit from round goby establishment than those in high productive systems such as the tidal Elbe River.

Conclusions

As hypothesized, zander in the Kiel Canal profit from the appearance of round goby by showing better growth patterns. From our data, we can safely conclude that round goby represents a major food taxon for zander in the canal and that growth has improved since 1995/1996 when no round goby were available as food.

We assume that negative effects on the zander in the Kiel Canal caused by the round goby invasion are marginal. A strong competition for food of young-of-the-year zander and round goby in the Kiel Canal seems unlikely. This is supported by the fact that in diet analyses of specimens caught at the study site, we found only a small overlap in the diets of round goby and young-of-the-year zander (unpublished data). In the inspected region, mysid shrimps, that are a main food for young-of-the-year zander, only occurred in low percentages in the stomachs of *N. melanostomus* (<5% of relative importance). Predation of round goby on zander eggs is likely infrequent due to nest guarding behaviour of zander. In contrast to the closely related walleye *Sander vitreus* (Mitchill, 1818) in North America, which is affected by egg predation from round goby (ROSEMAN ET AL. 2006) and does not exhibit parental care or nest guarding (NATE ET AL. 2001), zander defends its nest against predators (WUNDSCH 1963).

Overall, the establishment of round goby may lead to higher yields of the commercially important zander for fisheries at the Kiel Canal. Nevertheless, round goby populations, biomass/

growth of zander and trawl catch results should be monitored in the long term. The assumed biological control effect on round goby by zander may result in low abundance of the invader, and with a reduced availability of round goby, zander growth could decrease in the future again. Improvements for zander (and other predators) from round goby invasion are likely a phenomenon of water bodies that were limited by some kind of resource availability before the invasion.

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Appendices and supplementary Table

2

3

4 Age (years)

Appendix 7.1. Length-growth curve estimated for zander from the Kiel Canal (*n* = 945, years 2011–2013).

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Appendix 7.2. Length-weight relationship of the studied zander from the Kiel Canal (*n* = 961, years 2011–2013).

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A. Gut contents of Z _{<200}																								
Quarter		2/2011			\$/2011		4	/2011		1	/2012		2	/2012		3/	2012		4/	2012		1	2013	
Examined specimens		4			61			10			0			14			25			23			13	
	Z	В	F	Z	В	F	N	В	F	N	В	F	Z	В	F	N	В	F	N	В	F	N	В	F
Bream	0	0	0	0	0	0	0	0	0				0	0	0	0	0	0	0	0	0	0	0	0
Herring	80	66.6	66.7	4.46	16.2	13.8	0	0	0				.63	1.78	8.33	7.28	37.3	1.7	2.6	25.5	10.5	0	0	0
Perch	0	0	0	0	0	0	0	0	0				0	0	0	0	0	0	0	0	0	0	0	0
Zander	0	0	0	0	0	0	0	0	0	ī	ī	ī	0	0	0	0	0	0	0	0	0	0	0	0
Other gobies	0	0	0	15.8	27.5	39.7	0	0	0	ī	ī	ī	0	0	0	1.08	9.32	13	3.9	3.25	15.8	0	0	0
Roach	0	0	0	0	0	0	0	0	0		ī		0	0	0	0	0	0	1.3	11.7	5.26	0	0	0
Round goby	0	0	0	8.42	23.6	27.6	0	0	0				0	0	0	.81	3.18	8.7	5.49	16.2	10.5	0	0	0
Smelt	0	0	0	1.98	23.3	5.17	0	0	0		ī		0	0	0	0	0	0	1.3	20	5.26	0	0	0
Other fishes	0	0	0	ņ	.04	1.72	0	0	0	ī	ī	ī	0	0	0	0	0	0	0	0	0	0	0	0
Decapod shrimp	10	31.3	33.3	1.98	4.02	6.9	1.1	18.6	10				0	0	0	.27	8.6	t.35	0	0	0	0	0	0
Mysid shrimp	10	2.12	33.3	64.4	5.09	60.3	98.9	81.4	100				93.1	95.5	100	06	35.8	73.9	31.8	23.2	58.4	100	100	00
Other invertebrates	0	0	0	2.48	.25	8.62	0	0	0				6.25	2.68	33.3	.54	.76	8.7	2.6	.22	5.26	0	0	0

B. Gut contents of Z₂₀₀₋₃₉₉

Quarter	. 1	2/2011			3/2011		ч	1/2011		-	1/2012		0	/2012		ς.	/2012		4	/2012		Ч	/2013	
Examined specimens		40			37			18			0			12			37			32			17	
	Z	В	ч	Z	В	ч	Z	В	F	Z	В	ч	Z	В	ч	Z	В	ч	Z	В	F	Z	В	г
Bream	0	0	0	69.	9.	3.33	0	0	0			.	0	0	0	0	0	0	0	0	0	3.03	1.57	11.1
Herring	14.9	1.3	12.5	71.7	13.9	46.7	0	0	0	,	ī	,	13.7	4.91	20	5.48	26.4	32.4	4.81	13	15.8	0	0	0
Perch	0	0	0	0	0	0	1.47	11.9	10		ī		0	0	0	0.38	11.6	2.94	0	0	0	0	0	0
Zander	0	0	0	0	0	0	2.94	41.4	20	,	ī	,	0	0	0	0	0	0	0	0	0	0	0	0
Other gobies	2.03	۲.	7.5	0	0	0	0	0	0	,	ī		.85	3.08	10	2.65	7.82	14.7	8.65	6.38	36.8	3.03	1.75	11.1
Roach	0	0	0	0	0	0	0	0	0	,	ī	,	0	0	0	0	0	0	0	0	0	6.06	30.6	22.2
Round goby	35.1	90.1	70	19.3	82.8	70	1.47	16.2	10		ī		5.98	71.4	40	1.32	24.6	11.8	5.77	42.9	31.6	9.09	38.8	22.2
Smelt	0	0	0	69.	2.16	3.33	1.47	20.5	10		ī		0	0	0	0	0	0	2.88	9.65	15.8	0	0	0
Other fishes	0	0	0	0	0	0	0	0	0	,	ī		.85	6.86	10	0	0	0	0	0	0	3.03	20.1	11.1
Decapod shrimp	10.1	6.43	25	69.	.31	3.33	10.3	8.35	50	,	ı	,	.85	3.88	10	.57	4.53	5.88	8.65	24	42.1	3.03	4.04	11.1
Mysid shrimp	34.5	1.19	47.5	6.21	.13	23.3	79.4	1.55	20		ī		77.8	9.88	50	87.3	24.5	70.6	65.4	3.77	42.1	69.7	2.94	66.7
Other invertebrates	3.38	.24	12.5	69.	60.	3.33	2.94	.13	20		-		0	0	0	2.27	.62	23.5	3.85	.32	5.26	3.03	.26	11.1

Table 7.S1. (continued)

nts of Z _{≥400}	
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Quarter		2/2011			3/2011			1/2011			/2012		5	/2012		e contra	/2012		4	/2012			/2013	
Examined specimens		2			51			16			20			13			6			32			18	
	N	В	F	Z	В	F	Z	В	F	Z	В	F	N	В	F	N	В	F	N	В	F	N	В	F
Bream	0	0	0	2.17	.87	2.33	5.88	9.28	14.3	12.5	9.98	26.7	0	0	0	0	0	0	1.37	4.01	3.33	10	3.55	21.4
Herring	20	9.	50	3.26	1.13	4.65	5.88	32.9	7.14	7.5	45	20	43.6	1.29	8.33	14.3	3.25	25	11	1.75	23.3	0	0	0
Perch	0	0	0	2.17	2.13	2.33	0	0	0	0	0	0	0	0	0	0	0	0	5.48	13	13.3	0	0	0
Zander	0	0	0	17.4	41.6	30.2	29.4	40.9	64.3	42.5	33.6	66.7	0	0	0	7.14	7.16	12.5	27.4	48.7	43.3	17.5	48.2	21.4
Other gobies	0	0	0	0	0	0	2.94	.06	7.14	2.5	.14	6.67	0	0	0	0	0	0	13.7	.66	10	20	2.91	35.7
Roach	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	20	27	35.7
Round goby	60	99.2	100	52.2	52.3	67.4	0	0	0	2	5.1	13.3	53.9	98.4	100	28.6	75.4	50	9.59	8.81	16.7	0	0	0
Smelt	0	0	0	0	0	0	2.94	.61	7.14	2.5	5.19	6.67	0	0	0	0	0	0	9.59	20.2	23.3	2.5	9.88	7.14
Other fishes	0	0	0	0	0	0	5.88	14.1	14.3	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
Decapod shrimp	0	0	0	16.3	2	32.6	38.2	2.09	50	25	Ч	20	2.56	e.	8.33	50	14.1	50	17.8	2.83	30	27.5	8.5	35.7
Mysid shrimp	20	.17	50	5.43	.02	4.65	5.88	.01	14.3	2.5	.005	6.67	0	0	0	0	.12	0	2.74	.01	3.33	0	0	0
Other invertebrates	0	0	0	1.09	0	2.33	2.94	.002	7.14	0	0	0	0	0	0	0	0	0	1.37	.05	3.33	2.5	.03	7.14

Contribution of authors to Chapter 7

Mattias Hempel collected the examined material of zander in cooperation with co-author Rüdiger Neukamm, conducted the laboratory work for diet analysis and age determination and made all calculations. He made the map, prepared all Figures and Tables, wrote the initial draft of the manuscript, the comments to the editor and the reviewers and the revised version of the manuscript. Rüdiger Neukamm provides trawl net data for the years 2008–2010. Ralf Thiel assisted in the laboratory work, the calculations and the writing process.

8 General discussion

8.1 Further spread of round goby in general and in the study area

The invasive round goby *Neogobius melanostomus* (Pallas, 1814) is still extending its distribution area in the temperate zone of the northern hemisphere. It is generally accepted by scientists concerned with round goby that almost every reachable habitat (brackish and freshwater) is accessible for establishment of this invader (e.g. BALÁŽOVÁ-L'AVRINČÍKOVÁ & KOVÁČ 2007, KORNIS & VANDER ZANDEN 2010).

Especially large rivers and artificial canals seem to be vulnerable to a development of abundant populations of invasive gobiid species. The shorelines of both heavily modified and completely manmade systems such as many large rivers and artificial canals, respectively, provide conditions that can facilitate the invasion success (e.g. JUDE & DEBOE 1996, BORCHERDING ET AL. 2011, Chapter 4). Commonly lined by stone packaging along the banks, there are multiple possibilities to hide from predators and to spawn in cavities as common for speleophil gobiids (e.g. MILLER 1986, BELANGER & CORKUM 2003). Additionally, they can profit from earlier invasions of appropriate prey organisms, e.g. dreissenid mussels and gammarid amphipods such as the killer shrimp *Dikerogammarus villosus* (Sowinsky, 1894), which are often predominant in those modified ecosystems (LEUVEN ET AL. 2009, BRANDNER 2014).

Smaller, fast-moving confluents with a more natural morphology, as well as the head waters of rivers and streams are often described to be less inhabited by invasive gobiids, even if accessible (see KORNIS & VANDER ZANDEN 2010). In these environments, numerous native species, well adapted to the present conditions over eras, seem to be the better competitors in comparison to the invaders. However, a spread of round goby into high diversity lotic tributary systems was described for the Great Lakes Region (POOS ET AL. 2010). This spread is called a secondary invasion because it took place more than ten years after the first invasion of the species in that region. Round goby was detected in six streams draining into Lake Huron and Lake Erie. The morphology of the invaded sites was mainly characterised by pool and riffle structures typical for this lotic systems.

The above mentioned distribution patterns seem to be well reflected in the study area of this dissertation in north-western Germany. Nowadays, highly abundant populations of *N. melanostomus* in this region are found almost exclusively in artificial canals and in lower sections of large rivers (own observations and reports from fishermen and anglers). In the federal state of Schleswig-Holstein, especially the Kiel Canal and the lower Trave River are inhabited by dense populations of *N. melanostomus*. Close to these, in the area of the federal state of Hamburg, the Elbe River currently

holds a population of round goby that has grown rapidly from 2008 (first report, see Chapter 4) to 2014 (high abundance, own observations). A spread of round goby to parts of the Elbe River further upstream seems to be very likely. In the German part of the Elbe, there is only one barrier interfering with migration, the weir at the city of Geesthacht, equipped with fish passes at both banks. Until now there are sporadic reports of round goby specimens from directly upstream of this dam (see Chapter 4). In 2015, first records of *N. melanostomus* were stated for the upper Elbe River (ROCHE ET AL. 2015). The newly established population in the area of Ústí nad Labem, Czech Republic, was suggested to have its source in the tidal Elbe River and port-to-port transport was assumed as the most likely vector route. This was confirmed by morphological similarities but not verified by genetic analyses so far.

Round goby can reach a new system via different ways of introduction: By natural dispersal from nearby established populations or by direct introduction events, e.g. from ship-mediated transport or intentional releases. In the Kiel Canal and the Trave River system, the round goby invasion probably took place from the eastern openings of both waters into the Baltic Sea. In the Kiel Canal, first specimens were reported from close to its eastern end, especially from Flemhuder See, a lake adjacent to the canal in the eastern region, in 2007 (NEUKAMM 2009). A spread along the canal with about 20 km per year from east to west was detected in fishing surveys of the years 2011 and 2012 (THIEL & HEMPEL 2014). In the Trave River, first records were reported from a small confluent of the river, the Schwartau, in 2008 in its lower section near the mouth of the river into the Baltic Sea. In 2012 the species was caught during electro-fishing surveys about 30 km upstream (pers. comm. Dr. M. Brunke, State Agency for Agriculture, Environment and Rural Areas, Schleswig-Holstein, Germany). At present, a further spread of round goby from the Trave River can be seen in the Elbe-Lübeck-Canal, which connects the Trave River in the north with the Elbe River in the south. In 2012, the species N. melanostomus was present only at the northern end of the Elbe-Lübeck-Canal, close to the Trave River (NEUKAMM 2015). In 2015, round goby was found in large numbers about 20 km south of the Trave River (own observations).

In the tidal Elbe River, especially in the area of the port of Hamburg (see Chapter 4), introduction of round goby possibly took place in a different way than by natural dispersal from the established populations in the Baltic Sea area. The first record was reported in 2008 from the port area (Chapter 4) at a time when no round goby specimens were reported from down- or upstream sections of the Elbe River. That gives a hint for a direct introduction event into the port of Hamburg where at present an abundant population of this species exists (own observations). As the port of Hamburg is the 15th-largest port worldwide and is visited by about 12,000 ships every year (HPA 2013, Chapter 4), there are numerous possibilities for allochthonous species to reach this place, e.g. by transport via ballast water. About every 10th specimen caught in the port lacks one major characteristic of the

species: The black spot in the posterior part of the first dorsal fin. The lack of this spot was elsewhere reported only for Lake Erie, North America (ISSG 2013) and for the Weser River, Germany (personal communication Prof. Dr. H. Brunken, Bremen University of Applied Sciences, Germany). Therefore, a connection between the populations of round goby in north-western Germany (Weser River and Elbe River) and North America might exist. A back-import of *N. melanostomus* from North America to Europe, as mentioned for the quagga mussel *Dreissena bugensis* (Andrusov, 1897) (BOLTOVSKOY 2015), appears to be possible. However, first genetic analyses did not indicate a different origin of specimens with and without the black spot from the port of Hamburg (personal communication Prof. R. Thiel, University of Hamburg, Germany) indicating an identical source population.

The morphology of the inhabited water body has a strong effect on its potential for holding highly abundant round goby populations. This is mainly caused by the number of accessible spawning sites and possibilities to hide for the speleophil species (see MILLER 1986). The Kiel Canal and the Elbe River exhibit the typical image of artificial banks with the preferred stone packaging (see PILLE 2009, TONN 2012) in many areas. Therefore, it is not unusual that high densities of the invader are able to develop. The lower Trave River, however, holds high abundances of round goby even in the strongly brackish, lake-like areas near the Baltic Sea, which do not hold large areas covered by stone packaging or rocky substrate. The morphology of the habitat there is more similar to that of the Bay of Gdańsk at the Polish Baltic Sea coast. The examples of the Bay of Gdańsk (e.g. SAPOTA 2004) and the lower Trave River (THIEL & HEMPEL 2014) clarify that *N. melanostomus* can also build up dense populations in areas dominated by fine substrate. Because of the strong limitation of stony nesting substrate in these areas, round goby uses almost every solid material for egg deposition (see SAPOTA ET AL. 2014). Large patches of eggs can be found, e.g. under plastic waste (own observations) and large shells of bivalves may also be used.

The salinity tolerance is an important factor that can determine the physiological possibilities for a further spread of round goby. These possibilities comprise the dispersal by ship-mediated transport (ballast water) and by natural migration. Regarding current literature, the salinity tolerance of round goby seems not completely clarified. There are hints that the species must be somehow limited by high salinities. SAPOTA (2006) reported that no population exists in full oceanic salinity, but hypothetically *N. melanostomus* can live in such conditions. KORNIS ET AL. (2012) assume that *N. melanostomus* probably has a salinity tolerance <30. Transport via ballast water is supposed to be the most important way of round goby introductions worldwide (e.g. RICCIARDI & MACISAAC 2000, REID & ORLOVA 2002, SAPOTA 2004). Nowadays, stringent ballast water management requirements are in place to avoid introductions of alien species into the Great Lakes (BWWG 2015). Vessels have to conduct mid-ocean water exchange and to reach a salinity of at least 30 in the ballast water tanks before entering the

Great Lakes Seaway. Nevertheless, at least two Ponto-Caspian species, the amphipod Echinogammarus ischnus (Stebbing, 1899) and the fishhook waterflea Cercopagis pengoi (Ostroumov, 1891), have apparently entered the Great Lakes system after these regulations have been implemented (RICCIARDI & MACISAAC 2000). Thus, ballast water management requirements are either not realized by all vessels or not effective for all target species. In the salinity tolerance experiment, round goby specimens held at a salinity of 30 showed reduced growth but no mortality occurred in this approach (Chapter 5). With regard to this result it is possible that an oceanic water exchange which ensures a salinity of 30 in the tanks is not effective in killing all N. melanostomus specimens. Additionally, the rate of oceanic ballast water exchange depends on the used equipment and in reality the water exchange must work more or less like an acclimatization procedure. The salinity in ballast water tanks is in many cases continuously changed which could enable salinity-tolerant species to adapt to higher salinities. Considering the possibilities of further spread of round goby (and other gobiid species) in Eurasia, the procedure of oceanic water exchange seems to be irrelevant. Vessels that can carry round goby in their ballast tanks often travel only within fresh- and brackish waters (e.g. between ports of the Baltic Sea area or inland along canals and large rivers). A transport of gobiid eggs attached to ships hulls (BRANDNER ET AL. 2013b, HOLM ET AL. 2014, HIRSCH ET AL. 2016) may be another possible way of introductions, at least over short distances.

The possibilities to prevent further spread and introductions of *N. melanostomus* are discussed in many areas concerned. Existing dams are supposed as possible upstream migration barriers (e.g. KORNIS ET AL. 2012, VERREYCKEN 2013, HOLM ET AL. 2014). But at least in large rivers, dams are commonly equipped with fish passes that are desperately needed to secure up- and downstream fish migration e.g. to reach spawn areas or wintering habitats. For rivers and creeks in the European Union, the EU Water Framework Directive that was adopted in 2000, demands continuity as an important requirement to achieve a "good" river status. In the Alster River in Hamburg, an Elbe River tributary in the area of the study site of Chapter 4, a fish pass was newly constructed and finished in 2016 (HAMBURG.DE 2016). This can strongly enhance the possibilities of round goby from the Elbe River to reach the upper parts of the Alster River system.

An important task is preventing enclosed water bodies from the introductions of round goby for example by the release of bait or aquarium fish (e.g. VERREYCKEN 2013, HOLM ET AL. 2014).

Removal of highly abundant gobiids such as *N. melanostomus* from an invaded ecosystem seems to be almost impossible. At least in large waters it is extremely time-consuming, expensive and not realizable without harming other animals. Different piscicides were tested for their toxicity on round goby in comparison with other species, native to the Great Lakes (SCHREIER ET AL. 2008). No toxin was found that could solely eradicate *N. melanostomus* without killing other species. Bottom release formulations

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of Bayluscide[®] and antimycin were supposed to possibly have some use for selective removal of round goby. VERREYCKEN (2013) summarized that piscicides may be usable for eradication of emerging populations but only in confined areas and at high costs. Recent tests also evaluated the suitability of spawn traps for the removal of gobiid eggs (HOLM ET AL. 2014). For this purpose, packages of plastic pipes and clay pots were placed under water accessible as a spawning habitat for gobiids. Attached eggs can easily be detached from these packages when pulled out of the water. This procedure can decrease the number of hatched embryos in local populations but has little prospect of success for complete removal of those species.

8.2 Development of introduced round goby populations and the related consequences

Invasive species such as *N. melanostomus* are able to build up high abundances shortly after introduction into a new environment (see ABELE ET AL. 2013). These rapidly increasing population sizes are able to cause major environmental changes in the invaded ecosystems. Following the integrated conceptual model of a biological invasion (BRANDNER 2014), there is normally a lag-phase with low but stable abundance of the invader after its establishment. Then, a trigger (such as changes in water temperature) causes the further spread of the species and the impact-phase with a fast increasing abundance starts. In this phase, a heavy impact on the inhabited ecosystem is possible or even likely. After reaching extremely high population density, the extensive utilization of resources may limit further spread. A boom-and-bust cycle characterised by repeating increases and declines of the introduced species abundance seems to be the inevitable consequence (BRANDNER 2014). Not only the limitation of resources can counteract further increases of population size, additional effects from higher trophic levels due to predation can stabilise population sizes or even cause strong declines of the invader's abundance (e.g. HUO ET AL. 2014, Chapter 7).

In the tidal Elbe River in the area of Hamburg (study site of Chapter 4), the number of round goby records increased quickly in 2013 and 2014 (own observations and records provided by anglers and fishermen). Nowadays, round goby is quite common in catches of anglers at the banks of the main river and it can regularly be found in the stomachs of zander *Sander lucioperca* (Linnaeus, 1758). Just as the nearby situated Kiel Canal, the lower Elbe River holds an abundant population of zander as the major predatory species (see THIEL ET AL. 1995, EICK & THIEL 2014). As shown for the Kiel Canal (Chapter 7), the round goby abundance in the Elbe River may decrease in the near future as a consequence of predation by zander. On the other hand, reliance of zander on *N. melanostomus* as prey in the Elbe River might not be as important as in the Kiel Canal due to the higher productivity of the lower Elbe River ecosystem. Further investigations would help to track the development in the next years.

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Influences of tidal depth variations on round goby populations have not been described in literature yet. The Elbe River in the area of Hamburg shows mean variations in depth between low and high tide of almost four meters (BSH 2015). Large parts of the stone packaging that covers the banks in many areas fall dry during low tide, forcing the fishes out of cavities and making a large proportion of possible spawning places unusable. Regarding the high abundance of round goby despite to that fact, these tidal variations seem not to cause major problems for the establishment and/or breeding of the species. Again this highlights the high adaptability of *N. melanostomus*.

Round goby establishment in invaded ecosystems can strongly influence native fish species through competition. Highly abundant round goby populations can have negative impact on species with similar habitat requirements (e.g. SKÓRA & RZEZNIK 2001, LAUER ET AL. 2004, KARLSON ET AL. 2007, BERGSTROM & MENSINGER 2009, VAN KESSEL ET AL. 2016) and reduce the abundance of native species in invaded areas (pers. comm. G. Smith cited in CROSSMAN ET AL. 1992). In the Bay of Gdańsk, competition for food was supposed to exist between round goby and several native fish species (Skóra & RZEZNIK 2001). In this study, large specimens of N. melanostomus (total length (TL) > 100 mm) fed primarily on molluscs while small specimens (TL 20–40 mm) incorporated crustaceans and polychaetes into their diet. Therefore, round goby in the Bay of Gdańsk may compete for food with plaice Pleuronectes platessa Linnaeus, 1758, black goby Gobius niger Linnaeus, 1758 and flounder Platichthys flesus (Linnaeus, 1758), which all showed similarities in their diets. KARLSON ET AL. (2007) investigated the competition with flounder of the Bay of Gdańsk in detail. They found that flounder and round goby consumed similar species and sizes of prey and, therefore, resumed a high potential for food competition between both species. Additionally, the abundances of the two species were negatively correlated, which can be caused by high round goby abundance in shallow areas forcing migration of flounder to deeper parts.

LAUER ET AL. (2004) reported a rapid decline in the abundance of mottled sculpin *Cottus bairdii* Girard, 1850 and Johnny darter *Etheostoma nigrum* Rafinesque, 1820 in Lake Michigan, Great Lakes, after the establishment of round goby. This decline is supposed to have been caused by biological interaction with round goby, e.g. by competition for food, shelter and spawning sites. Aquaria experiments conducted by BERGSTROM & MENSINGER (2009) indicate that round goby is able to outcompete native fish species such as slimy sculpin *Cottus cognatus* Richardson, 1836, spoonhead sculpin *Cottus ricei* (Nelson, 1876) and logperch *Percina caprodes* (Rafinesque, 1818) in interspecific resource competition. In the Lower Rhine, a large European stream, the percentages of different fish species were inspected in nearshore habitats in 2009 (BORCHERDING ET AL. 2011). That was about three years after the first record of invasive gobiids in this area. In the catches of electro-fishing surveys mainly bighead goby *Ponticola kessleri* (Günther, 1861) and round goby were present. Non-gobiid species made up less than

20% in both sampled months (August and October). Due to the dominance of gobiids it is supposed that they will have a strong effect on the biocoenosis of the river through competition for habitat and/or food or through feeding on eggs or juveniles of other fish species. In the Meuse River, the Netherlands, the occurrence of round goby and a thereby caused predation and competition for shelter and food was supposed to be the reason for a rapid decline of native river bullhead *Cottus perifretum* Freyhof, Kottelat & Nolte, 2005 (VAN KESSEL ET AL. 2016). In contrast, the two other invasive gobiid species occurring in that region, tubenose goby *Proterorhinus semilunaris* (Heckel, 1837) and bighead goby, do not have such strong impact on the densities of river bullhead.

In the Kiel Canal and the lower Trave River, a diet overlap between small native gobiid species of the genus *Pomatoschistus* (sand goby *P. minutus* (Pallas, 1770) and common goby *P. microps* (Krøyer, 1838)) and at least smaller individuals of round goby was found. Both species preyed mainly on small crustaceans, chironomids and polychaetes (KNÖRR 2013, THIEL & HEMPEL 2014). Related to this dietary overlap and due to the possibility of competition for habitat and spawning sites, it seems likely that the abundance of *Pomatoschistus* spp. decreased due to the establishment of *N. melanostomus* in the Kiel Canal. This was also indicated by abundance data of both taxa from beach seining in nearshore areas of the canal, before and after the invader's occurrence (Table 8.1). In contrast, the total fish abundance in the area studied decreased nearly as much as the abundance of *Pomatoschistus* spp. did, which indicates that the development could also be a general pattern.

Table 8.1. Total fish abundance and abundance of present gobiid species at the study site	"Schirnaumündung",
Kiel Canal, Germany. Data 1995/1996 by KAFEMANN (2000) and 2011/2012 by THIEL & HEMPEL (2014). All abundance
data in Ind. ha ⁻¹ .	

Year	1995	1996	before establishment 1995/1996 (mean)	2011	2012	after establishment 2011/2012 (mean)
Total fish abundance	95 <i>,</i> 896	20,229	58,063 (±53,505)	19,678	10,823	15,251 (±6,261)
P. minutus	30,057	4,698	17,378 (±17,932)	10,843	1,323	6,083 (±6,732)
P. microps	16,197	500	8,349 (±11,099)	1,353	1,503	1,428 (±106)
Pomatoschistus spp.	46,254	5,198	25,726 (±29,031)	12,196	2,826	7,511 (±6,626)
N. melanostomus	0	0	0	1,251	734	993 (±366)

Other fishes in the studied region that may be affected by competition with the round goby, especially for food, are, e.g. flounder, European perch *Perca fluviatilis* Linnaeus, 1758, several cyprinid species, black goby and European eel *Anguilla anguilla* (Linnaeus, 1758). At least in regions with highly abundant populations of *N. melanostomus* it seems likely that abundance of some of those species will decrease.

Growth and maximum size of *N. melanostomus* are of particular interest because they can differ strongly between ecosystems and populations. In the study area, major differences can be seen in the

populations of round goby from the Kiel Canal and the lower Trave River regarding the growth of the species. Inspected specimens from the lower Trave River (Figure 8.1) have the potential to grow almost twice as big as specimens from the Kiel Canal (THIEL & HEMPEL 2014, Chapter 6). The asymptotic length L_{∞} (*TL*, mm), calculated by the von Bertalanffy growth equation, was 237 mm for round goby from the lower Trave River and 133 mm for those from the Kiel Canal. Calculations were performed using FISAT II version 1.2.2 (FAO, Rome, Italy) as described in Chapter 6 and took place with specimens caught in 2011/2012. The differences in growth could have been associated with the local conditions of both ecosystems, especially considering the diet of *N. melanostomus*. Round goby in the Trave River mainly ingested mussels, barnacles and sea squirts as prey whereas fish from the Kiel Canal mainly consumed smaller invertebrates such as isopods, annelids and chironomids (see Chapter 6).



Figure 8.1. Length-growth curve estimated for round goby from the lower Trave River (*n* = 155, year 2012).

Round goby populations from different areas (both native and introduced) vary strongly in the maximum length of the specimens. Current literature gives several possible explanations for this phenomenon. CORKUM ET AL. (2004) indicate the salinity of the inhabited water body as an explanation for the maximum size of round goby. From unpublished data by Svetlana Rudnicka (Institute of Fisheries, Bulgaria) they report that in freshwater and slightly brackish water (salinity 4.5–7.8) dwarf forms of this species occur whereas under more marine conditions (salinity 15–17) larger sizes and faster growth were common. Small maximum size was also found by MACINNIS & CORKUM (2000) for the pure freshwater population of round goby in the Detroit River, Great Lakes area. The same observation was made for the studied areas in the Kiel Canal (salinity of about 5–7) and the lower Trave River (salinity of up to about 15 depending on the influx from the Baltic Sea). BRANDNER ET AL. (2013b) found that round goby specimens at the invasion front in the Danube River reached larger size and had higher condition in comparison to stretches of the river with established populations. The situation cannot be

compared to the studied areas of the Kiel Canal and the lower Trave River as here both populations can be designated as being established. Highly abundant populations have been present at least since 2008.

Similar to growth, the age at maturity of round goby can differ between introduced and native populations. The species was found to mature earlier in introduced populations than in native ones (e.g. MACINNIS & CORKUM 2000). In our study areas we observed one-year-old mature females and males that participated in spawning at an age of less than two years. In contrast, age at maturity for round goby in its native area in the Pontocaspian region is 2–3 years for females and 3–4 years for males (MILLER 1986).

Fish in the diet of highly abundant invasive gobiids could be a possible reason for the decline of native species. Especially in non-scientific publications, the first aspect associated with them is their role as a predator of fish eggs and larvae. The newly established species were held responsible for strong declines in the abundance of, e.g. cyprinid species through direct predation.

Numerous authors in Europe and North America have inspected the diet of invasive gobiids from the Pontocaspian area, especially that of round goby. Two examples from brackish waters of the Baltic Sea come from Denmark and Poland. While inspecting the diet of 1,080 specimens of round goby from the Puck Bay and the Gulf of Gdańsk, Poland, Baltic Sea, in 2001 (WANDZEL 2003), fishes were found as a food taxon throughout the whole sampled period from February to September. Fish species consumed were ruffe *Gymnocephalus cernua* (Linnaeus, 1758) and species of the genus *Pomatoschistus*. In addition, spawn of round goby was cannibalized by males. Depending on the season fish made up 2–14% wet weight of the diet, mussels made up the main part with 82–95%. In the brackish waters of the east coast of Denmark, Baltic Sea, the diet of round goby was inspected in November 2010 (Azour 2011). Only eight of the 474 inspected fishes had eaten three-spined stickleback *Gasterosteus aculeatus* Linnaeus, 1758 which was the only fish species found in the diet. No eggs were found, however, this was unlikely due to the chosen time anyway, which did not coincide with the typical spawning season of many fish species in that region.

Considering the large streams in Europe, data on the diet of introduced gobies are available especially from the Rhine River and the Danube River. In the upper Danube River in early and late summer of 2010, 283 specimens of *N. melanostomus* and 235 specimens of bighead goby were inspected for food consumed (BRANDNER ET AL. 2013a). Round goby preyed only marginally on fish in late summer. In contrast, fish represented an important prey item with 5% of index of food importance in early summer and 10% in late summer in bighead goby. The major part of the diet of both species was comprised of crustaceans, while chironomids and molluscs represented minor parts.

ADÁMEK ET AL. (2007) inspected the diet of three invasive gobiids (*P. kessleri*, Monkey goby *Neogobius fluviatilis* (Pallas, 1814) and *N. melanostomus*) in the area of the confluence of the Danube and Hron rivers near the town of Štúrovo, Slovakia. Fish were found only in the diet of two individuals of bighead goby. Monkey goby preyed mostly on Corophiidae and chironomid larvae. The amphipod *D. villosus* appeared with about 90% of mass in the food taxa of round goby, but only five individuals of round goby were examined.

In the Lower Rhine the three invasive gobiid species *N. fluviatilis, N. melanostomus* and *P. kessleri* were inspected for food consumed (BORCHERDING ET AL. 2013). Food consumed was comparable to the situation in the Danube River. *N. fluviatilis* and *N. melanostomus* did not prey on fish while the third species *P. kessleri* consumed up to more than 10% of fish depending on the season. Major food taxa of all three species were crustaceans and chironomids. The obvious decline in native species after establishment of invasive gobiid species is thus likely to be rather the result of competition for food and habitat than a matter of predation by invasive species (see e.g. DUBS & CORKUM 1996, JANSSEN & JUDE 2001, KARLSON ET AL. 2007, BRANDNER 2014).

In the study area we investigated the prey of round goby in 2011 and 2012 for the lower Trave River and the Kiel Canal. The relative importance (*RI*) of fish and especially fish eggs in the diet was minor in both ecosystems (Figure 8.2, Chapter 6). In the Trave River only *N. melanostomus* of \geq 100 mm *TL* preyed on fish and the *RI* for this taxon was less than 7%. In the Kiel Canal (Chapter 6) *N. melanostomus* of the length classes 50–99 and \geq 100 mm preyed on fish (1.7 and 4.4% of *RI* respectively) and fish eggs (0.3 and 2.1% of *RI* respectively; only one specimen in each size class). Fish species eaten were youngof-the-year Atlantic herring *Clupea harengus* Linnaeus, 1758 (in seasons when larvae and juveniles of this species occurred in high abundances) and small gobies of the genus *Pomatoschistus*. These results clearly reflect the results of other studies described above concerning the diet of round goby.

In the tidal Elbe River (study site of Chapter 4) the diet of the newly established round goby was investigated in autumn 2014 and spring 2015 (GIESLER 2015). In autumn no fish or eggs were found in the diet of *N. melanostomus*. While this was consistent with three out of four study areas in spring, in the fourth area every second round goby had eaten percid eggs (probably *G. cernua*). Predation of eggs is also confirmed for different fish species with benthic eggs in the Great Lakes area, North America (e.g. STEINHART ET AL. 2004a, ROSEMAN ET AL. 2006). Thus, a seasonally increased predation on fish eggs by round goby seems possible for numerous ecosystems. The high seasonal and geographical variability of the opportunistic species diet was confirmed also by surveys in its native area, the Black Sea (BĂNARU & HARMELIN-VIVIEN 2009). There, in an area which was hydrologically influenced by the Danube River delta, one study site was situated directly seawards of the delta (northern site) and one further away from it (southern site). At the northern site, *N. melanostomus* consumed high proportions

(64% of *RI*) of European sprat *Sprattus sprattus* (Linnaeus, 1758) in spring, while at the southern site no fish were found in the diet throughout the year.



Figure 8.2. Relative importance index (*RI*) of prey taxa classified into three length classes by total length (*TL*) of round goby from the lower Trave River. Numbers of non-empty guts examined are given above bars.

8.3 Predatory fish influenced by round goby

Round goby can contribute important parts to the diet of predators and numerous authors describe positive effects of incorporating the newly available gobiids into the diet of predators (e.g. JAKUBAS 2004, STEINHART ET AL. 2004b, KING ET AL. 2008, Chapter 7). The relatively small size of *N. melanostomus* and the tubular shape makes it easy to swallow and its common occurrence and high abundance make it an easily accessible prey, at least for bottom orientated predators. In addition, the reproductive behaviour including multiple spawning events from April until September if water temperature lies between 9–26°C (see CHARLEBOIS ET AL. 1997, GERTZEN ET AL. 2016), ensures the availability of multiple differently sized specimens of round goby all year round (see STEINHART ET AL. 2004b).

While predators may benefit from gobiids as easily available prey throughout the year, they may simultaneously suffer from them at earlier life stages. That is, invasive gobiids may prey on eggs (see CHOTKOWSKI & MARSDEN 1999) and larvae, compete with juveniles for food and habitat and finally may act as vectors for parasites (EMDE ET AL. 2014). Additionally, not each predatory fish species that latterly uses *N. melanostomus* as a major prey organism benefits from this diet shift by faster growth or better condition (e.g. POTHOVEN & MADENJIAN 2013, CRANE ET AL. 2015).

In northern Europe, e.g. the following predatory fish species could be strongly influenced by round goby establishment:

The European perch, identified as a major predator of round goby (e.g. ALMQVIST ET AL. 2010, MIKL ET AL. 2017, own observations), is a common species almost Europe-wide. Predation on eggs by round goby seems to be more likely for perch than for the nest-guarding zander (see Chapter 7). In the latitude of northern Germany, *P. fluviatilis* spawns in March–May (WUNDSCH 1963) and does not conduct any parental care of the eggs or larvae (THORPE 1977). Eggs are deposited in shallow water on plants, wood and stones, easily accessible for *N. melanostomus*. On the other hand, perch eggs are covered by a thick protective mucilaginous layer and it was reported that they were not eaten by any fish (THORPE 1977). It is unclear whether this is transferable to round goby or only valid for species native to the distribution area of perch and should be clarified, e.g. by performing aquaria experiments with round goby offered perch eggs as diet. In addition to a possible egg predation, an overlap in the diet of perch and round goby (e.g. SkóRA & RZEZNIK 2001, COPP ET AL. 2008, HOLM ET AL. 2014) can result in strong competition for resources and thereby negatively influence perch populations in invaded areas (see GERTZEN 2016).

Northern pike *Esox lucius* Linnaeus, 1758 represents one of the largest European predatory fishes. *E. lucius* is known from several European ecosystems invaded by the round goby, e.g. it is common in the brackish coastal waters of Rugia Island, German Baltic Sea, and in the Danube River. In Lake St. Pierre, the largest fluvial lake of the St. Lawrence River system in northern America, northern pike incorporates round goby in its diet (REYJOL ET AL. 2010), 11% of inspected stomachs solely contained round gobies and 15% contained this species as part of a mixed diet. In the Dyje River, Czech Republic, MIKL ET AL. (2017) found that *E. lucius* consumed 19.8% round goby by mass. A strong diet overlap between *E. lucius* and round goby is not likely because of the almost exclusively piscivorous behaviour of pike (e.g. MANN 1982). Predation of pike eggs and larvae by round goby seems possible as the pike spawns in flat areas on macrophytes.

Another predator, which possibly profits from *N. melanostomus* as prey, is the European eel. Supported by our own observations and information from fishermen at the Kiel Canal, at least larger specimens of European eel use round goby as a part of their diet. Due to its snakelike appearance, European eel should be able to prey on gobies even if they try to hide in the interstitial of the stone packaging or in rocky substrate. However, the establishment of round goby can also have negative consequences for *A. anguilla*. Usage of this prey may result in higher infection rates of European eel with the eel parasite *Anguillicoloides crassus* (Kuwahara, Niimi & Itagaki, 1974). EMDE ET AL. (2014) found each third round goby infected by *A. crassus* larvae, which occur exclusively inside of acanthocephalan cysts (another parasite). They supposed that the eel parasite might bypass the
immune system of *N. melanostomus* by using the acanthocephalan cysts as a "Trojan horse". This may explain the significantly higher infection rate of European eel from the brackish Kiel Canal in comparison to specimens from the nearby freshwater system Elbe-Lübeck-Canal (KULLMANN 2014). Generally, the infection rate of eels with *A. crassus* appears to be higher in freshwater ecosystems than in brackish or oceanic habitats (JAKOB ET AL. 2009). However, in the years under study the abundance of round goby in the Kiel Canal was much higher than in the Elbe-Lübeck-Canal, where the population had just started to increase (NEUKAMM 2015).

Cod *Gadus morhua* Linnaeus, 1758 is a typical inhabitant of the Baltic Sea. Thus, there is a large overlap in the distribution areas of this species and round goby. In the Gulf of Gdańsk, Baltic Sea, round goby represented on average 22% of cod diet by mass, varying between the size classes of the predator (ALMQVIST ET AL. 2010). GRUSZKA & WIĘCASZEK (2011) reported from the same region that cod of <450 mm *TL* consumed 42% round goby by mass while cod >450 mm consumed 32%. Reports of local fishermen from the Bay of Lübeck, western coast of the Baltic Sea, suggest round goby to be an important prey of cod for this region as well. A diet overlap between *G. morhua* and *N. melanostomus* is likely, at least for smaller specimens of both species. In the Bay of Gdańsk diet of small cod of 50– 150 mm *TL* comprised 30% decapods and 42% polychaetes by mass (ALMQVIST ET AL. 2010). These taxa also represent important prey items of round goby in the Baltic Sea (SKÓRA & RZEZNIK 2001, KARLSON ET AL. 2007, AZOUR 2011) and adjacent brackish waters (THIEL & HEMPEL 2014, Chapter 6).

The European flounder is likely to be negatively influenced by *N. melanostomus*. In the Bay of Gdańsk, KARLSON ET AL. (2007) found a high diet overlap between small flounder and round goby: Bivalves, polychaetes and gastropods were important food items of both species. Referring to their data, KARLSON ET AL. (2007) concluded that high abundance of *N. melanostomus* might restrict habitat utilization of flounder and therefore could affect this commercially important species negatively. In contrast, we found juvenile specimens of round goby in the stomachs of flounder from the Kiel Canal demonstrating that this species incorporates the newly available prey in its diet as well.

Wels catfish *Silurus glanis* Linnaeus, 1758 is another predatory fish species in European waters that is likely to be influenced by the round goby. In the Dyje River, Czech Republic, invasive gobiids (round goby and tubenose goby) represented 41.5% of diet by mass for subadult wels catfish (MIKL ET AL. 2017). In addition, numerous reports from anglers confirm the usage of *N. melanostomus* as prey for *S. glanis* (e.g. ANGELFIEBER.COM 2010, ANGELFORUM.AT 2012).

For zander, which highly relies on round goby as prey in the Kiel Canal, we found a faster growth and better body condition in years after the round goby invasion (compare Table 8.2, Chapter 7). Before the establishment of round goby in the Kiel Canal specimens of zander in this water body showed slow growth reaching only a low hypothetic asymptotic length (L_{∞}) with 685 mm of *TL* (KAFEMANN & THIEL 1998, KAFEMANN 2000). Only for one lake in southern Finland, a slightly lower L_{∞} (653 mm *TL*) was reported, which was supposed to be mainly caused by the high abundance of zander in the lake (MILARDI ET AL. 2011, compare Table 8.2). After the establishment of *N. melanostomus*, the L_{∞} in the studied region of the Kiel Canal reached 937 mm of *TL* (Chapter 7), which represents an increase of about one third. Thus, L_{∞} was shifted much more to the upper bound for the species zander, comparable to L_{∞} of relatively fast growing zander in the highly productive tidal Elbe River (Table 8.2).

Considering all available data it appears that almost every predatory fish species that is known from European fresh and brackish waters can potentially act as a predator of round goby.

Regarding different predatory species in different ecosystems, the newly established round goby has diverse effects due to the use as food. CRANE ET AL. (2015) published a long-term study of the body condition of four predators from Lake Erie and Lake Ontario, northern America, comparing the situation before and after the round goby invasion. One of the four inspected species (smallmouth bass *Micropterus dolomieu* Lacepède, 1802) showed a general increase in condition after the establishment of round goby. The other three species only showed minor increases in condition, only for some of the inspected length classes or in only one of the two inspected lakes. Condition of large sized burbot *Lota lota* (Linnaeus, 1758) even decreased after incorporating round goby in its diet.

Amongst others, the above mentioned results suggest that only those species with limited food resources in former time are able to benefit as shown, e.g. by increased condition, faster growth, higher abundance, range expansion or lower age at maturity. This effect was not only limited to predatory fishes (e.g. STEINHART ET AL. 2004b, JOHNSON ET AL. 2005, CRANE ET AL. 2015, Chapter 7), but has also been demonstrated for birds (JAKUBAS 2004, TUCKER & SEEFELT 2014) and snakes (KING ET AL. 2008).

In addition to the availability of resources, the geographical location of the inhabited water body can strongly influence the growth of the fishes living therein. For zander, a fast growth was reported for populations in the southern part of its distribution area (e.g. BíRó 1970, HARKA 1977, POULET ET AL. 2004, PÉREZ-BOTE & ROSO 2012, Table 8.2). This was implicated to be at least partially caused by the comparably high water temperatures that are close to a physiological optimum (27°C) of zander in summer. A positive effect on growth due to higher yearly average temperatures was also found by BUIJSE ET AL. (1992) for zander in Lake Ijssel, the Netherlands. However, faster growth in warmer conditions cannot be applied for every water body, which is obvious, e.g. in the slow growing population of Lake Eğirdir, Turkey (BALIK ET AL. 2004) and the fast growing population of the southern Baltic Sea (HAHLBECK & MÜLLER 2003). However, if the right combination of at least the availability of

resources, the density of competitors for food and the temperature is not given, growth cannot reach a species maximum (see NEUHAUS 1934, BORCHERDING ET AL. 2007).

Water body	Age group						Author (Course)
	Ι	II	111	IV	V	VI	Author (Source)
Kiel Canal (2011–2013)	219	343	446	531	601	659	Chapter 7
Kiel Canal (1995/1996)	141	266	353	424	474	524	Kafemann & Thiel (1998)
Tidal Elbe River, Germany	151	315	430	533	611	664	Kafemann & Thiel (1998)
Fumemorte canal, France	275	431	605	734	796	-	Poulet et al. (2004)*
Alcántara Reservoir, Spain	242**	344**	431**	506**	571**	627**	Pérez-Bote & Roso (2012)
Lake Eğirdir, Turkey	197**	263**	324**	380**	431**	479**	BALIK ET AL. (2004)*
Lake Balaton, Hungary	210	301	376	440	507	562	Bíró (1970)*
Tisza River, Hungary	205	320	407	486	562	635	Harka (1977)*
Lake Sahajärvi, Finland	74**	133**	186**	235**	279**	318**	MILARDI ET AL. (2011)
Southern Baltic Sea, Germany/Poland	257**	352**	439**	519**	592**	659**	Hahlbeck & Müller (2003)

Table 8.2. Back calculated total length (mm) of zander (age groups I–VI) for different water bodies.

*Some authors provide fork length (*FL*) or standard length (*SL*) of zander. These were converted into total length (*TL*) after TURKI ET AL. (2009) using the mean data of female and male specimens (*TL* / *SL* = 1.198, *TL* / *FL* = 1.066). **calculated based on the von Bertalanffy growth equation given by the authors.

Abundance control of *N. melanostomus* by predators with strong reliance on the non-indigenous species is supposed to take place in different ecosystems (e.g. VERREYCKEN 2013, HUO ET AL. 2014). In Lake Michigan, northern America, the annual mortality rate of *N. melanostomus* is estimated from 0.79 to 0.84 (HUO ET AL. 2014). Top-down predatory control by burbot as the main predator was suggested to be the most likely reason for this high mortality rate. For the Rhine River, Germany, reintroduction of burbot was mentioned as a way to control the density of invasive gobiids. This was confirmed by aquaria experiments in which burbot preyed successfully on offered monkey goby and round goby (KUNDRUHN 2014). The effect of predatory control was also supposed to take place in the Kiel Canal (Chapter 7). The high abundance of zander in this ecosystem is probably the reason for the strong decrease in round goby abundance after peak values.

8.4 Outlook

Caused by the still ongoing spreading of round goby into many different ecosystems of the northern hemisphere, the species and its impact are still of concern for current and future research projects. A lot of issues have not been addressed so far.

Regarding the findings of Chapter 4 and the round goby population in the tidal Elbe River, which is still growing since that research, questions about the future development of the species in the Elbe River

system are mandatory: How will the spread of the species in regions up- and downstream of Hamburg take place? Will there be an "invasion front" of upstream migrating specimens as described for the Danube River by BRANDNER (2014)? Or will there be multiple places of introduction with the first example at Ústí nad Labem, upper Elbe River, Czech Republic, in 2015 (ROCHE ET AL. 2015)? Another possible place of introduction is the connection with the Mittelland Canal situated about 270 km upstream of Hamburg, nearby the city of Magdeburg. This canal nowadays holds an abundant population of round goby in parts situated west of this connection with the Elbe River (see MATTEIKAT 2015, own observations). In the area of the most upstream record from Chapter 4, the Mittelland Canal is also connected with the Elbe River by the Elbe Lateral Canal with a length of about 100 km. For the Elbe Lateral Canal itself, first catches of round goby were reported by anglers in 2014 (FEHMARN-ANGLER.NET 2014). Downstream of Hamburg, the estuary of the Elbe River begins. It drains into the North Sea with full oceanic salinity. The question here is: Is N. melanostomus able to build up an abundant population in the high saline parts close to the North Sea as it does in estuaries of the Baltic Sea with less strong gradients in salinity? To answer these questions, further research activities concerning this species should be carried out. In addition, the origin of the round goby population in the area of the Hamburg Harbour should be clarified by genetic analyses comparing specimens from the tidal Elbe River with those from different native and introduced populations in the world.

Until now, in smaller confluents of the distribution area, a high abundance of *N. melanostomus* was reported only nearby their entries into the densely populated larger rivers or canals. Further upstream, the occurrence of round goby was reported from single specimens only, caught for example during electro-fishing surveys. Monitoring measures, for example in the course of the EU Water Framework Directive, will show if the observed distribution patterns are stable or only a snapshot of a continuing development.

In addition, it seems to be only a matter of time until other invasive pontocaspian gobiid species establish and build up high densities in the studied ecosystems with all their connections to other systems and the high shipping traffic. One of these species was already present: The bighead goby, was first observed in the Elbe River close to the city of Hamburg in 2015 (THIEL ET AL. 2017).

In Chapter 5 we investigated the salinity tolerance of juvenile round goby. Based on the results of this research, waters with nearly ocean-like salinity seem to be accessible for establishment of the species, but no oceanic population of *N. melanostomus* is known (SAPOTA 2006). Given that the salinity tolerance of fish species can differ between juveniles and adults and that the eggs may need lower salinities to develop (e.g. LEHTONEN ET AL. 1996), this distribution pattern can be triggered by the first life stage. The conduction of an experiment, dealing with the development of round goby eggs under different salinities, would be helpful to clarify this question. In such an approach the salinity levels of

Chapter 5 (0, 7.5, 15 and 30) can be used and the percentages of successfully hatched embryos should be compared. In this way it is possible to find out if the development of round goby eggs may requires a salinity considerably lower than 30.

Within the work on Chapter 6 it becomes apparent that the possibilities of competition between native fish species and round goby are numerous, also in the case of the ecosystem Kiel Canal. It will be important to monitor the future development, e.g. of the population of small native gobiids from the genus *Pomatoschistus*. The high diet overlap with round goby and the risk for direct predation and/or competition for spawning sites makes the small native gobiids vulnerable for negative impacts of the established round goby population.

Considering Chapter 7, it is of interest in which way the populations of zander and round goby in the Kiel Canal will develop in the future. A re-assessment of growth and diet of zander should be performed about 10 years after the present study. Then it would be possible to detect whether zander growth was inevitably linked to goby peak abundance or could remain on a high level. Biomasses of zander and round goby are determined within the long-term research program that takes place in the area from June to October each year (for details see Chapter 7).

Diet analyses of N. melanostomus were performed by numerous authors (e.g. WANDZEL 2003, CARMAN ET AL. 2006, AZOUR 2011, BRANDNER ET AL. 2013a, Chapter 6). Therein, a high plasticity of prey organisms was found and the prey almost matched the available spectrum of possible prey organisms in the considered ecosystem (see Кидгелко & Кvасн 2005, Borza et al. 2009, Brandner 2014). This high plasticity of prey was confirmed in aquaria experiments inspecting the food intake of round goby when offering different food organisms (THIEL & HEMPEL 2014). In these experiments, the possible prey (zebra mussel Dreissena polymorpha (Pallas, 1771) in acceptable sizes, killer shrimp, ragworm Hediste diversicolor (O.F. Müller, 1776) and gudgeon Gobio gobio (Linnaeus, 1758)) was offered ad libitum. If only one prey taxon was available, the food intake was comparable for each of the offered prey items. In contrast, when offered all four prey taxa at the same time, preferences for certain taxa (killer shrimp and ragworm) could be detected. Similar to these findings, TRÜBNER (2010) found a preference of round goby for European smelt Osmerus eperlanus (Linnaeus, 1758) when offered in aquaria experiments at the same time with zebra mussel and European nightcrawler Eisenia hortensis (Michaelsen, 1890). BAUER ET AL. (2007) conclude also from aquaria experiments that round goby may prefer soft-bodied invertebrate prey over the hard-shelled zebra mussels. Based on these findings it seems necessary to conduct more research into the diet preferences of round goby, especially regarding possible threats for taxa preferred by round goby.

9 References

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Hiermit erkläre ich an Eides statt, dass ich die vorliegende Dissertationsschrift selbst verfasst und keine anderen als die angegebenen Quellen und Hilfsmittel benutzt habe.

Hamburg, 27. September 2017

Mattias Hempel